Safety Assessment of *Olea europaea* (Olive)-Derived Ingredients as Used in Cosmetics

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All interested persons are provided 60 days from the above release date (i.e., August 26, 2023) to comment on this safety assessment, and to identify additional published data that should be included or provide unpublished data which can be made public and included.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 All unpublished data submitted to the Cosmetic Ingredient Review (CIR) will be discussed in open meetings, will be available for review by any interested party, and may be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 Executive Director, Dr. Bart Heldreth.

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ABBREVIATIONS

ALP = alkaline phosphatase
CAE = catechin equivalents
CIR = Cosmetic Ingredient Review
Council = Personal Care Products Council
CPSC = Consumer Product Safety Commission
DART = developmental and reproductive toxicity
dw = dry weight
ECE = epicatechin equivalents
EPA = Environmental Protection Agency
FDA = Food and Drug Administration
FEMA = Flavor and Extract Manufacturers Association
GAE = gallic acid equivalents
HRIPT = human repeated-insult patch test
GRAS = generally recognized as safe
HS-SPME-GC-FID = headspace solid-phase micro-extraction coupled with gas chromatography with flame ionized detector
IC_{50} = half-maximal inhibitory concentration
IgE = immunoglobulin E
MEA = monoethanolamine
LDH = lactate dehydrogenase
LOAEL = lowest-observable-adverse-effect level
LPS = lipopolysaccharide
NOAEL = no-observable-adverse-effect level
OECD = Organization for Economic Co-Operation and Development
Panel = Expert Panel for Cosmetic Ingredient Safety
PEG = polyethylene glycol
PMNC = polymorphonuclear cells
QAE = quillaja equivalents
QE = quercetin equivalents
RE = rutin equivalents
SIOPT = single-insult occlusive patch test
TG = test guideline
US = United States
VCRP = Voluntary Cosmetic Registration Program
wINCI *Dictionary* = web-based *International Cosmetic Ingredient Dictionary and Handbook*

ABSTRACT

The Expert Panel for Cosmetic Ingredient Safety (Panel) assessed the safety of 23 *Olea europaea* (olive)-derived ingredients, most of which are reported to function as skin-conditioning agents in cosmetic products. Industry should minimize impurities that could be present in cosmetic formulations, such as heavy metals and pesticide residues, according to limits set by the US Food and Drug Administration (FDA) and the Environmental Protection Agency (EPA). The Panel reviewed all relevant data and concluded that 16 *Olea europaea* (olive)-derived ingredients (i.e., the fruit-, leaf-, husk-, and seed-derived ingredients) are safe in cosmetics in the present practices of use and concentration described in this safety assessment. Additionally, the Panel also concluded that the available data are insufficient to make a determination of safety for the remaining 7 *Olea europaea* (olive)-derived ingredients under the intended conditions of use in cosmetic formulations.

INTRODUCTION

This assessment reviews the safety of the following 23 *Olea europaea* (olive)-derived ingredients as used in cosmetic formulations:

Hydrolyzed Olive Fruit
Hydrolyzed Olive Fruit Extract
Hydrolyzed Olive Leaf Extract
Olea Europaea (Olive) Bark Extract
Olea Europaea (Olive) Branch Extract
Olea Europaea (Olive) Bud Extract
Olea Europaea (Olive) Flower Extract
Olea Europaea (Olive) Flower Water
Olea Europaea (Olive) Fruit
Olea Europaea (Olive) Fruit Extract
Olea Europaea (Olive) Fruit Juice
Olea Europaea (Olive) Fruit Juice Extract
Olea Europaea (Olive) Fruit Unsaponifiables
Olea Europaea (Olive) Fruit Water
Olea Europaea (Olive) Husk Powder
Olea Europaea (Olive) Leaf
Olea Europaea (Olive) Leaf Extract
Olea Europaea (Olive) Leaf Powder
Olea Europaea (Olive) Leaf Water
Olea Europaea (Olive) Sap Extract
Olea Europaea (Olive) Seed
Olea Europaea (Olive) Seed Powder
Olea Europaea (Olive) Wood Extract

Most of the *Olea europaea* (olive)-derived ingredients detailed in this safety assessment are reported to function in cosmetics as skin-conditioning agents (emollient, humectant, or miscellaneous), according to the web-based *International Cosmetic Ingredient Dictionary and Handbook* (wINCI; *Dictionary*; see Table 1).[1] Olea Europaea (Olive) Husk Powder and Olea Europaea (Olive) Seed Powder are reported to only function as abrasives, and Olea Europaea (Olive) Flower Water and Olea Europaea (Olive) Fruit Juice are reported to only function as antioxidants. The reported function as a skin bleaching agent (for Olea Europaea (Olive) Fruit Extract and Olea Europaea (Olive) Leaf Extract) is considered a drug effect in the United States (US) and, therefore, is not addressed in this assessment as it is not under the purview of the Panel.

