Safety Assessment of Apple-derived Ingredients as Used in Cosmetics

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All interested persons are provided 60 days from the above release date to comment on this safety assessment and to identify additional published data that should be included or provide unpublished data which can be made public and included. Information may be submitted without identifying the source or the trade name of the cosmetic product containing the ingredient. All unpublished data submitted to CIR will be discussed in open meetings, will be available at the CIR office for review by any interested party and may be cited in a peer-reviewed scientific journal. Please submit data, comments, or requests to the CIR Director, Dr. Lillian Gill.

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Abstract: The Cosmetic Ingredient Review (CIR) Expert Panel (Panel) reviewed the safety of 26 apple-derived ingredients, which function mostly as skin conditioning agents in cosmetic products. Because apple-derived ingredients may be obtained from different apple cultivars, the composition of ingredients derived from different cultivars should be similar to that of ingredients reviewed in this safety assessment. Additionally, industry should use good manufacturing practices to limit impurities that could be present in botanical ingredients. The Panel reviewed relevant data relating to the safety of these ingredients, and concluded that 21ingredients are safe in the present practices of use and concentration in cosmetics as described in this safety assessment, when formulated to be non-irritating and non-sensitizing; the available data are insufficient for determining the safety of pyrus malus (apple) root extract, pyrus malus (apple) stem extract, malus domestica (apple) callus extract, malus domestica (apple) oil, and malus domestica (apple) stem extract.

INTRODUCTION

The safety of the following 18 *Pyrus malus* (apple)-derived ingredients and 8 *Malus domestica* (apple)-derived ingredients as used in cosmetics is reviewed in this safety assessment:

- Pyrus Malus (Apple) Fruit Extract
- Pyrus Malus (Apple) Bark Extract
- Pyrus Malus (Apple) Carpel Powder
- Pyrus Malus (Apple) Fiber
- Pyrus Malus (Apple) Flower Extract
- Pyrus Malus (Apple) Fruit
- Pyrus Malus (Apple) Fruit Water
- Pyrus Malus (Apple) Juice
- Pyrus Malus (Apple) Leaf Extract
- Pyrus Malus (Apple) Pectin Extract
- Pyrus Malus (Apple) Peel Extract
- Pyrus Malus (Apple) Peel Powder
- Pyrus Malus (Apple) Peel Wax
- Pyrus Malus (Apple) Pulp Extract
- Pyrus Malus (Apple) Root Bark Powder
- Pyrus Malus (Apple) Root Extract
- Pyrus Malus (Apple) Seed Extract
- Pyrus Malus (Apple) Stem Extract
- Malus Domestica (Apple) Fruit Extract
- Malus Domestica (Apple) Fruit Water
- Malus Domestica (Apple) Fiber
- Malus Domestica (Apple) Juice
- Malus Domestica (Apple) Oil
- Malus Domestica (Apple) Stem Extract
- Malus Domestica (Apple) Fruit Cell Culture Extract
- Malus Domestica (Apple) Callus Extract

The definitions of these ingredients are included in the *International Cosmetic Ingredient Dictionary and Handbook (Dictionary)*, except for the following five:[1] malus domestica (apple) seed oil, malus domestica (apple) fiber, malus domestica (apple) juice, malus domestica (apple) oil, and malus domestica (apple) stem extract. However, uses of these 5 ingredients in cosmetic products are reported in the Food and Drug Administration's (FDA's) Voluntary Cosmetic Registration Program (VCRP) database.[2]

The following functions of *Pyrus malus* (apple)-derived ingredients and *Malus domestica*-derived ingredients in cosmetic products are reported in the *Dictionary*:[1] skin conditioning agents, binders, emulsion stabilizers, viscosity increasing agents, astringents, fragrance ingredients, antioxidants, exfoliants, and skin bleaching agents. Pyrus malus (apple) root bark powder is the only ingredient in this group that is reported to function as a skin bleaching agent (skin bleaching is not regarded as a cosmetic use in the U.S.,[3] and the Panel will not evaluate safety for that use).

As stated above, *Pyrus malus* and *Malus domestica* are genus and species names for apple that appear within the names of apple-derived ingredients that are listed in the *Dictionary*.[1] Though most of the ingredients reviewed in this safety assessment are identified as *Pyrus malus* (apple)-derived ingredients, CIR has been informed by the cosmetics industry that *Pyrus malus* is a genus and species name for apple that is not in current use, and that the *Pyrus malus*-derived ingredients listed above are under consideration by the cosmetics industry for new name assignments.[4] Regarding additional names for apple, *Malus domestica* and *Malus sylvestris* are listed as other names for *Pyrus malus* in the Germplasm Resources Information Network online database.[5] The name *Malus sylvestris* is identified as the botanical name for pyrus malus (apple) fruit extract in data on trade name mixtures provided by industry and included in this safety assessment.[6,7] Furthermore, the International Cosmetic Ingredient Nomenclature Committee (INCI), sponsored by the Personal Care Products Council (Council), has determined that the accepted scientific name for apple is *Malus pumila*.

Some of the ingredients (e.g., fruit/fruit-derived) reviewed in this safety assessment may be consumed as food, and daily exposure from food use would result in much larger systemic exposures than those from use in cosmetic products. The

primary focus of the safety assessment of these ingredients as used in cosmetics is on the potential for effects from topical exposure.

The Panel has evaluated the safety of pyrus malus (apple) seed oil and other plant-derived fatty acid oils in cosmetics, and issued a final report with the conclusion that these oils are safe in the present practices of use and concentration.[8] Malus domestica (apple) seed oil was initially included in this safety assessment on apple-derived ingredients, but was removed after it was determined that pyrus malus (apple) seed oil and malus domestica (apple) seed oil are different names for the same ingredient.

In the current safety assessment, if a substance tested in a study is not clearly a cosmetic ingredient, because of the absence of information on the genus and species from which the substance was derived and/or the method of extraction used, the test substance will be referred to by a common name (e.g., apple, apple juice, apple fruit extract, apple seed extract, or pyrus malus).

Data on procyanidin B-2 (epicatechin-(4β→8)-epicatechin, found in apple fruit)[9] are also included for use in this safety assessment. Procyanidins are members of the procyanidin or condensed tannins class of flavonoids.[10] Procyanidin B-2 is not a cosmetic ingredient.

Because apple-derived ingredients may be obtained from different apple cultivars, the Panel noted that the composition of ingredients derived from different cultivars should be similar to the composition of ingredients reviewed in this safety assessment.

Data on skin depigmentation effects are included in this safety assessment; however, a review of ingredients for drug effects (i.e., noncosmetic effects) is not within the Panel's purview.

CHEMISTRY

Definition and Structure

Pyrus Malus

The definitions and functions of apple-derived cosmetic ingredients reviewed in this safety assessment are presented in Table 1.[1] The ingredients in this report are related by source, as each is a derivative of apple. While the identity and concentrations of ingredient components may vary from plant part to plant part, and from extract method to extract method, those component identities and concentrations cannot be known for these industry-specific ingredients until such information is provided as outlined in the industry's botanical framework. Differences in those components do not neccessitate the regrouping of such ingredients, but instead warrant a comparison/contrast effort as to how those differences affect safety. Those differences are likely to be informative.

Chemical and Physical Properties

Pyrus Malus (Apple) Fruit Extract

Properties of 2 trade name mixtures containing 10-25% pyrus malus (apple) fruit extract are presented in Table 2.[6,7] Composition data on both mixtures are provided in the section on Composition/Impurities – Pyrus Malus (Apple) Fruit Extract.

Method of Manufacture

Pyrus Malus (Apple) Fiber

Information provided by the cosmetics industry indicates that pyrus malus (apple) fiber is derived from residue formed during apple juice production. The residue is dried and mechanically ground and sieved.[11]

Pyrus Malus (Apple) Fruit Extract

Pyrus malus fruit extract has been prepared according to the following procedure:[12] Pyrus malus fruit was peeled manually and the pericarp was separated, dried at room temperature (26-28°C), and powdered. The powder was extracted

with 95% ethanol. The ethanol was evaporated using a rotary evaporator, and the fruit extract was stored at -20°C prior to use.

According to information provided by the cosmetics industry, pyrus malus (apple) fruit extract is made by extracting apples with a 50% vol% 1,3-butylene glycol solution (the extraction solution is 50% butylene glycol and 50% water).[13] The extract is then treated as follows: filtration, sedimentation, filtration, adjustment of the concentration with additional 50% butylene glycol and 50% water solution, and packaging.

The method of manufacture of a trade name material containing 10%-25% pyrus malus (apple) fruit extract (provided by the cosmetics industry) has been described as follows:[6] The plant material is extracted with 1,2-propylene glycol at "considerate" temperatures during a fixed time, and is sterile-filtered at the end of the fabrication. Lactic acid (0.1-1%) is used for pH regulation, and the product contains 0.6% Bactiphen 250G (phenoxyethanol [75%-100%], methylparaben [10-25%], ethylparaben [1%-5%], propylparaben [1%-5%], and butylparaben [1%-5%]) as the preservative. Information provided by the cosmetics industry indicates that another trade name material containing 10%-25% pyrus malus (apple) fruit extract is manufactured using the same procedure, except that glycerin (vegetable origin) is the extraction solvent. Again, lactic acid (0.1%-1%) is used for pH regulation, but the product contains potassium sorbate (0.35%) and sodium benzoate (0.35%) as preservatives.

According to information provided by the cosmetics industry, the process of manufacturing a trade name material containing 20% pyrus malus (apple) fruit extract begins with the mechanical grinding/milling of pyrus malus fruit.[14] This is followed by extraction in butylene glycol (at specific pH and temperature), the addition of phenoxyethanol, and filtration. Another tradename material containing 20% pyrus malus (apple) fruit extract is manufactured differently.[15] Processing (mechanical grinding and milling), is followed by extraction in propylene glycol (at specific pH and temperature) and, then, filtration.

The method of manufacture of a product described as an aqueous solution containing 20% pyrus malus (apple) fruit extract is:[16] (1) solubilization of apple powder in water; (2) separation of soluble and insoluble phases by filtration; and (3) sterilizing filtration.