The Expert Panel for Cosmetic Ingredient Safety (Panel) has previously reviewed the safety of *Olea europaea* (olive) fruit oil, *Olea europaea* (olive) oil unsaponifiables, hydrogenated olive oil, hydrogenated olive oil unsaponifiables, potassium olivate, sodium olivate, *Olea europaea* (olive) husk oil, and olive acid.[2] The Panel concluded these ingredients are safe in the present practices of use and concentration, as described in the safety assessment.

Some of the ingredients 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.

Botanicals, such as *Olea europaea* (olive)-derived ingredients, may contain hundreds of constituents. Thus, in this assessment, the Panel will assess the safety of each of these ingredients as a whole, complex substance; toxicity from single components may not predict the potential toxicity of botanical ingredients.

This safety assessment includes relevant published and unpublished data that are available for each endpoint that is evaluated. Published data are identified by conducting an extensive search of the world's literature, last performed April 2023. A listing of the search engines and websites that are used and the sources that are typically explored, as well as the endpoints that the Panel typically evaluates, is provided on the Cosmetic Ingredient Review (CIR) website (https://www.cir-safety.org/supplementaldoc/preliminary-search-engines-and-websites; https://www.cir-safety.org/supplementaldoc/cir-report-format-outline). Unpublished data are provided by the cosmetics industry, as well as by other interested parties.

Note: The cosmetic ingredient names, according to the *Dictionary*, are written as listed above, without italics and without abbreviations. When referring to the plant from which these ingredients are derived, the standard scientific practice of using italics will be followed (i.e., *Olea europaea*). Often in the published literature, the general name "olive" is used, and it is not known how the substance being tested compares to the ingredient as used in cosmetics. Therefore, if it is not known whether the material being discussed is a cosmetic ingredient, the generic terminology, in all lowercase (e.g., olive leaf extract or olive fruit), will be used. However, if it is known that the material is a cosmetic ingredient, the naming convention provided in the *Dictionary* (e.g. Olea Europaea (Olive) Leaf Extract or Olea Europaea (Olive) Fruit) will be used.

CHEMISTRY

Definition and Plant Identification

The definitions of the ingredients included in this review are provided in Table 1.[1] The generic CAS number for several olive ingredients in this report is 84012-27-1.

Olea europaea L. is an evergreen tree or shrub native to the Mediterranean region of the world, and is one of the earliest domesticated fruit trees in the world, used for its oil, edible fruit, and medicinal properties since antiquity.[3-5] There are at least 30 species within the genus *Olea*, but only *Olea europaea* is cultivated.[6]

Table 2 lists the generic definitions of the parts of plants that are most pertinent to the ingredients in this report.[1] The olive tree is short and thick, averaging about 10 m in height.[7] The tree has a large diameter trunk and is bent and twisted. Branches are reedy with opposite branchlets, and the leaves are shortly-stalked, narrow, oblong, and leathery, and are pale green on the top-side and silvery-whitish on the bottom-side in color. The bark is pale grey in color. The fruit is small, ovoid, and blackish-violet when ripe. The fruit and seed, or drupe, is comprised of an external epicarp, a middle mesocarp, and an internal endocarp, which becomes totally lignified at the end of the epi-mesocarp expansion growth.[8] The seed coat encloses the endosperm and embryo.

Chemical Properties

Chemical properties for the *Olea europaea* (olive)-derived ingredients are summarized in Table 3. Specific gravity (at 25º C) for Olea Europaea (Olive) Fruit Extract (prepared in butylene glycol/water) and Olea Europaea (Olive) Leaf Extract (prepared in water) were reported to be 1.02 and 1.00, respectively.[9,10] Both of these preparations are reported to be soluble in any proportion of water.

Method of Manufacture

Unpublished data were submitted describing methods of manufacture for some ingredients. For the general methodologies of processing *Olea europaea* (olive)-derived ingredients described below, it is unknown if these methodologies apply to cosmetic ingredient manufacturing. In several cases, the definition of the ingredients, as given in the *Dictionary*, provides insight as to the method of manufacture.

Olea Europaea (Olive) Flower Water

Olea Europaea (Olive) Flower Water is obtained through steam distillation of the flowers of *Olea europaea*.[1] (No further details are provided.)

Olea Europaea (Olive) Fruit Extract

A standardized aqueous olive pulp (fruit) extract was reported to be prepared as a byproduct during the processing of the pulp of olives (*Olea europaea* L.) for oil extraction.[11] The extract was produced as a freeze-dried powder.