A product containing pyrus malus (apple) fruit extract (18.5%), water (81.445%), and potassium sorbate (0.055%) is manufactured as follows:[16] (1) solubilization of fruit of *Pyrus malus* in water; (2) enzymatic hydrolysis, separation of soluble and insoluble phases; (3) inactivation by thermic treatment; (4) purification of soluble phase; and (5) concentration of soluble phase and membrane sterilization.

The method of manufacture of a pyrus malus (apple) fruit extract trade name material (identified as apple polyphenol extract) has been described as follows:[17] Unripe apples were crushed and pressed while 10% sodium metabisulfite solution was added. Pectolytic enzyme was used to clarify the juice obtained, and the mixture was centrifuged and/or filtered with diatomaceous earth. The clarified juice was passed through a column with aromatic synthetic adsorbents, after which the column was washed with distilled water to to remove sugars and organic acids. Apple polyphenol extract was eluted with approximately 50% ethanol and concentrated using an evaporator. The concentrated fraction was dried (using a spray drier) to obtain the apple polyphenol extract as a brown powder. Toxicity data on this material (apple polyphenol extract [brown powder]) are summarized in the Toxicology section of this safety assessment.

Malus Domestica (Apple) Fruit Water

Malus domestica (apple) fruit water is prepared by dehydrating the fruit under vacuum at low temperature.[18]

Apple Juice (genus and species of apple not stated)

Apple juice (from Fuji and Gala varieties; genus and species not stated) was obtained by mechanical pressure, depectinized in a water bath (2 h at 45˚C), filtered through paper, bottled, closed, and stabilized by freezing.[19]

Pyrus Malus (Apple) Pectin Extract

A simple procedure for the extraction of pectin has been described as follows:[20] Approximately 40 g of fruit were washed with normal saline (0.90% w/v sodioum chloride in water)], after which the fruit sample was crushed and homogenized in a blender. The resulting materials were allowed to dry in a hot air (60°C) oven for approximately 2 h until the pectin extracted turned into powder. To obtain sterilized pectin, the powdered pectin was sterilized with N-saline.

Apple Peel Extract

Prior to analysis for anthocyanin content, apples (*Malus domestica*) were peeled and the peel was extracted with methanol containing 0.1% hydrochloric acid.[21]

Pyrus Malus (Apple) Root Bark Powder

According to information provided by the cosmetics industry, pyrus malus (apple) root bark powder is extracted with carbon dioxide, and is then purified.[22] Another source indicates that the air-dried, powdered bark of *Pyrus malus* is extracted with petroleum, chloroform, or 90% ethanol.[23] The petroleum extract yielded a faintly yellow crystalline mass, the chloroform extract yielded a dark yellow solid, and the ethanolic fraction provided a very small amount of a brown alkali-soluble residue.

Pyrus Malus (Apple) Seed Extract

Pyrus malus seed extract has been prepared according to the following procedure:[24] After 3 months of stratification (steady exposure to moist environment before germination becomes possible), the seeds (not dehydrated) were extracted. The extraction procedure was performed using chilled 80% redistilled methanol, to which butylated hydroxytoluene (BHT, 10 mg) was added as an antioxidant. Extraction was followed by homogenization in a chilled blender for 10 minutes. The homogenate was allowed to stand overnight (at 4°C) in the dark, filtered, and the residue was reextracted. The filtrate was evaporated (at 35°C) in a rotary evaporator, avoiding direct sunlight, and an aqueous extract remained.

The method of manufacture of pyrus malus (apple) seed extract that is being marketed to the cosmetics industry has been described as follows:[25] Ground apple seeds are extracted with alcohol (95% non-denatured alcohol). The solvent is evaporated to remove any residual alcohol. The residue is a paste that is sold in solution of various solvents.

Pyrus Malus (Apple) Seed Oil

Crude oil from the ground seeds (~ 5 g) has been extracted over a 6 h period using n-hexane.[26] The solvent was removed using a rotary evaporator under reduced pressure, and the oil was flushed with a stream of nitrogen and stored at -20°C in sealed tubes.

Composition/Impurities

The composition of various components of the apple tree will vary based on where the tree is grown, the maturity of the tree, and storage conditions for the parts of the tree that are used to produce cosmetic ingredients. The method of extraction will also affect the composition of ingredients derived from the parts of the apple tree.

Pyrus malus

The following sugars were detected in the nectar of *Pyrus malus* using gas chromatography: fructose, glucose, and sucrose.[27] Quantitative data were not reported.

Pyrus Malus (Apple) Bark Extract

According to information provided by the cosmetics industry, pyrus malus (apple) root bark powder extracted with carbon dioxide is purified to contain 95% to 98% phloridzin, a dihydrochalcone.[22] According to another source, the following 3 compounds were isolated from the bark of *Pyrus malus* extracted with different solvents (petroleum, chloroform, or 90% ethanol): β-sitoserol, friedelin, and epi-friedelinol.[23]

Malus Domestica (Apple) Flower Extract

The extract of flowers from the redgold apple cultivar was analyzed using gas chromatography (GC) and GC-MS (mass spectrometry), and the following compounds identified were classified as making a major contribution to the odor of apple flowers:[28]

cis-hex-3-en-1-ol
benzyl alcohol
2-phenylethyl alcohol
cinnamyl alcohol

nonanal
octanol
indole

Pyrus Malus (Apple) Fruit

Composition data on different varieties of *Pyrus malus* are included in Tables 3 and 4.[29]

A number of sweet and sour varieties of apples from the hilly parts of Northwest India have been found to contain malvidin monoglycoside.[30] Additionally, the skin of Grimes Golden, Jonathon, and Stayman wine sap apples have been reported to contain idaein (3-β-galactosidyl cyanidin), and the related yellow varieties yielded quercetin-3-galactoside.

The following 3 predominant hydroxycinnamic derivatives in the apple (*Pyrus malus* L., var. Calville blanc) have been reported:[31] *p*-coumarylquinic acid, *p*-coumarylglucose, and chlorogenic acid. The concentration of each derivative, some weeks after blossoming, is highest in young fruits. The amount per fruit increases during approximately 1 month and 2 months for *p*-coumarylquinic acid and chlorogenic acid, respectively. During this period, a relative accumulation of the *o*-diphenolic compound is observed, in comparison with *p*-coumaric derivatives.

Assay results for the presence of amino acids in *Pyryus malus* were as follows:[32] alanine, γ-amino butyric acid, asparagine, cysteine or cystine, and glutamic acid. Quantitative data were not reported. The presence of an unidentified substance that reacted with ninhydrin was also reported.

Pyrus Malus (Apple) Fiber

Composition data on pyrus malus (apple) fiber provided by the cosmetics industry are included in Table 5.[33,34]

Malus Domestica (Apple) Fruit
and Malus Domestica (Apple) Peel

For 20 apple cultivars, the concentrations of 5 classes of polyphenols were significantly different, across cultivar, for both the peel and flesh.[35] The total polyphenol concentration ranged from 0.9 µg/g wwb (wet weight basis) in the flesh of Newtown Pippin to 453 µg/g wwb in the peel of Red Delicious. Harrison, Granny Smith, Rome, Winesap, and Black Twig cultivars contained the highest concentration of total flavan-3-ols in flesh.

Apple Fruit (genus and species not stated)

Data on the average content of phenolic compounds in fruit from apple trees (genus and species not stated; semi-dwarf and super-dwarf rootstocks) grown in Lithuania are as follows:[36] Whether or not the fruit was ripe was not stated.

- Chlorogenic acid (729-1047 µg/g dry weight)
- Phloridzin (83.7-122 µg/g dry weight)
- Procyanidin B1 (33.5-81.5 µg/g dry weight)
- Procyanidin B2 (504-920 µg/g dry weight)
- Σ (Σ = total) Procyanidins (558-1001 µg/g dry weight)
- **(+)**-Catechin (35.6-77.3 µg/g dry weight)
- (-)-Epicatechin (217-329 µg/g dry weight)
- Σ Catechins (254-406 µg/g dry weight)
- Hyperoside (87-147 µg/g dry weight
- Isoquercitrin (15.4-23.3 µg/g dry weight)
- Rutin (15.1-21.6 µg/g dry weight)
- Avicularin (59.9-94 µg/g dry weight)
- Quercitrin (86.1-130 µg/g dry weight)
- Σ Quercetin glycosides (264-416 µg/g dry weight)
- Σ Phenolic compounds (1976-2943 µg/g dry weight)

Super-dwarf rootstocks had the highest content of all phenolic compounds tested, and semi-dwarf rootstocks had the lower content of all phenolic compounds tested. The content of (+)-catechin, procyanidin B1, and total procyanidins in apple fruits depended on the rootstock genotype. The rootstock genotype had a lesser effect on the content of quercitrin, (-)-epicatechin,

total catechins, phloridzin, and chlorogenic acid. It was noted that conditions during the growing season, yield, and fruit weight had an impact on the content of phenolic compounds.

Pyrus Malus (Apple) Fruit Extract

Using paper chromatography, the extracts (extractant not stated) of fruits of *Pyrus malus* have been found to contain chlorogenic and isochlorogenic acids.[23] Some glycosides of quercetin were also detected. The total polyphenols (~ 200 mg/100 g dry weight) and total flavonoids (~ 25 mg/100 g dry weight) content of pyrus malus fruit extract have also been reported.[12] According one of the suppliers of pyrus malus (apple) fruit extract for use in cosmetic products, this ingredient contains sugar and organic acids.[13]

According to information provided by the cosmetics industry, *Malus sylvestris* has been identified as the botanical name for pyrus malus (apple) fruit extract in 2 pyrus malus (apple) fruit extract trade name mixtures containing 10%-25% pyrus malus (apple) fruit extract.[6,7] Composition data on *Malus sylvestris* include (quantitative data not reported):

- Quercetin
- Mineral substances
- Essential oil
- Enzymes
- Fruit acids
- Amino acids
- Vitamins
- Arabane
- Galactane
- Tannins
- Pectin
- Sugars
- Wax

One trade name mixture contains pyrus malus (apple) fruit extract (10%-25%) and propylene glycol (75%-100%),[6] and the other contains pyrus malus (apple) fruit extract (10%-25%), glycerin (75%-100%), and aqua (water) (10%-25%).[7]

The composition (polyphenol profiles) of a pyrus malus (apple) fruit extract trade name material (identified as apple polyphenol extract) has been described as follows:[17] procyanidins (63.8%), which comprised 11.1% dimers, 12.3% trimers, 8.7% tetramers, 5.9% pentamers, 4.9% hexaers, and 20.9% other polymers. It also contained 12.4% flavan-3-ols (monomers), 6.5% other flavonoids, 10.8% non-flavonoids. In addition to the polyphenols, apple polyphenol extract also contained 1.8% moisture, 2.1% protein, and 0.4% ash.

Pyrus Malus (Apple) Juice

Juice from the Fuji and Gala apple varieties (genus and species not stated) in Brazil has the following composition:[19] malic acid (0.18 to 0.389 g/100 mL), total reducing sugar (8.65 to 15.18 g/100 mL), and total phenolic compounds (100 to 400 mg/L). In comparison, apple juice from the Golden delicious variety contains: malic acid (0.312 g/100 mL), total reducing sugar (10.533 g/100 mL), and total phenolic compounds (535.082 mg/L).

Pyrus Malus (Apple) Leaf and Pyrus Malus (Apple) Leaf Extract

The partition of nitrogen in various parts of *Pyrus malus* throughout a one-year cycle has been studied.[37] The study involved seedling apple trees that received heavy applications of sodium nitrate at regular intervals throughout the vegetative period. Positive tests for nitrates (or nitrites) were reported for one tissue only, the leaf buds, just as they were opening. Flavone glycosides have also been detected in the leaves of *Pyrus malus*.[23] The sugar alcohol sorbitol has been detected in *Pyrus malus* leaves at a concentration of 0.45%.[38]

Although data on the composition of the cosmetic ingredient, pyrus malus leaf extract were not found, composition data on the volatile oil obtained from fresh leaves of the *Malus domestica* tree are available. This oil is a complex mixture of mono-, sesqui-, and di-terpenes, phenolics, and aliphatic hydrocarbons, and has been classified as cytotoxic to animal and human cancer cell lines.[39] The major compounds of this oil have been characterized as: eucalyptol, (43.7%), phytol (11.5%), α-farnesene (9.6%), and pentacosane (7.6%).

Malus Domestica (Apple) Leaf Extract

A study relating to the composition and content of phenolic compounds in ethanol extracts of apple (*Malus domestica*) leaves was performed.[40] Phloridzin (a phloretin 2'-glucoside) was a predominant component in the ethanol extracts of apple leaves from all of the cultivars that were analyzed by high-performance liquid chromatography (HPLC). Additionally, the following quercetin glycosides were identified in these extracts:

hyperoside
isoquercitrin
avicularin
rutin
quercitrin (major compound among the quercetin glycosides identified)

Using an ultra-performance liquid chromatography-diode array detection (UPLC-DAD) quantification method, the following 4 compounds were identified in apple leaves of two different varieties, golden and royal:[41]

rutin
3-hydroxyphloridzin
phloridzin
quercetin-3-O-arabinoside

Pyrus Malus (Apple) Pectin Extract

In the absence of data on the extract, it should be noted that pectin is a complex mixture of polysaccharides that comprises approximately one third of the cell wall dry substance of higher plants.[20] The highest concentrations of pectin are found in the middle lamella of the cell wall, with concentrations gradually decreasing from the primary wall toward the plasma membrane.

Malus Domestica (Apple) Peel Extract

The total phenolic and anthocyanin content of apple peels from 6 apple (*Malus domestica*) cultivars grown in southern Brazil was studied.[21] Total phenolic content varied from 105.4 to 269.7 mg gallic acid equivalents (GAE)/100 g of fresh mass (FM). The differences in phenolic content among the apple cultivars were statistically significant ($p < 0.05$). Similarly, the differences in anthocyanin composition (in apple peels) among the apple cultivars were statistically significant, and values ranged from 4.79 to 41.96 mg cyanidin-3-galactoside (cy-3-gal)/100 g of FM. Cy-3-gal is the major anthocyanin that is present in red or partially red genotype apples.

In another study, variations in the content of phenolics, antioxidant activity, and minerals in the peel and pulp of 5 apple (*Malus domestica* Borkh.) cultivars from Pakistan were studied.[42] The mean extract yield of antioxidant components obtained with 80:20 methanol-water (v/v) was determined to be 22.1 g/100 g for the peel and 14.2 g/100 g for pulp on a dry weight (DW) basis. Ranges of total phenolics (1,907.5-2,587.9 mg GAE/100 g DW) and total flavonoids (1,214.3-1,816.4 mg catechin equivalent (CE)/100 g DW) have been reported for the peel of different cultivars of apple. Similarly, ranges of total phenolics (1,185.2-1,475.5 mg GAE/100 g DW) and total flavonoids (711.8-999.3 mg CE/100 g DW) have been reported for the pulp of different cultivars of apple. An analysis for minerals content was also performed. In both the peel and pulp, potassium (K)-containing minerals content was highest, followed by minerals containing Mg, Ca, Fe, Na, and Zn.

Pyrus Malus (Apple) Seed Extract

Endogenous levels of both cis- and trans-isomers of free and bound abscisic acid were studied in dormant and after-ripened *Pyrus malus* embryos.[24] In bioassays, the level of free abscisic acid was very high in dormant embryos, mainly in cotyledons (4800 ng), but became very low after 3 months of stratification. A considerable increase in bound abscisic acid (1450 ng) was noted, and bound abscisic acid became dormant in after-ripened embryos. Bound trans-abscisic acid was detected only in the cotyledons (100 pg) of after-ripened embryos.

The extracts prepared from dry, dormant *Pyrus malus* L. seed (120 g) were found to contain gibberellins (GAs, which are growth promoting hormones).[43] These extracts contained GA_4 (3.080 mg total; 26 µg/g dry seed) and GA_7 (0.960 mg total; 8 µg/g dry seed). The ratio of GA_4 to GA_7 was 76:24. The ratios for seeds stratified for 35 days and 90 days were 81:19 and 64:36, respectively. Another study was performed to identify free GAs in dormant embryos of *Pyrus malus* L. cV Golden delicious using different extraction procedures.[44] Using an ethanolic extraction procedure, minute quantities of free gibberellins (traces to 50 pg/embryo) were detected. Extraction with Tris buffer (pH 7.2) yielded slightly higher quantities of gibberellins (traces to 134 pg). Very large amounts of gibberellins, especially GA_1 and GA_4 (560 and 1560 pg/embryo, respectively), were detected when the embryos were crushed in Tris buffer and treated with Triton X 100.

Residual level of pesticides in pyrus malus (apple) seed extract is controlled to comply with European food limits.[25]

Apple Seed Extract

Royal Gala apple seeds (genus and species not stated) were collected from apple pomace.[45] The methylated hexane extract of the seeds consisted mainly of fatty acids (80.9%) in its volatile fraction and benzaldehyde (< 0.2%) was also detected. Of the fatty acids identified, linoleic acid content was highest (51.2%), followed by palmitic acid (10.5%), linolenic acid (5.6%), stearic acid (4.3%), and oleic acid (4.1%). Fatty acid composition data are included in Table 6.

Further extraction of the seed with 70% aqueous acetone yielded the following 2 major compounds: [(6-*O*-β-D-glucopyranosyl)oxy]benzeneacetonitrile (amygdalin) and phloretin-2'-β-D-glucopyraide (phloridzin). The minor polyphenols identified were: chlorogenic acid, *p*-coumarylquinic acid, 3-hydroxyphloridzin, phloretin-2'-xyloglucoside, and quercetin glycosides.[45]

USE

Cosmetic

The safety of Pyrus malus (apple)-derived ingredients is evaluated based on the expected use of these ingredients in cosmetics. The Panel uses data received from FDA and the cosmetics industry to determine cosmetic use. Use frequencies of individual ingredients in cosmetics are collected from manufacturers and reported by cosmetic product category in FDA's VCRP database. Use concentration data are submitted by Industry in response to surveys of maximum reported use concentrations, by product category, that are conducted by the Council. Collectively, the use frequency and use concentration data indicate that 16 of the 26 apple-derived ingredients are used in cosmetic products.

According to information supplied to the FDA's VCRP by industry, and the results from a survey of ingredient use concentrations conducted by the Council, 9*Pyrus malus* (apple)-derived ingredients and 7 *Malus domestica* (apple)-derived ingredients are being used in cosmetic products, and malus domestica (apple) fruit extract has the highest reported use frequency (382 products)[2,46] The following 10 ingredients are not reported as being used in cosmetic products:

Pyrus malus (apple) bark extract
Pyrus malus (apple) carpel powder
Pyrus malus (apple) fiber
Pyrus malus (apple) leaf extract
Pyrus malus (apple) peel extract
Pyrus malus (apple) peel wax
Pyrus malus (apple) pulp extract
Pyrus malus (apple) root bark powder
Pyrus malus (apple) stem extract
Malus domestica (apple) callus extract

The Council survey data also indicate that apple-derived ingredients are being used in leave-on cosmetic products at maximum ingredient use concentrations up to 9% (i.e., for pyrus malus (apple) fruit water in face and neck products [not sprays]), and in rinse-off cosmetic products at maximum ingredient use concentrations up to 0.75% (i.e., for pyrus malus (apple) fruit extract in hair conditioners).[46] Frequency of use/use concentration data for *Pyrus malus* (apple)- and *Malus domestica* (apple)-derived ingredients are summarized in Table 7.

Cosmetic products containing *Pyrus malus* (apple)-derived or *Malus domestica* (apple)-derived ingredients may be applied to the skin and hair or, incidentally, may come in contact with the eyes and mucous membranes. Products containing these ingredients may be applied as frequently as several times per day and may come in contact with the skin or hair for variable periods following application. Daily or occasional use may extend over many years.