Another supplier reported that Olea Europaea (Olive) Fruit Extract is manufactured by extracting olive fruit with specified eluent/s (water, butylene glycol, safflower seed oil, glycerin, and/or propylene glycol) under appropriate temperature conditions, to yield a concentrate.[9] The concentrate is then blended with the desired diluent/s and preservation system to produce the final ingredient. The ingredient is evaluated for physicochemical properties according to the specification requirements for the batch to be released. In addition, the concentrate is also evaluated for contaminants and physicochemical properties as needed.

A supplier reported that it sells olive oil under the INCI name Olea Europaea (Olive) Fruit Extract.[12] The material can be extracted through several processes, including pressing and filtering, using hexane, or through super critical carbon dioxide extraction.

Olea Europaea (Olive) Fruit Unsaponifiables

Olea Europaea (Olive) Fruit Unsaponifiables is the remaining fraction of olive fruit remaining after fractional distillation.[1] (No further details are provided.)

Olea Europaea (Olive) Fruit Water

Olea Europaea (Olive) Fruit Water is obtained through steam distillation of the fruits of *Olea europaea*.[1] (No further details are provided.)

Olea Europaea (Olive) Husk Powder

Olea Europaea (Olive) Husk Powder is obtained from drying and grinding the husks of *Olea europaea*.[1] (No further details are provided.)

Olive husk may also be called olive pomace or olive cake.[13] Olive husk is the solid residue obtained after olive oil extraction. It consists of the crushed hull, the skin, and the pulp of the olive, as well as some oil and water.

Olea Europaea (Olive) Juice Extract

A supplier reported that Olea Europaea (Olive) Juice Extract is produced from concentrated olive juice that is extracted with 50 vol% 1,3-butylene glycolic solution.[14] The resulting material then undergoes sedimentation, filtration, and adjustment prior to packaging.

Olea Europaea (Olive) Leaf Extract

Olea Europaea (Olive) Leaf Extract is manufactured by extracting olive leaves with specified eluent/s (water, butylene glycol, safflower seed oil, glycerin and/or propylene glycol) under appropriate temperature conditions, to yield a concentrate.[10] The concentrate is then blended with the desired diluent/s and preservation system to produce the final ingredient. The ingredient is evaluated for physicochemical properties according to the specification requirements for the batch to be released. In addition, the concentrate is also evaluated for contaminants and physicochemical properties as needed.

A supplier reported that Olea Europaea (Olive) Leaf Extract is manufactured by extracting the leaves of *Olea europaea* with water/glycerin or sunflower oil. The process involves maceration and filtration.[15]

Another supplier reported that Olea Europaea (Olive) Leaf Extract is produced by extracting dried raw olive leaves with 50 vol% ethanol solution and concentrating.[14] The resulting material is then dissolved in 50 vol% 1,3-butylene glycolic solution and then undergoes sedimentation, filtration, and adjustment prior to packaging.

A microwave-assisted aqueous extract of olive leaves produced for research was made by first oven-drying leaves before grinding them and running them through a metal mesh sieve.[16] The resulting material was then microwaved with distilled water, vacuum-filtered, and lyophilized.

Olea Europaea (Olive) Leaf Powder

Olea Europaea (Olive) Leaf Powder is obtained from drying and grinding the leaves of *Olea europaea*.[1] A supplier reported that Olea Europaea (Olive) Leaf Powder is manufactured by grinding dry olive leaves prior to sieving and sterilization (by gamma ray or heat).[17]

Olea Europaea (Olive) Leaf Water

Olea Europaea (Olive) Leaf Water is obtained through steam distillation of the leaves of *Olea europaea*.[1] A supplier reported that Olea Europaea (Olive) Leaf Water is manufactured through hydrodistillation of the leaves of *Olea europaea* in water.[15]

Olea Europaea (Olive) Seed Powder

Olea Europaea (Olive) Seed Powder is obtained from drying and grinding the seeds of *Olea europaea*.[1] (No further details are provided.)

Composition and Impurities

The composition of constituents of *Olea europaea* (olive)-derived ingredients can vary annually, and is dependent on the cultivar, production area, climate, season, and soil characteristics.[18,19] Composition may also vary with use of fresh versus dried raw materials.[20] Oleuropein is the main phenolic component of the unprocessed fruit and leaves of *Olea europaea* L.[18] Content of oleuropein in leaves is dependent on the leaf tissue conditions (i.e., fresh, frozen, dried, or lyophilized). One study of leaf extracts with different solvents and two different cultivars found the total phenolic content, total flavonoids, and oleuropein content to be similar between cultivars, but it was noted that the leaves had been harvested from the same location in Australia.[21]

Olea Europaea (Olive) Bark Extract

Mineral content of the powdered bark of a subspecies of *Olea europaea* was 18.31 ppm calcium, 9.63 ppm magnesium, 8.94 ppm potassium, 0.22 ppm iron, 0.08 pm copper, 0.03 ppm lead, and below the threshold of detection for zinc.[22] From phytochemical analysis, the primary constituents of the powdered bark were reported as 36.01% total proteins, 0.82% total lipids, and 43.68% total carbohydrates. The yield of secondary constituents, described in Table 4, varied with the type of solvent used; for example, total flavonoids was 64.44 mg/g for a chloroform extract and 8.11 mg/g for a water extract.