The following ingredients are being used in products that may be inhaled: pyrus malus (apple) fruit extract, pyrus malus (apple) flower extract, pyrus malus (apple) fruit, malus domestica (apple) fruit extract, and malus domestica (apple) fruit water. The highest maximum use concentration that is being reported for *Pyrus malus* (apple)- or *Malus domestica* (apple)-derived ingredients in these types of products is 0.1% for pyrus malus (apple) fruit extract in perfumes and in body and hand sprays. The product types and maximum use concentrations reported for all of the apple-derived ingredients that are used in products that may be inhaled are presented in Table 7. In practice, 95% to 99% of the droplets/particles released from cosmetic sprays have aerodynamic equivalent diameters >10 μm, with propellant sprays yielding a greater fraction of droplets/particles below 10 μm, compared with pump sprays.[47,48,49,50] Therefore, most droplets/particles incidentally inhaled from cosmetic sprays would be deposited in the nasopharyngeal and bronchial regions and would not be respirable (i.e., they would not enter the lungs) to any appreciable amount.[47,48]

Noncosmetic

Apples are among the 20 most frequently consumed raw fruits in the United States.[51]

TOXICOKINETICS

Apple Juice

Unfiltered apple juice (500 ml) was consumed by human subjects, and blood and urine samples were analyzed for total phenolic content and the concentration of selected individual polyphenolic compounds.[52] Large differences in apple polyphenol pharmacokinetics between subjects were observed. Pharmacokinetic data were classified into subgroups according to fast or slow rates of polyphenol metabolism. For some of the subjects, metabolism was undetectable within the time frame of the study. No differences in renal excretion were detected when male and female subjects were compared. However, relative concentrations of polyphenolic compounds were slightly higher in male subjects. Thus, apple-derived polyphenols can be readily detected in human blood and urine after apple juice consumption. The authors noted that the existence of subpopulations with different pharmacokinetics was suggestive of significant variations in individual metabolism rates of polyphenolic substances.

TOXICOLOGY

As noted earlier, some of the ingredients (e.g., fruit/fruit-derived) reviewed in this safety assessment may be consumed as food, and daily exposure from food use would result in much larger systemic exposures than those from use in cosmetic products. The primary focus of the safety assessment of these ingredients as used in cosmetics is on the potential for local effects from topical exposure.

Toxicity data on procyanidin B-2 (epicatechin-(4β→8)-epicatechin, found in apple fruit) were identified in the published literature, and are included for use in the safety assessment of apple fruit extract. As mentioned previously, procyanidins are members of the procyanidin or condensed tannins class of flavonoids.[10] Procyanidin B-2 is not a cosmetic ingredient.

Acute Toxicity

Oral

Apple Fruit Extract

Apple polyphenol extract (dissolved in 0.5% sodium carboxymethyl cellulose solution) was administered intragastrically to Sprague-Dawley (Crj:CD) rats (5 males, 5 females) at an oral dose of 2,000 mg/kg body weight (dose volume = 10 ml/kg).[17] The production of apple polyphenol extract is described in the Method of Manufacture section of this safety assessment. The animals were necropsied after a 14-day observation period. None of the animals died, and the authors noted that there were no significant changes in organs examined. It was concluded that the acute oral minimum fatal dose of apple polyphenol extract was > 2,000 mg/kg body weight.

Pyrus Malus (Apple) Seed Extract

The acute oral toxicity of pyrus malus (apple) seed extract (1% w/v in hexyldecanol) was studied using 10 male mice (IFFA CREDO OF 1).[25] The test substance was administered at a maximal oral dose of 20 ml/kg, and dosing was followed by an 8-day observation period. Gross necropsy was performed on day 8. There was no evidence of test substance-related clinical signs in the study, and the behavior of animals was considered normal. Gross necropsy did not reveal any evidence of modification of the main organs. Particularly, there were no signs of necrosis or ulceration of the digestive tract.

Subcutaneous

Procyanidin B-2

Procyanidin B-2 (> 94% pure) was administered to groups of ten 6-week-old Sprague-Dawley rats of the Crj: CD, SPF/VAF strain (5 males, 5 females/group).[10,9] The test material (in purified water) was administered subcutaneously to 3 groups at doses of 500 mg/kg, 1,000 mg/kg, and 2,000 mg/kg, respectively (dose volume = 10 ml/kg). The single dose was injected into the neck. A fourth group (control) was dosed with purified water. None of the animals died. Hair loss and crust formation (in all 3 dose groups), and swelling (2,000 mg/kg group) were reported. The following observations were made at necropsy on day 14: thickening of the subcutis with granulomatous inflammation in the 1,000 mg/kg group (4 of 5 males; 2 of 5 females) and 2,000 mg/kg group (all animals); accumulation of pigment-laden macrophages in the duodenal

mucosa in the 500 mg/kg group (1 of 5 females), 1,000 mg/kg group (4 of 5 males; all females), and 2,000 mg/kg group (all animals).

Repeated Dose Toxicity

Oral

Apple Fruit Extract

Apple polyphenol extract was administered intragastrically to 3 groups of Sprague-Dawley (Crj:CD) rats (10 males, 10 females) at doses of 500; 1,000; and 2,000 mg/kg/day (dose volume = 10 ml/kg).[17] The doses were administered daily for 90 days. A fourth group served as the untreated control. Necropsies were performed on all animals at the end of the study. Body weight gain was unaffected by treatment, and changes in food consumption were attributed to normal biological variation. Statistically significant differences ($P < 0.05$ or $P < 0.01$) in hematological, clinical chemistry, and urinary tests were reported. However, these changes were within the normal range of physiological background data and were not correlated with the apple polyphenol extract dosage. Gross necropsy findings were not indicative of adverse changes in any of the organs examined. A statistically significant increase in lung weight was found in the 1,000 mg/kg/day dose group, but this change was within the normal physiological range. Histopathological examination of organs did not reveal any changes that were related to dosing with the test material.

Antimicrobial Activity

Pyrus Malus (Apple) Pectin Extract

Pectin was extracted from several varieties of apple, i.e., American, Delicious, and Maharaj-ji (all obtained from Kashmir).[20] Pectin content was found to be maximum in Maharaj-ji (20.60%), followed by Delicious (14.4%) and American (11.60%). The pectin extracted was evaluated for *in vitro* antibacterial activity against different pathogenic bacterial cultures and antifungal activity. Pectin extracted from the Delicious variety had potent antibacterial activity against *Klebsiella pneumonia* (mean inhibitory concentration [MIC] = 0.8 mg/ml), followed by *Streptococcus pyogenes* (MIC = 0.3 mg/ml), *E. coli* (MIC = 0.7 mg/ml), and *Lactococcus* sp. (MIC = 0.7 mg/ml). There was no evidence of antibacterial activity in any of the bacterial strains tested with pectin extracted from the other varieties. Regardless of the variety from which pectin was extracted, there was no evidence of antifungal activity against the following fungal strains: *Aspergillus niger*, *Candida albicans*, or *Saccharomyces cerevisiae*.

REPRODUCTIVE AND DEVELOPMENTAL TOXICITY

Data on the reproductive and developmental toxicity of apple-derived ingredients were not found in the published literature nor were unpublished data provided.

GENOTOXICITY

In Vitro

Apple Fruit Extract

The genotoxicity of apple fruit extract was evaluated in the Ames test using *Escherichia coli* strain WP2*uvr*A and *Salmonella typhimurium* strains TA100, TA98, TA1535, and TA1537.[10] The test material was evaluated at doses up to 5,000 µg/plate with and without metabolic activation. Slight genotoxicity was observed at a dose of 2,500 µg/plate without metabolic activation, but not at the other doses, with or without metabolic activation.

Apple fruit extract (concentrations up to 0.313 mg/ml) was evaluated for genotoxic activity in Chinese hamster CHL/IU mammalian cells using a chromosomal aberrations assay with and without metabolic activation.[10] Genotoxicity was not observed, with or without metabolic activation.

Pyrus Malus (Apple) Seed Extract

The genotoxicity of pyrus malus (apple) seed extract (1% w/v in hexyldecanol) was studied using the following *S. typhimurium* strains: TA98, TA100, TA1535, TA1537, and TA1538.[25] The test substance was evaluated at doses up to 5,000 µg/plate with and without metabolic activation. Revertant frequencies in treated strains tested were similar to vehicle control values. All positive control data were within acceptable ranges. The test substance was classified as non-genotoxic.

Procyanidin B-2

The genotoxicity of procyanidin B-2 (> 94% pure) was evaluated using *S. typhimurium* strains TA98, TA100, TA1535, and TA1537, and *E. coli* strain WP2*uvrA*.[9] Procyanidin B-2 was tested at doses up to 5,000, µg/plate with or without metabolic activation. Test results were negative.

In the chromosome aberrations assay using Chinese hamster lung cells, procyanidin B-2 (> 94% pure) was evaluated with (concentrations up to 4.8 mM) and without (concentrations up to 1.8 mM) metabolic activation.[9] Neither structural aberrations such as chromosome-type aberrations or chromatid-type aberrations were observed with or without metabolic activation. However, polyploidy was observed with metabolic activation.

In Vivo

Apple Fruit Extract

The genotoxicity of apple fruit extract was evaluated in the micronucelus test.[17] Three groups of 5 male Sprague-Dawley rats (7 weeks old) were dosed orally with 500 mg/kg body weight; 1,000 mg/kg body weight; and 2,000 mg/kg body weight (dose volume = 10 ml/kg), respectively. Genotoxicity was evaluated by measuring the frequency of polychromatic erythrocytes cells in bone marrow. Test results were negative.

Procyanidin B-2

In a micronucleus test, groups of 8-week-old mice [Crj: CD-1 (ICR), specific pathogen free (SPF)/virus antibody free (VAF)] received single subcutaneous injections of 500 mg/kg, 1,000 mg/kg, and 2,000 mg/kg procyanidin B-2, respectively.[9] The frequency of micronucleated polychromatic erythrocytes in the bone marrow of mice dosed with procyanidin B-2 was not significantly different from that of the negative control. Whether or not a statistical analysis of the data was performed was not stated.

CARCINOGENICITY

Data on the carcinogenicity of apple-derived ingredients were not found in the published literature nor were unpublished data provided.

ANTICARCINOGENICITY

The anticarcinogenicity of apples and their components has been the subject of a review article.[53] In *in vitro* studies, apple extracts and components, especially oligomeric procyanidins, have been shown to influence multiple mechanisms that are relevant for cancer prevention. Additionally, apple products have been shown to prevent skin, mammary, and colon carcinogenesis in animal models, and epidemiological observations indicate that regular consumption of one or more apples a day may reduce the risk for lung and colon cancer.

IRRITATION AND SENSITZATION

Skin Irritation and Sensitization

In Vitro

Pyrus Malus (Apple) Fruit Extract

The skin irritation potential of a tradename material containing 20% pyrus malus (apple) fruit extract was evaluated using the Epiderm™ model, a reconstructed human epidermal model consisting of human-derived epidermal keratinocytes.[54] This is a European Center for the Validation of Alternative Methods (ECVAM)-validated test method.[55] The keratinocytes have been cultured to form a multilayer, highly differentiated model of the human epidermis. In the Epiderm™ model assay, a chemical is classified as an irritant if the mean relative tissue viability of exposed tissues is reduced by 50% of the mean viability of the negative controls. A non-irritant's viability is > 50%. Sterile Dulbecco's phosphate-buffered saline (DPBS) and sterile deionized water served as negative controls. Sodium dodecyl sulfate solution (5%) served as the positive control. The test material was classified as a non-irritant in this assay. The positive control was classified as an irritant.[54] In the same assay, negative results were also reported for another tradename material containing 20% pyrus malus (apple) fruit extract.[56]

Non-Human

Pyrus Malus (Apple) Fruit Extract

A product described as an aqueous solution containing 20% pyrus malus (apple) fruit extract was applied for 4 h (under a dressing [type not stated]) to the skin of 3 albino rabbits according to the Organization for Economic Co-operation and Development (OECD) 404 protocol.[16] Test sites were examined for up to 72 h after removal of the dressing. The test substance was classified as having slight skin irritation potential.

The skin sensitization potential of a product described as an aqueous solution containing 20% pyrus malus (apple) fruit extract was studied in the maximization test (OECD 406 protocol) using albino guinea pigs (number not stated).[16] Positive reactions were not observed. However, it was concluded that the test substance had slight skin sensitization potential, but did not induce skin irritation.

Pyrus Malus (Apple) Seed Extract

The skin irritation potential of pyrus malus (apple) seed extract (1% w/v in hexyldecanol) was evaluated using 6 New Zealand white rabbits.[25] Prior to application, the test substance was diluted to a concentration of 23% in sterile distilled water (effective test substance concentration = 1% x 23% = 0.23%). An occlusive patch containing the diluted test substance (0.5 ml) was applied for 24 h to abraded and intact skin on the right and left side, respectively. The dose per cm^2 was not stated. Reactions were scored according to the Draize scale at 24 h, 48 h, and 72 h post-application. Mild erythema was observed at intact and abraded test sites of all animals, but was completely reversible between 48 h and 72 h post-application. Edema was not observed. The test substance was classified as a slight skin irritant (index of cutaneous irritation = 0.58).

The guinea pig maximization test was used to evaluate the skin sensitization potential of pyrus malus (apple) seed extract (1% w/v in hexyldecanol) in 20 male guinea pigs (strain not stated).[25] During induction, the animals were injected intradermally with the test substance, followed by topical dermal application of the test substance (25% in Vaseline) under an occlusive patch for 48 h. The effective concentration of the test substance applied dermally during induction was 0.25% (1% x 25% = 0.25%). The dose per cm^2 was not stated. During the challenge phase, the test substance (0.25%) was applied to the flank, under an occlusive, patch for 24 h. An additional 20 guinea pigs served as controls (treated similarly, but without test substance application). Reactions were scored at 24 h and 48 h post-application. Mild erythema was observed in treated and control animals, and reactions had cleared by 48 h. The erythema observed was interpreted as a consequence of an irritation phenomenon, rather than as a sensitization phenomenon. This was so because the erythema observed at 24 h was reported for an equal number of treated and control animals, and because the percentage of the reaction observed was extremely low. The test substance was classified as a non-sensitizer.

Procyanidin B-2

Procyanidin B-2 (vehicle not stated) was applied topically to dorsal abraded and intact skin of 12 New Zealand white rabbits (Kbs: NZW, SPF) using an occlusive patch (0.5 ml on 2.5 x 2.5 cm patch).[9] Sodium lauryl sulfate (8% aqueous) and physiological saline served as positive and negative controls, respectively. The patches remained in place for 24 h, and reactions were scored at 24.5 h and 72 h post-application. Skin irritation was not observed at abraded or intact sites treated with procyanidin B-2, the vehicle, or saline. Significant inflammation (severe irritation) was observed at sites treated with 8% sodium lauryl sulfate.

The skin sensitization potential of procyanidin B-2 was evaluated in a maximization test using 24 male guinea pigs (Crj: Hartley,SPF/VAF).[9] A 2 x 4 cm area of dorsal skin was used in the induction phase of the study, and the test procedure involved a combination of subcutaneous injections and topical applications. During the induction phase, procyanidin B-2 (1%) was injected subcutaneously, and, after 24 h, an occlusive patch containing 0.2 ml of 10% procyanidin B-2 was applied for 24 h. The challenge phase was initiated after a 2-week non-treatment period. A 2 x 2 cm occlusive patch containing 10%

procyanidin B-2 (0.1 ml) was applied for 24 h. Reactions were scored at 24.5 h, 48 h, and 72 h post-application. 2,4-Dinitrochlorobenzene (1% DNCB) served as the positive control. Procyanidin B-2 did not induce sensitization. DNCB was a strong sensitizer.

Human

Pyrus Malus (Apple) Fruit Water

The skin irritation and sensitization potential of a face and neck product containing 8.7835% pyrus malus (apple) fruit water was evaluated in a human repeated insult patch test involving 49 male and female subjects.[57] During induction, an occlusive patch (2 cm^2) containing the test substance (0.2 ml) was applied for 24 h to an area between the scapula and waist that was adjacent to the midline. A total of 9 induction applications were made. A 2-week non-treatment period was followed by the challenge phase. A challenge patch was applied for 24 h to a new test site. Reactions were scored at 24 h and 72 h post-application. Transient, barely perceptible (+) to mild (1-level) reactions (all non-specific patch test responses) were observed in 5 or 49 subjects during the induction and/or challenge phase of the study. It was concluded that the test substance did not induce skin irritation or cause allergic contact dermatitis.

Phototoxicity

Pyrus Malus (Apple) Seed Extract

The phototoxicity of pyrus malus (apple) seed extract (1% w/v in hexyldecanol) was studied using albino guinea pigs.[25] The animals (10 per group) received an application of the test substance (1 ml) with or without UVA irradiation. The dose per cm^2 was not stated. Test sites were examined at 1 h, 6 h, and 24 h post-irradiation. There was no evidence of dermal reactions in irradiated or non-irradiated animals, and the test substance was classified as non-phototoxic.

Ocular Irritation

In Vitro

Pyrus Malus (Apple) Fruit Extract

The ocular irritation potential of a tradename material containing 20% pyrus malus (apple) fruit extract was evaluated using the Epiocular™ model, a multilayer, highly differentiated model of the human corneal epithelium.[54] In the Epiocular™ model assay, a chemical is classified as an irritant if the mean relative tissue viability of the exposed tissues is reduced by 60% of the mean viability of the negative controls. A non-irritant's viability is > 40%. Sterile DPBS and sterile deionized water served as negative controls. Methyl acetate served as the positive control. The test material was classified as a non-irritant in this assay. The positive control was classified as an irritant. In the same assay, negative results were also reported for another tradename material containing 20% pyrus malus (apple) fruit extract.[56]

Using the method of neutral red release, the ocular irritation potential of a product described as an aqueous solution containing a maximum percentage of 20% pyrus malus (apple) fruit extract was determined using a monolayer of rabbit corneal fibroblasts.[16] The product was tested at concentrations up to 50% (effective test substance concentration = 20% x 50% = 10%) In this assay, cytotoxicity was evaluated by determining the test concentration that caused 50% cell death (IC_{50}). Test results indicated that the IC_{50} was above a concentration of 50%. The % cell death that was observed at a dilution of 50% was 15%, and, thus, cytotoxicity was said to have been negligible.

In Vivo

Pyrus Malus (Apple) Fruit Extract

A product described as an aqueous solution containing a maximum percentage of 20% pyrus malus (apple) fruit extract (0.1 ml) was instilled into one eye of each of 3 rabbits according to the OECD 405 protocol.[16] Instillation was not followed by ocular rinsing. Untreated eyes served as controls. The test substance was slightly irritating to the eye and did not cause lesions of the ocular mucosa that were considered significant.

Pyrus Malus (Apple) Seed Extract

Pyrus malus (apple) seed extract (0.1 ml; 1% in hexyldecanol) was instilled into the right eye of each of 6 New Zealand white rabbits, and remained for 24 h.[25] Ocular irritation was scored according to the method of Kay and Calandra at 24 h, 48 h, 72 h, 96 h, and 12 h post-instillation. At 24 h post-instillation, conjunctival redness, swelling, and discharge were observed in 6 rabbits. These reactions were completely reversible at 48 h and 96 h post-instillation. Lesions of the cornea or iris were not observed. The test substance was classified as slightly irritating to the eyes (accurate ocular irritation index at 24 h = 0.58).

Procyanidin B-2

The ocular irritation potential of procyanidin B-2 (> 94% pure) was evaluated using twelve male New Zealand white rabbits (Kbs: NZW, SPF).[9] The test material (100 µl) was instilled in the right eye of each animal. The left eye served as the untreated control. Half of the animals were subjected to ocular rinsing after instillation. Ocular reactions were scored for up to 120 h post-instillation. No changes were observed in the cornea or iris, but slight irritation of the conjunctiva was observed. Procyanidin B-2 was classified as minimally irritating to the eyes of rabbits.

MISCELLANEOUS STUDIES

Phloridzin

A study on phloridzin-induced melanogenesis was performed using B16 melanoma cells.[58] Phloridzin (found in many parts of the apple tree) induced a dose-dependent increase (100 µg/ml and 500 µg/ml doses) in tyrosinase activity and melanin content, and these changes were accompanied by an increase in the levels of tyrosinase and the tyrosine-related proteins, TRP-1 and TRP-2. Also, the cAMP-dependent protein kinase A (PKA) inhibitor H89 impaired the response of the tyrosinase activity and melanin synthesis to phloridzin. Collectively, the results of this study indicated that phloridzin increased tyrosinase gene expression through the cAMP signaling pathway, leading to stimulation of melanogenesis.