In crude stem bark extracts of a subspecies of *Olea europaea*, the total phenolic content of methanol, ethanol, and chloroform extracts were 399, 351, and 312 µg/mg (catechol equivalents), respectively.[23] A methanol extract of the bark of a subspecies of *Olea europaea* was reported to have the following classes of bioactive compounds: alkaloids, tannins, and flavonoids.[24] Further description was not provided.

Olea Europaea (Olive) Bud Extract and Olea Europaea (Olive) Flower Extract

Phenolic compounds identified in both the methanol extracts of dried buds and open flowers of one Tunisian olive cultivar included secoiridoids, flavonoids, simple phenols, cinnamic acid derivatives, and lignans.[25] Secoiridoids were measured at a higher percentage of total phenols in open flowers (41.7%) than in buds (30.5%). Conversely, flavonoids were

measured at a higher percentage of total phenols in buds (38.1%) than in open flowers (26.7%). Cinnamic acid derivative and simple phenols were comparable. Lignans were measured at 0.4% and 1.0% of total phenols in buds and open flowers, respectively.

Olea Europaea (Olive) Flower Extract

In an 80% ethanol extract of olive flowers, phenolic acids (vanillic acid, *p*-coumaric acid, vanillin, caffeic acid), flavonoids (luteolin, apigenin, rutin, diosmetin), simple phenols (hydroxytyrosol, tyrosol), secoiridoids (oleuropein, ligstroside), and the cinnamic acid derivative, verbascoside, were identified using liquid chromatography with tandem mass spectrometry.[26] The flavonoids (9.4 mg/g dry matter) and secoiridoids (7.7 mg/g dry matter) comprised most of the phenols; total phenols were determined to be 22.7 mg/g dry matter.

Olea Europaea (Olive) Fruit

Constituents of olive fruit are reported to include monounsaturated fatty acids, aliphatic and triterpene alcohols, sterols, hydrocarbons, and several antioxidants.[27] Pentacyclic triterpenes in olive fruit include maslinic acid (1.2 - 1.8 mg/g dry weight (dw)) and oleanolic acid (0.4 - 0.6 mg/g dw), which are exclusively located in the epicarp and decrease as the fruit ripen.[28] Total phenolics in 10 types of commonly consumed olives ranged from 0.21 mg gallic acid equivalents (GAE)/g to 2.20 mg GAE/g.[29]

Through headspace solid-phase micro-extraction coupled with gas chromatography with flame ionized detector (HS-SPME-GC-FID) of fruit homogenates, the ethanol content in olive fruit was found to vary between different cultivars (0.56 to 58 mg/kg for 3 different cultivars).[30] Regardless of cultivar, ethanol content of fruit increased during the ripening process.

Olea Europaea (Olive) Fruit Extract

A comparison of the constituent composition between cultivars and production area for olive fruit extracts is found in Table 5.[31] Total polyphenol content for Italian cultivars ranged from 182.35 - 290.21 mg GAE/g, while for Algerian cultivars, the total polyphenol content ranged from 147.13 - 272.83 mg GAE/g.

Several biphenols have been identified in methanol:water extracts of drupes, including oleuropein, hydroxytyrosol, tyrosol, vanillin, apigenin, luteolin, and quercetin.[32] Oleuropein, tyrosol, and hydroxytyrosol content in these extracts ranged as follows, respectively: < 0.037 - 145 mg/kg, < 0.045 - 40.3 mg/kg, and < 0.048 - 426 mg/kg. An ethanolic extract of olive fruit was approximately 11.25% hydroxytyrosol.[33]

Ethanol:water extracts (80:20) of olive fruit were analyzed for hydroxycinnamic acids and flavonoids.[34] Measured values of hydroxycinnamic acids included trace amounts of ferulic acid and *p*-coumaric acid, trace to1.0 mg/kg dw caffeic acid, and 3.6 - 60.1 mg/kg dw chlorogenic acid. Flavonoids measured values were 36.7 - 583.9 mg/kg dw rutin, 0.5 - 2.7 mg/kg quercetin, 20.9 - 121.0 mg/kg luteolin, 1.6 - 8.7 mg/kg luteolin-7-*O*-rutinoside, and trace to 1.3 mg/kg naringenin.