SUMMARY

Pyrus malus (apple)-derived and malus domestica (apple)-derived ingredients (26 total) are reviewed in this safety assessment. The following functions of these ingredients in cosmetic products are being reported: skin conditioning agents, binders, emulsion stabilizers, viscosity increasing agents, astringents, fragrance ingredients, antioxidants, exfoliants, and skin bleaching agents (skin bleaching is not regarded as a cosmetic use in the U.S.) Pyrus malus (apple) root bark powder is the only ingredient in this group that is reported to function as a skin bleaching agent in cosmetic products.

According to information supplied to FDA by industry as part of the VCRP, and the results from a survey of ingredient use concentrations conducted by the Council, the following 9 *Pyrus malus* (apple)-derived ingredients and 7 *Malus domestica* (apple)-derived ingredients are being used in cosmetic products, and malus domestica (apple) fruit extract has the highest reported use frequency (382) products):

Pyrus malus (apple) fruit extract
Pyrus malus (apple) flower extract
Pyrus malus (apple) fruit
Pyrus malus (apple) fruit water
Pyrus malus (apple) juice
Pyrus malus (apple) pectin extract
Pyrus malus (apple) peel powder
Pyrus malus (apple) root extract
Pyrus malus (apple) seed extract
Malus domestica (apple) fruit extract
Malus domestica (apple) fruit water
Malus domestica (apple) fiber
Malus domestica (apple) juice
Malus domestica (apple) oil
Malus domestica (apple) stem extract
Malus domestica (apple) fruit cell culture extract

Pyrus malus (apple)-derived ingredients are being used in leave-on cosmetic products at maximum ingredient use concentrations up to 9% (i.e., for pyrus malus (apple) fruit water in face and neck products [not sprays]), and in rinse-off cosmetic products at maximum ingredient use concentrations up to 0.75% (i.e., for pyrus malus (apple) fruit extract in hair conditioners).

The available information on the composition of apples indicates that phenolic compounds are present in the fruit, juice, leaves, peel, and seeds. Phloridzin (found in many parts of the apple tree) induced a dose-dependent increase in tyrosinase activity and melanin content, and these changes were accompanied by an increase in the levels of tyrosinase and the tyrosine-related proteins, TRP-1 and TRP-2.

Apple-derived polyphenols were readily detected in human blood and urine after apple juice consumption.

In an acute oral toxicity study of pyrus malus (apple) seed extract (1% w/v in hexyldecanol), there was no evidence of test substance-related clinical signs. At gross necropsy, there were no signs of necrosis or ulceration of the digestive tract, or modification of the main organs. In acute oral and repeated dose toxicity tests (rats) on apple fruit extract, there were no significant hematological, clinical, chemical, histopathological, or urinary effects at a dose of 2000 mg/kg.

A single subcutaneous injection of procyanidin B-2 into rats did not produce signs of significant injury, and a lethal dose of > 2,000 mg/kg was reported.

In the Ames test without metabolic activation, apple fruit extract was slightly genotoxic when tested at a high concentration of 2500 µg/plate, but significant genotoxic activity was not found in the chromosomal aberration test or the micronucleus test. In another Ames test, pyrus malus (apple) seed extract (1% w/v in hexyldecanol) was non-genotoxic in the following *Salmonella typhimurium* strains, with and without metabolic activation: TA98, TA100, TA1535, TA1537, and TA1538. Results were negative for procyanidin B-2 in the Ames test and in a chromosome aberrations assay involving Chinese hamster lung cells.

An aqueous solution containing 20% pyrus malus (apple) fruit extract was slightly irritating to the skin of rabbits. In the guinea pig maximization test, this solution did not induce skin irritation, but had slight skin sensitization potential. The skin irritation potential of a tradename material containing 20% pyrus malus (apple) fruit extract was evaluated using the Epiderm™ model, and results were negative.

Pyrus malus (apple) seed extract (1% w/v in hexyldecanol) caused slight skin irritation when diluted with water to a concentration of 0.23% and applied to the skin of rabbits. In the guinea pig maximization test, pyrus malus (apple) seed extract (1% w/v in hexyldecanol) caused skin irritation, but not sensitization, when tested at an ingredient concentration of 0.25% during induction and challenge. A preparation containing procyanidin B-2 did not cause skin irritation in rabbits. Additionally, procyanidin B-2 did not induce skin sensitization in guinea pigs in the maximization test.

A face and neck product containing 8.7835% pyrus malus (apple) fruit water did not cause skin irritation or allergic contact dermatitis in an RIPT involving 49 subjects.

Pyrus malus (apple) seed extract (1% w/v in hexyldecanol) was classified as non-phototoxic in a study involving albino guinea pigs.

An aqueous solution containing a maximum percentage of 20% pyrus malus (apple) fruit extract (0.1 ml) was slightly irritating to the eyes of rabbits. The same was true after instillation of pyrus malus (apple) seed extract (1% in hexyldecanol) into the eyes of rabbits. In primary ocular irritation tests involving rabbits, both a preparation containing procyanidin B-2 and the vehicle induced slight conjunctival irritation; however, ethanol was presumed to have been the causative agent.

The ocular irritation potential of an aqueous solution containing a maximum percentage of 20% pyrus malus (apple) fruit extract, tested at ingredient concentrations up to 10%, was evaluated using the method of neutral red release. Cytotoxicity was evaluated in this assay and found to be negligible; thus, ocular irritation was negligible. A tradename material containing 20% pyrus malus (apple) fruit extract was evaluated for ocular irritation potential using the Epiocular™ model, and results were negative.

DISCUSSION

The Panel discussed different apple cultivars as sources of apple-derived ingredients, the stimulatory effect of phloridzin (component of apple fruit) on melanogenesis, the presence of impurities in botanical ingredients, and the potential for inhalation exposure to apple-derived cosmetic ingredients. The data needed for completion of the safety assessment on 5 of the apple-derived ingredients were also determined.

Considering that apple-derived ingredients may be obtained from different apple cultivars, the Panel noted that the composition of ingredients derived from different cultivars should be similar to the composition of ingredients reviewed in this safety assessment.

The Panel stated their awareness of data indicating that phloridzin, a component of apple fruit, apple bark extract, apple root bark powder, apple leaf extract, and apple seed extract, had a stimulatory effect on melanogenesis *in vitro*, but

agreed that a review of ingredients for drug effects, i.e., noncosmetic effects, is not within the Panel's purview. However, the Panel noted that an effect on cutaneous pigmentation would not be expected at the use concentrations of apple-derived ingredients in cosmetic products. As a precaution, the Panel noted that manufacturers should be aware of this effect and, thus, the ingredients containing phloridzin in the formulation should not have a stimulatory effect on melanogenesis when applied to the skin.

The Panel expressed concern about pesticide residues and heavy metals that may be present in botanical ingredients. They stressed that the cosmetics industry should continue to use current good manufacturing practices (cGMPs) to limit impurities. The Panel noted the USDA designation of ≤ 15 ppb as corresponding to "negative" aflatoxin content. Additionally, the Panel stated that aflatoxins should not be present at levels of toxicological concern in apple-derived ingredients.

Additionally, the Panel discussed the potential for incidental inhalation exposures to apple-derived ingredients in products that are sprayed or in powder form and agreed that, based on likely airborne particle size distributions and concentrations in the breathing zone and ingredient use, incidental inhalation would not lead to local respiratory effects or systemic effects.

The Panel determined that the following data are needed to evaluate the safety of pyrus malus (apple) root extract, pyrus malus (apple) stem extract, malus domestica (apple) callus extract, malus domestica (apple) oil, and malus domestica (apple) stem extract in cosmetic products:

- Method of manufacture and impurities
- 28-day dermal toxicity study; if any adverse effects noted, genotoxicity and reproductive and developmental toxicity data may be needed

CONCLUSION

The CIR Expert Panel concluded that the following 21 apple-derived ingredients are safe in the present practices of use and concentration in cosmetics as described in this safety assessment, when formulated to be non-irritating and non-sensitizing.

Pyrus Malus (apple) bark extract*
Pyrus Malus (apple) carpel powder*
Pyrus Malus (apple) fiber*
Pyrus Malus (apple) flower extract
Pyrus Malus (apple) fruit extract
Pyrus Malus (apple) fruit
Pyrus Malus (apple) fruit water
Pyrus Malus (apple) juice
Pyrus Malus (apple) leaf extract*
Pyrus Malus (apple) pectin extract
Pyrus Malus (apple) peel extract*
Pyrus Malus (apple) peel powder
Pyrus Malus (apple) peel wax*
Pyrus Malus (apple) pulp extract*
Pyrus Malus (apple) root bark powder*
Pyrus Malus (apple) seed extract
Malus Domestica (apple) fiber
Malus Domestica (apple) fruit extract
Malus Domestica (apple) fruit water
Malus Domestica (apple) fruit cell culture extract
Malus Domestica (apple) juice

The Panel concluded that the available data are insufficient for evaluating the safety of the following 5 apple-derived ingredients in cosmetic products:

Pyrus Malus (apple) root extract
Pyrus Malus (apple) stem extract*
Malus Domestica (apple) callus extract*
Malus Domestica (apple) oil
Malus Domestica (apple) stem extract

*Not reported to be in current use. Were ingredients in this group not in current use to be used in the future, the expectation is that they would be used in product categories and at concentrations comparable to others in this group.