A commercial olive fruit extract (prepared for analysis in 50% ethanol) was determined to have a total phenol content of 4.64 mg GAE/g and a total flavonoid content of 24.17 mg quercetin equivalent (QE)/g.[35] The major phenolic components included hydroxytyrosol, elenolic acid, verbascoside, luteolin-7-*O*-glucoside, secoiridoids, and oleuropein.

A standardized aqueous olive pulp (fruit) extract powder was composed of 98% - 99% dry solids, including 1% - 2% citric acid and 6% polyphenols.[11] Other constituents included protein, fat, and carbohydrates. Of the polyphenols, the major constituent was hydroxytyrosol (50% - 70%), with oleuropein (5% - 10%), tyrosol (0.3%), and oleuropein aglycone + gallic acid (~20% combined) also present.

A supplier reported the microbial plate count for Olea Europaea (Olive) Fruit Extract prepared in butylene glycol and water to be less than 100 organisms/g.[9] No further details provided.

Olea Europaea (Olive) Husk Powder

Raw olive husk contains the crushed hull of the fruit, skin, pulp, water (~25%) and residual oil (4.5 - 9%).[13] Cis-Oleic acid is the most abundant fatty acid. Olive husk contains small amounts of nitrogen (crude protein) and a high proportion of fiber, consisting of 10% hemicellulose, 15% cellulose, and 27% lignin. Additional constituents include soluble phenols, calcium, magnesium, potassium, sodium, and iron. Lead, cadmium, chromium, and mercury content is reported to be below 1 mg/kg.

Olea Europaea (Olive) Juice Extract

A supplier reported that Olea Europaea (Olive) Juice Extract is comprised of saccharides and tannin.[14] Heavy metals content is not more than 20 ppm and arsenic content is not more than 2 ppm. No further details provided.

Olea Europaea (Olive) Leaf

Pentacyclic triterpenes found in olive leaf include oleanolic acid (29.2 - 34.5 mg/g), maslinic acid (4.8 - 7.3 mg/g), ursolic acid (2.0 - 2.5 mg/g), erythrodiol (0.8 - 1.5 mg/g), and uvaol (0.7 - 1.5 mg/g). These quantities change in abundance and profile as leaves mature.[28]

Olea Europaea (Olive) Leaf Extract

Olive leaf extract contains several biphenols, including oleuropein, tyrosol, hydroxytyrosol, apigenin, luteolin, quercetin, pinoresinol, catechin, ferulic acid, gallic acid, and vanillic acid.[32,36] Yields of constituents are dependent on solvent type and extraction methods. For example, oleuropein content of olive leaf extract in methanol:water (80:20, v/v) ranged from < 0.00013 – 0.29 mg/g,[32] while the oleuropein content from a microwave assisted aqueous extract was 11.59 mg/g (dry base),[16] and an ultrasound-assisted extraction of olive leaves produced 13.39 mg/g oleuropein.[37]

Constituent levels in olive leaves by extract type, cultivar, and production area are described in Table 6.[3,38] Ethanolic extracts of Italian olive cultivars had higher levels of oleuropein than methanolic extracts of Tunisian olive cultivars (7.49 - 30.46 g/kg dw versus 0.246 - 0.520 g/kg dw, respectively). Total phenolic content for the ethanolic extracts of Italian cultivars ranged from 11.39 - 48.62 g GAE/kg dw, while the methanolic extracts of Tunisian cultivars ranged from 18.96 - 47.47 g GAE/kg and total flavonoid content ranged from 3.08 - 7.29 mg catechin equivalents (CAE)/g.

The major phenolic compounds in methanolic leaf extracts of Tunisian olive cultivars were identified as hydroxytyrosol, tyrosol, 4-hydroxybenzoic acid, rutin, luteolin-7-*O*-glucoside, apigenin-7-*O*-glucoside, oleuropein, apigenin, and catechin hydrate.[3] Aqueous extracts of leaves from Tunisian olive cultivars had total phenolic content of 480.3 - 546.1 mg GAE/g, flavonoid content of 506.4 - 605.3 mg CAE/g, and flavonol content of 73.0 - 109.4 mg rutin equivalents (RE)/g.[39]

Aqueous extracts of olive leaves from Turkey yielded a total phenolic content of 92.13 mg GAE/g, a total flavonoid content of 21.64 mg RE/g and a total saponin content of 180.04 quillaja equivalents (QAE)/g.[40] In a methanol extract (70:30 methanol: water) of olive leaves, total phenols were 23.52 mg GAE/g dw, ortho-diphenols were 58.74 mg GAE/g dw, total flavonoids were 16.96 mg CAE/g dw, and tannins were 7.09 mg epicatechin equivalents (ECE)/g dw.[41]

A commercial olive leaf extract (prepared for analysis in 50% ethanol) was determined to have a total phenol content of 7.87 mg GAE/g and a total flavonoid content of 32.03 mg QE/g.[35] The major phenolic components included hydroxytyrosol, oleuropein aglycone-1, elenolic acid, verbascoside, luteolin-7-*O*-glucoside, flavonoid glucosides, and oleuropein. In another ethanolic extract of olive leaves, hydroxytyrosol was measured at 7.26%.[33]

An aqueous extract of olive leaves was determined to have the following soluble carbohydrates: myo-inositol, mannitol, galactose, glucose, fructose, sucrose, raffinose, and stachyose.[42] Of these carbohydrates, glucose and mannitol were present at the highest percentages (49.2% and 41.0%, respectively).

A supplier reported that Olea Europaea (Olive) Leaf Extract is comprised of organic acid and tannin.[14] Heavy metals content is not more than 20 ppm and arsenic content is not more than 2 ppm. Oleuropein content is not less than 0.03% w/v. No further details provided.

Another supplier reported that the following heavy metals were not detected at respective reporting limits for Olea Europaea (Olive) Leaf Extract (testing conducted on concentrate in alcohol base): antimony, arsenic, cadmium, chromium, iron, lead, mercury, and nickel.[10] Additionally, no residual pesticides were detected. The microbial plate count for Olea Europaea (Olive) Leaf Extract prepared in water was reported to be less than 100 organisms/g.

Olea Europaea (Olive) Leaf Powder

A supplier reported that Olea Europaea (Olive) Leaf Powder is 100% olive leaves.[17] No further details provided.

Olea Europaea (Olive) Sap Extract

Constituents of olive sap include terpenoids, phytohormones, alkaloids, sterols/steroids, retinols/retinoids, tocopherols, and carotenoids.[19]

Olea Europaea (Olive) Seed

Methanol and methanol/water extracts of olive stones and seeds were found to have hydroxycinnamic acid derivatives, phenolic alcohols, flavonoids and flavonoid glucosides, secoiridoids, fatty acids, and terpenes.[43] The main bioactive component of olive seeds has been identified as hydroxytyrosol.[44]

Olea Europaea (Olive) Wood Extract

The main constituents of olive wood chips extracted with ethyl acetate have been identified as tyrosol, hydroxytyrosol, cycloolivil, ligustroside, oleuropein, and 7-deoxyloganic acid.[45] Secoiridoids determined from the same extract are as follows: oleuropein-3”-methyl ether (0.7 mg/g), 7”(*S*)-hydroxyoleuropein (2.8 mg/g), jaspolyanoside (2.2 mg/g), ligustroside 3’-*O*-β-D-glucoside (1.3 mg/g), jaspolyoside (3.3 mg/g), isojaspolyoside A (0.6 mg/g), and oleuropein 3’-*O*-β-D-glucoside (0.7 mg/g).[46]

USE

Cosmetic

The safety of the cosmetic ingredients addressed in this assessment is evaluated based on data received from the US FDA and the cosmetics industry on the expected use of these ingredients in cosmetics and does not cover their use in airbrush

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

According to 2023 VCRP survey data, Olea Europaea (Olive) Leaf Extract has the highest frequency of use; it is reported to be used in 170 formulations, with a majority of uses in leave-on skin care preparations (Table 7).[47] Olea Europaea (Olive) Fruit Extract is reported to be used in 124 formulations, also with the majority of uses in leave-on skin care preparations. All other in-use ingredients are reported to be used at much lower numbers. The results of the concentration of use surveys conducted by the Council in 2020 and 2023 indicate that Olea Europaea (Olive) Leaf Extract has the highest concentration of use in a leave-on formulation; it is used at up to 2% in suntan preparations.[48,49] The highest concentration of use reported for products resulting in rinse-off dermal exposure is 10% in Olea Europaea (Olive) Fruit Unsaponifiables in shaving cream. The 14 ingredients not in use, according to the VCRP and industry survey, are listed in Table 8.[47-49]

Some *Olea europaea* (olive)-derived ingredients may be incidentally ingested or be used near the eye or mucous membranes. For example, Olea Europaea (Olive) Fruit Extract is reported to be used in lipstick (0.24%), eye lotion and other eye makeup preparations (concentration not reported), and bar soaps and detergents (up to 0.11%).[47,48] Additionally, some of the ingredients are used in cosmetic sprays and powders and could possibly be inhaled; for example, Olea Europaea (Olive) Leaf Extract is used at 0.018% in hair spray and at 0.0002% in aerosol deodorant and Olea Europaea (Olive) Fruit Extract is used in face powders (no concentration reported).[47,48] In practice, as stated in the Panel's respiratory exposure resource document (https://www.cir-safety.org/cir-findings), most droplets/particles incidentally inhaled from cosmetic sprays would be deposited in the nasopharyngeal and tracheobronchial regions and would not be respirable (i.e., they would not enter the lungs) to any appreciable amount. There is some evidence indicating that deodorant spray products can release substantially larger fractions of particulates having aerodynamic equivalent diameters in the range considered to be respirable. However, the information is not sufficient to determine whether significantly greater lung exposures result from the use of deodorant sprays, compared to other cosmetic sprays. Conservative estimates of inhalation exposures to respirable particles during the use of loose powder cosmetic products are 400-fold to 1000-fold less than protective regulatory and guidance limits for inert airborne respirable particles in the workplace.