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

Ingredient/CAS No.	Definition	Function
Pyrus malus (apple) fruit extract	Pyrus malus (apple) fruit extract is the extract of the fruit of *Pyrus malus.*	Skin-Conditioning Agents - Miscellaneous

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

Ingredient/CAS No.	Definition	Function
Pyrus malus (apple) bark extract	Pyrus malus (apple) bark extract is the extract of the bark of *Pyrus malus.*	Cosmetic Astringents
Pyrus malus (apple) carpel powder	Pyrus malus (apple) carpel powder is the powder obtained from the dried, ground carpels of *Pyrus malus.*	Skin-Conditioning Agents - Miscellaneous
Pyrus malus (apple) fiber	Pyrus malus (apple) fiber is the finely ground fiber obtained from the dried fruit of *Pyrus malus.*	Binders; Emulsion Stabilizers; Viscosity Increasing Agents - Aqueous
Pyrus malus (apple) flower extract	Pyrus malus (apple) flower extract is the extract of the flowers of *Pyrus malus.*	Skin-Conditioning Agents - Miscellaneous
Pyrus malus (apple) fruit	Pyrus malus (apple) fruit is the fruit of the apple, *Pyrus malus.*	Cosmetic Astringents
Pyrus malus (apple) fruit water	Pyrus malus (apple) fruit water is an aqueous solution of the steam distillate obtained from the fruit of *Pyrus malus.*	Fragrance Ingredients
Pyrus malus (apple) juice	Pyrus malus (apple) juice is the liquid expressed from the fresh pulp of the apple, *Pyrus malus.*	Skin-Conditioning Agents - Miscellaneous
Pyrus malus (apple) leaf extract	Pyrus malus (apple) leaf extract is the extract of the leaves of *Pyrus malus.*	Skin-Conditioning Agents - Miscellaneous
Pyrus malus (apple) pectin extract	Pyrus malus (apple) pectin extract is the extract of the pectin of *Pyrus malus.*	Skin-Conditioning Agents - Miscellaneous
Pyrus malus (apple) peel extract	Pyrus malus (apple) peel extract is the extract of the peel of *Pyrus malus.*	Antioxidants
Pyrus malus (apple) peel powder	Pyrus malus (apple) peel powder is the powder obtained from the dried, ground peel of *Pyrus malus.*	Exfoliants
Pyrus malus (apple) peel wax	Pyrus malus (apple) peel wax is a wax obtained from the peel of the apple, *Pyrus malus.*	Not Reported
Pyrus malus (apple) pulp extract	Pyrus malus (apple) pulp extract is the extract of the pulp of *Pyrus malus.*	Skin-Conditioning Agents - Miscellaneous
Pyrus malus (apple) root bark powder	Pyrus malus (apple) root bark powder is the powder obtained from the dried, ground root bark of *Pyrus malus.*	Skin Bleaching Agents*
Pyrus malus (apple) root extract	Pyrus malus (apple) root extract is the extract of the roots of *Pyrus malus.*	Skin-Conditioning Agents - Miscellaneous
Pyrus malus (apple) seed extract	Pyrus malus (apple) seed extract is the extract of the seeds of *Pyrus malus.*	Skin-Conditioning Agents - Miscellaneous
Pyrus malus (apple) stem extract	Pyrus Malus (Apple) Stem Extract is the extract of the stems of *Pyrus malus.*	Skin-Conditioning Agents - Miscellaneous
Malus domestica (apple) callus extract	Malus Domestica Callus Extract is the extract of the callus of *Malus domestica* grown in culture.	Antioxidants; Skin Protectants
Malus domesatica (apple) fruit extract	Malus Domestica Fruit Extract is the extract of the fruit of *Malus domestica*.	Antioxidants
Malus domestica (apple) fruit water	Malus Domestica Fruit Water is an aqueous solution of the steam distillates derived from the fruit of *Malus domestica.*	Fragrance Ingredients; Skin-Conditioning Agents - Emollient
Malus domestica (apple) fruit cell culture extract	Malus Domestica Fruit Cell Culture Extract is the extract of a culture of the fruit cells of *Malus domestica*.	Skin-Conditioning Agents - Miscellaneous

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

Ingredient/CAS No.	Definition	Function
Malus domestica (apple) callus extract	Malus Domestica Callus Extract is the extract of the callus of *Malus domestica* grown in culture.	Antioxidants; Skin Protectants

*Skin bleaching agent is not regarded as a cosmetic use in the U.S.,[3] and the Panel will not evaluate safety for that use.

Table 2. Properties of Pyrus Malus (Apple) Fruit Extract Trade Name Mixtures.[6,7]

A mixture containing 10-25% pyrus malus (apple) fruit extract and 75-100% propylene glycol	A mixture containing 10-25% pyrus malus (apple) fruit extract, 10-25% water, and 75-100% glycerin
Clear, brownish yellow colored liquid	Clear, brownish yellow colored liquid
Faint fruity odor	Faint fruity odor

Table 3. Composition Data on Different Varieties of *Pyrus malus*.[29]

Variety	Fat (%)	Fiber (%)	Protein (%)	Pectin (%)	Reducing Sugar	Non-reducing Sugar	Total Sugar
Amri	0.22	5.89	0.47	0.58	10.34	3.87	14.21
Bluish Golden	5.15	3.61	0.29	1.29	9.71	2.58	11.83
Bonza	0.21	4.41	0.74	1.63	11.18	4.28	15.46
Discovery	0.66	8.6	0.44	1.6	10.59	6.16	16.75
Elster	0.25	7.46	0.36	0.88	9.51	7.88	16.59
Florina	0.24	4.17	0.48	0.62	11.84	5.93	17.77
Gala	0.26	4.35	0.27	0.53	11.95	5.78	17.73
Gloster 69	3.38	4.03	0.17	1.07	10.76	3.75	14.51
Golden Delicious	0.23	5.06	0.44	0.51	11.29	4.41	15.7
Granny Smith	0.28	3.8	0.48	0.99	11.14	4.32	15.46
Green Sleeves	0.24	6.14	0.37	1.05	11.37	7.92	19.29
Ida Red	0.21	1.91	0.36	0.69	9.95	3.57	13.52
Kaja	0.08	1.13	0.38	0.43	7.1	0.57	7.67
Kids Orange	0.21	1.16	0.35	1.07	11.37	7.51	18.88
Mushadi	0.23	5.55	0.45	0.52	8.69	3.75	12.44
Red Delicious	0.07	2.56	0.47	0.58	10.74	4.09	14.83
Samootree	0.02	3.92	0.34	1.29	13.14	6.99	20.13
Summer Red	0.13	5.26	0.46	0.69	12.88	6.28	19.16
Tyees Men Early	0.23	2.94	0.38	0.5	11.66	6.47	18.13
Spartan	0.28	3.8	0.48	0.99	10.28	1.97	12.25

Table 4. Mineral Composition/Chemistry Data on Different Varieties of *Pyrus malus*.[29]

Variety	Sodium (mg/100 g)	Potassium (mg/100 g)	Phosphorus (mg/100 g)	Calcium (mg/100 g)	Magnesium (mg/100 g)	Iron (mg/100 g)	Vitamin A (mg/100 g)	Ascorbic Acid (mg/100 g)	Moisture (%)	Ash (%)	pH Value
Amri	8.69	82.52	15.09	15.31	8.11	5.63	0.94	3.44	80.5	1.29	4.07
Bluish Golden	8.31	75.19	8.34	16.29	14.88	7.47	0.89	3.78	83.39	3.07	3.78
Bonza	14.27	127.34	4.2	13.99	10.97	4.2	0.91	3.91	71.46	1.84	3.76
Discovery	11.01	109.36	16.34	20.79	14.62	7.23	0.95	4.2	75.91	1.14	3.87
Elster	10.14	96.94	11.25	19.89	11.7	7.09	0.99	5.93	79.72	1.51	3.75
Florina	12.8	114.63	16.98	17.85	12.19	2.46	0.8	4.28	75.4	1.71	4.19
Gala	11.4	82.27	13.31	21.99	15.58	7.6	0.79	3.22	81	1.29	4.22
Gloster 69	11.84	93.76	16.75	17.04	14.82	10.86	0.92	3.29	80.26	1.53	3.79
Golden Delicious	7.75	63.43	8.2	15.51	13.69	3.88	0.84	3.51	84.49	1.06	4.03
Granny Smith	10.37	73.82	13.1	14.55	11.73	10.73	0.87	3.91	82.71	1.73	3.94
Green Sleeves	9.12	75.19	13.59	7.87	4.44	10.92	0.92	2.74	81.75	1.61	3.79
Ida Red	7.7	65.76	9.44	9.06	6.26	3.08	0.94	3.32	84.6	1.63	3.78
Kaja	9.79	102.87	15.25	14.74	10.26	1.57	0.96	3.33	83.67	1.82	3.74
Kids Orange	8.2	72.98	11.97	16.41	14.4	1.25	0.99	7.38	83	1.26	4.44
Mushadi	11.2	64.89	9.58	13.28	7.12	7.08	0.9	5.33	84.25	2.01	4.08
Red Delicious	12.49	73.01	4.56	16.11	7.72	4.46	0.97	4.32	82.15	1.69	4.59
Samootree	10.49	84.69	16.66	13.74	11.63	1.96	0.79	3.95	78.12	1.45	3.93
Summer Red	6.4	79.72	12.24	17.21	10.74	5.16	0.88	1.38	87.1	2.6	3.89
Tyees Men Early	9.01	76.44	13.95	17.33	9.07	1.8	0.76	3.66	81.5	1.63	3.57
Spartan	10.37	73.82	13.14	14.55	11.73	10.73	0.81	2.99	82.71	1.73	4.07
Mean Values	10.06	84.43	12.19	15.67	11.08	5.78	0.89	3.8			
Median Values									81.95	1.63	3.91

Table 5. Composition Data on Pyrus Malus (Apple) Fiber.[33,34]

Composition	Impurities
Water (1.5%)	Ash (1.3%)
Fat (1.9%)	Not dioxin-like polychlorinated biphenyls (PCBs, 3 µg/kg; not detected)
Saturated fatty acids (0.3%, calculated as triglyceride)	Lead (0.36 mg/kg)
Monounsaturated fatty acids (0.6%, calculated as triglyceride)	Cadmium (< 0.01 mg/kg)
Polyunsaturated fatty acids (1%, calculated as triglyceride)	Mercury (< 0.30 mg/kg)
Total protein content (4.5%)	Arsenic (0.02 mg/kg)
Carbohydrate (25.5%, calculated)	Ochratoxin A (< 0.5 µg/kg)
Sucrose (enzymatical, 1609 mg/100 g)	Patulin (< 20 µg/kg)
Glucose (enzymatical, 2989 mg/100 g)	Aflatoxin B1 (< 0.7 µg/kg)
Fructose (enzymatical, 7406 mg/100 g)	Aflatoxin B2 (< 0.2 µg/kg)
Sodium (< 50 mg/kg)	Aflatoxin G1 (< 0.7 µg/kg)
Dietary fibers (65.3 g/100 g)	Aflatoxin G2 (< 0.2 µg/kg)
	Total aflatoxin (B1, B2, G1, and G2, not detected)
	Nitrite (as Na-nitrite, <3 mg/kg)
	Nitrate (as Na-nitrate, 29 mg/kg)
	Total nitrite/nitrate (as $NaNO_2$, 23.5 mg/kg)
	Total nitrite/nitrate (as $NaNO_3$, 29 mg/kg)
	Pesticide Residues
	Boscalid (0.164 mg/kg)
	Chlorantraniliprole (0.024 mg/kg)
	Chlorpyrifos (0.072 mg/kg)
	Cyprodinil (0.032 mg/kg)
	Etofenprox (0.020 mg/kg)
	Fenoxycarb (0.032 mg/kg)
	Fludioxonil (0.028 mg/kg)
	Hexythiazox (0.020 mg/kg)
	Indoxacarb (0.020 mg/kg)
	Methoxyfenozide (0.052 mg/kg)
	Myclobutanil (0.028 mg/kg)
	Pendimethalin (0.020 mg/kg)
	Phosalon (0.020 mg/kg)
	Primicarb (0.032 mg/kg)
	Propargite (0.080 mg/kg)
	Pyraclostrobin (0.116 mg/kg)
	Tebufenpyrad (0.024 mg/kg)
	Trifloxystrobin (0.160 mg/kg)
	Captan (0.628 mg/kg)
	Iprodion (0.036 mg/kg)

Table 6. Fatty Acid Composition Data on Apple Seed Extract.[45]

Fatty Acids	% Composition
Butyl linoleate	1.50%
2-Dodecenal	< 0.2%
Deca-2,4-dienal	0.49%
Ethyl linoleate	4.31%
Ethyl oleate	< 0.2%
Ethyl palmitate	0.56%
Ethyl stearate	< 0.2%
Hexyl hexanoate	0.54%
Hexyl linoleate	3.30%
Hexyl octanoate	0.49%
Hexyl palmitate	0.61%
Linoleic acid	51.15%
Linolenic acid	5.60%
Methyl docosanoate	0.72%
Methyl eicosanoate	2.18%
Methyl eicosenoate	1.05%
Methyl heneeicosenoate	< 0.2%
Methyl heptadecanoate	0.28%
Methyl linoleate	37.71%
Methyl linolenate	5.60%
Methyl nonodecanoate	< 0.2%
Methyl nonanoate	< 0.2%
Methyl octanoate	< 0.2%
Methyl oleate	4.12%
Methyl palmitate	9.93%
Methyl palmitoleate	< 0.2%
Methyl pentadecanoate	< 0.2%
Methyl pentanoate	< 0.2%
Methyl stearate	4.33%
Methyl tetracosanoate	< 0.2%
Methyl tricosanoate	< 0.2%
Myristyl myristate	< 0.2%
Nonacosane	3.59%
Oleic acid	4.12%
Palmitic acid	10.49%
Squalene	3.40%
Stearic acid	4.33%
Tricosene	4.29%
Total fatty acids	80.91%

Table 7. Current Frequency and Concentration of Use According to Duration and Type of Exposure.[2,46]

	Pyrus Malus (Apple) Fruit Extract		**Malus Domestica (Apple) Fruit Extract**		**Pyrus Malus (Apple) Fruit**	
	# of Uses	Conc. (%)	# of Uses	Conc. (%)	# of Uses	Conc. (%)
Totals/Conc. Range	125	0.000002-1	382	0.8	NR	0.00005-0.03
Duration of Use						
Leave-On	116	0.000002-1	283	NR	NR	0.00005
Rinse off	9	0.000075-0.75	98	0.8	NR	0.0018-0.03
Diluted for (bath) Use	NR	0.1	1	NR	NR	NR
Exposure Type						
Eye Area	4	0.018-0.29	28	NR	NR	NR
Incidental Ingestion	3	0.001-0.0036	6	NR	NR	NR
Incidental Inhalation- Sprays	106	0.00083-0.1	1	NR	NR	NR
Incidental Inhalation- Powders	106	0.00003-0.12**	1	NR	NR	0.00005
Dermal Contact	121	0.00003-1	90	0.8	NR	0.00005
Deodorant (underarm)	1*	NR	NR	NR	NR	NR
Hair - Non-Coloring	NR	0.0002-0.75	73	NR	NR	0.0018-0.03
Hair-Coloring	NR	0.1	NR	NR	NR	NR
Nail	NR	0.000002-0.00007	1	NR	NR	NR
Mucous Membrane	7	0.001-0.1	19	NR	NR	NR
Baby Products	NR	NR	NR	NR	NR	NR
	Pyrus Malus (Apple) Fruit Water		**Pyrus Malus (Apple) Juice**		**Malus Domestica (Apple) Juice**	
	# of Uses	Conc. (%)	# of Uses	Conc. (%)	# of Uses	Conc. (%)
Totals/Conc. Range	NR	0.008-9	NR	0.0003-0.7	11	NR
Duration of Use						
Leave-On	NR	0.008-9	NR	0.07	2	NR
Rinse off	NR	0.008-0.5	NR	0.0003-0.7	9	NR
Diluted for (bath) Use	NR	NR	NR	0.007	NR	NR
Exposure Type						
Eye Area	NR	0.2	NR	NR	2	NR
Incidental Ingestion	NR	0.008	NR	NR	NR	NR
Incidental Inhalation- Sprays	NR	NR	NR	NR	NR	NR
Incidental Inhalation- Powders	NR	0.76-9**	NR	0.07**	NR	NR
Dermal Contact	NR	0.008-9	NR	0.0025-0.07	1	NR
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	NR	NR	NR	0.0003-0.07	7	NR
Hair-Coloring	NR	NR	NR	NR	1	NR
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	NR	0.008	NR	0.0025-0.007	1	NR
Baby Products	NR	NR	NR	NR	NR	NR
	Pyrus Malus (Apple) Peel Powder		**Pyrus Malus (Apple) Root Extract**		**Pyrus Malus (Apple) Seed Extract**	
	# of Uses	Conc. (%)	# of Uses	Conc. (%)	# of Uses	Conc. (%)
Totals/Conc. Range	2	NR	4	NR	16	0.001-0.6
Duration of Use						
Leave-On	1	NR	1	NR	13	0.001-0.6
Rinse off	1	NR	NR	NR	3	NR
Diluted for (bath) Use	NR	NR	3	NR	NR	NR
Exposure Type						
Eye Area	NR	NR	NR	NR	2	0.6
Incidental Ingestion	NR	NR	NR	NR	2	0.001
Incidental Inhalation- Sprays	1***	NR	2***	NR	2***	NR
Incidental Inhalation- Powders	1***	NR	2***	NR	2***	NR
Dermal Contact	1	NR	4	NR	9	0.0015-0.6
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	NR	NR	NR	NR	NR	NR
Hair-Coloring	1	NR	NR	NR	NR	NR
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	NR	NR	2	NR	2	0.001
Baby Products	NR	NR	NR	NR	NR	NR

Table 7. Current Frequency and Concentration of Use According to Duration and Type of Exposure.[2,46]

	Pyrus Malus (Apple) Flower Extract		Pyrus Malus (Apple) Pectin Extract		Malus Domestica (Apple) Fruit Water	
	# of Uses	Conc. (%)	# of Uses	Conc. (%)	# of Uses	Conc. (%)
Totals/Conc. Range	NR	0.0005	9	NR	113	2
Duration of Use						
Leave-On	NR	0.0005	5	NR	106	2
Rinse off	NR	NR	4	NR	7	NR
Diluted for (bath) Use	NR	NR	NR	NR	NR	NR
Exposure Type						
Eye Area	NR	NR	NR	NR	1	NR
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation- Sprays	NR	0.0005	3*	NR	2	2
Incidental Inhalation- Powders	NR	NR	1	NR	2	NR
Dermal Contact	NR	0.0005	5	NR	105	2
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	NR	NR	1	NR	NR	NR
Hair-Coloring	NR	NR	NR	NR	NR	NR
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	NR	NR	NR	NR	2	NR
Baby Products	NR	NR	1	NR	NR	NR
	Malus Domestica (Apple) Oil		**Malus Domestica (Apple) Stem Extract**		**Malus Domestica (Apple) Fruit Cell Culture Extract**	
	# of Uses	Conc. (%)	# of Uses	Conc. (%)	# of Uses	Conc. (%)
Totals/Conc. Range	1	NR	1	2	29	0.00001-1
Duration of Use						
Leave-On	1	NR	1	2	27	0.00001-1
Rinse off	NR	NR	NR	NR	2	0.00001-0.2
Diluted for (bath) Use	NR	NR	NR	NR	NR	NR
Exposure Type						
Eye Area	1	NR	NR	NR	9	0.009-0.089
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation- Sprays	NR	NR	1***	NR	9***	NR
Incidental Inhalation- Powders	NR	NR	1***	NR	9***	0.0001-1
Dermal Contact	1	NR	1	NR	28	0.00001-1
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	NR	NR	NR	NR	NR	0.18-0.2
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
	Malus Domestica (Apple) Fiber					
	# of Uses	Conc. (%)				
Totals/Conc. Range	3	NR				
Duration of Use						
Leave-On	3	NR				
Rinse off	NR	NR				
Diluted for (bath) Use	NR	NR				
Exposure Type						
Eye Area	NR	NR				
Incidental Ingestion	NR	NR				
Incidental Inhalation- Sprays	3***	NR				
Incidental Inhalation- Powders	3***	NR				
Dermal Contact	23	NR				
Deodorant (underarm)	NR	NR				
Hair - Non-Coloring	NR	NR				
Hair-Coloring	NR	NR				
Nail	NR	NR				
Mucous Membrane	NR	NR				
Baby Products	NR	NR				

Table 7. Current Frequency and Concentration of Use According to Duration and Type of Exposure.[2,46]

NR = Not Reported; Totals = Rinse-off + Leave-on Product Uses.

*It is possible that these products may be sprays, but it is not specified whether the reported uses are sprays.

**It is possible that these products may be powders, but it is not specified whether the reported uses are powders.

***Not specified whether a powder or spray, so this information is captured for both categories of incidental inhalation.

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

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