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

The *Olea europaea* (olive)-derived ingredients named in the report are not restricted from use in any way under the rules governing cosmetic products in the European Union.[50]

Non-Cosmetic

Different parts of the olive tree have been used for centuries for nutritional properties and protective health effects.[43] The leaves of the olive tree have been historically used as an herbal drug in folk medicine, with use as therapy for chronic conditions like gout, diabetes, and hypertension.[18,51,52] Leaves, fruit, and their constituents have been studied for health benefits such as antioxidant,[18,27,36,53] antimicrobial,[36,54-56] (including anti-malarial),[57] anti-inflammatory,[18,27,35,58,59] antiviral (including anti-HIV activity),[60] cardioprotective,[18,61] hepatoprotective,[62] neuroprotective[18,63,64], and anti-cancer effects[18,65] Olive leaves, extracts, and constituents have also been studied as potential treatments for diabetes (types 1 and 2),[66-68] hypertension,[69,70] and for protective effects against oxidative stress on kidneys and liver.[71] Additional therapeutic uses for olive leaf and olive fruit have been studied for the treatment of wounds,[72] intestinal morphological injuries,[27] and multiple sclerosis and other neurodegenerative diseases.[36] Olive drupes (fruit, pit and seed) have been studied for treating gastric disturbances,[44] reducing blood sugar, cholesterol, and uric acid;[43] and for protective effects on the tissues and functions of the liver, kidneys, and heart.[43,73] Olive pits (including the seed) have been used in folk medicine to treat gastric disturbances.[44] Olive bark and wood have been studied for antioxidant,[23,74] antidiabetic and anticancer activity,[22] as well as antimicrobial activity[23,24] (including anti-malarial).[75]

Olive leaves and fruit extracts have been studied for use in natural food preservation and packaging.[16,33,76,77] The Expert Panel for the Flavor and Extract Manufacturers Association (FEMA) generally recognized as safe (GRAS) program has provided recommended use levels for olive fruit extract as a flavor ingredient based on the average usual use level of 120 ppm and the average maximum use level of 720 ppm.[78]

TOXICOKINETIC STUDIES

No relevant toxicokinetic studies on *Olea europaea* (olive)-derived ingredients were found in the published literature, and unpublished data were not submitted. In general, toxicokinetics data are not expected to be found on botanical ingredients because each botanical ingredient is a complex mixture of constituents.

TOXICOLOGICAL STUDIES

Acute Toxicity Studies

Acute toxicity studies on *Olea europaea* (olive)-derived ingredients are summarized in Table 9. In mouse studies of olive stem bark extract, an aqueous hydrolyzed olive pulp (fruit) extract, and olive leaf extracts, the LD_{50} was greater than 2000 mg/kg, which was the maximum dose tested for each ingredient.[14,57,75,79] In rat studies, an aqueous hydrolyzed olive pulp (fruit) extract had an LD_{50} greater than 5000 mg/kg, and olive leaf extract had an LD_{50} greater than 2000 mg/kg.[79,80]

Short-Term and Subchronic Toxicity Studies

Repeated-dose oral toxicity studies on *Olea europaea* (olive)-derived ingredients are summarized in Table 10. No treatment-related mortalities were observed in rats that received olive fruit extract (up to 1381 mg/kg bw/d) or hydrolyzed olive pulp (fruit) extract (aqueous; up to 2000 mg/kg/d) via gavage for 90 d.[79,81] The lowest-observable-adverse-effect level (LOAEL) was 1381 mg/kg bw/d and the no-observable-adverse-effect level (NOAEL) was 691 mg/kg bw/d in the olive fruit extract study, and the NOAEL for the hydrolyzed olive pulp (fruit) extract was 2000 mg/kg/d. In studies of a proprietary olive leaf extract (0, 360, 600, 1000, or 2000 mg/kg/d) in rats, dose-dependent hyaline droplet nephropathy was observed in males in the 1000 and 2000 mg/kg dose groups, but not in lower dose males or in any females in a 14-d study.[82] No mortality, clinical signs of toxicity, or abnormalities in liver and kidneys were observed in a 28-d study with olive leaf extract (ethanol) at up to 400 mg/kg, but the concentration of blood urea nitrogen was significantly increased in males in the 100 and 400 mg/kg dose groups when compared to controls.[80] In a 42-d rat study with dietary concentrations of up to 0.9% olive leaf extract (aq.), livers had fatty changes and hepatocellular necrosis was observed in all test groups, but the effects were more prominent in the 0.7% and 0.9% dose groups.[4] Kidneys in the treated groups had streaky hemorrhages and congestion in the cortical region, with more severe hemorrhage in the two higher dose groups. The NOAEL in a 90-d rat study was the maximum test dose of 1000 mg/kg bw/d for a proprietary olive leaf extract.[82]

DEVELOPMENTAL AND REPRODUCTIVE TOXICITY (DART) STUDIES

DART studies on *Olea europaea* (olive)-derived ingredients are summarized in Table 11. In male rats treated at up to 450 mg/kg olive fruit extract (hydroalcoholic) for 48 d, a significant decrease in testicle weights (all treatment groups) and seminal vesicle weight (150 mg/kg dose group only) was observed, as were significant decreases in testosterone hormone levels, sperm counts, and sperm motility (all treatment groups for each end point).[83] Hydrolyzed olive pulp (fruit) extract (aqueous; up to 2000 mg/kg/d) produced no treatment-related mortalities in F_0 mature rats or F_1 rat pups, and produced no adverse effects in fertility or reproduction.[79] The NOAEL for developmental toxicity in rats was greater than 2000 mg/kg/d when dams received the same test material during gestation days 6 through 20.[11,79]

GENOTOXICITY STUDIES

In vitro and in vivo genotoxicity studies on *Olea europaea* (olive)-derived ingredients are summarized in Table 12. Mutagenic activity was observed in a bacterial reverse mutation assay of a hydrolyzed olive pulp (fruit) extract (aqueous; tested up to 5000 µg/plate with metabolic activation); however, inconsistencies between trials, antibacterial properties of the test material, and positive findings in only two concentrations complicated the interpretation of the findings.[79] Mutagenic activity was also observed in a chromosome aberration assay (aqueous; tested up to 1000 µg/ml) of the hydrolyzed olive pulp (fruit) extract when tested with metabolic activation; however, this test material was not mutagenic in an in vivo micronucleus assay (aqueous; tested up to 5000 mg/kg/d via gavage) in rats. A proprietary olive leaf extract was not considered genotoxic in a bacterial reverse mutation assay (tested up to 5000 µg/plate or in a mammalian chromosome aberration test (tested up to 1500 µg/ml) in V79 Chinese hamster lung cells.[82] A bacterial Vitotox™ test and an alkaline comet assay in human hepatic cells performed on different olive leaf extracts from Tunisia were negative in 3 of the 4 extracts tested (up to 5000 µg/ml); however, borderline genotoxicity was observed in the 4th extract.[84] A proprietary olive leaf extract was not genotoxic in an in vivo micronucleus assay (tested up to 200 mg/ml) in mice.[82]

CARCINOGENICITY STUDIES

Relevant carcinogenicity data for the *Olea europaea* (olive)-derived ingredients were not found in the published literature, and unpublished data were not submitted.

OTHER RELEVANT STUDIES

Cytotoxicity

Olea Europaea (Olive) Fruit Extract

The effects of the extract of olive fruit skins on cell proliferation and apoptosis was studied in HT-29 human colon cancer cells.[85] Olive fruit was extracted with chloroform and methanol. The pentacyclic triterpene profile of the extract was 73.25% maslinic acid, 25.75% oleanolic acid, 1% erythrodiol, and trace amounts of maslinic acid derivatives. Dose-dependent effects showed antiproliferative activity without displaying necrosis. Apoptosis was observed through microscopic changes in membrane permeability and detection of DNA fragmentation in cells that were incubated for 24 h with olive fruit extract. Caspase-3 was activated in a dose-dependent manner after a 24-h incubation, with up to 6-fold increased activity over the control cells. The production of superoxide anions in the cell mitochondria of the treated cells indicated that programmed cell death was induced by the intrinsic pathway. The authors concluded that olive fruit extract inhibited cell proliferation without cytotoxicity and the restoration of apoptosis in this study with human colon cancer cells.

Olea Europaea (Olive) Leaf Extract

In a cytotoxicity study, olive leaf extract was added to polymorphonuclear cells (PMNC) at a concentration of 320 µg/ml for 16 h after stimulation with 1 µg/ml of lipopolysaccharide (LPS).[36] The test material was extracted in ethanol. No significant effect on cell viability was observed when compared with cell culture with or without LPS stimulation. The test material was not cytotoxic.

DERMAL IRRITATION AND SENSITIZATION STUDIES

Dermal irritation and sensitization data for the *Olea europaea* (olive)-derived ingredients are summarized in Table 13. Olea Europaea (Olive) Leaf Extract, tested at 100% in an in vitro primary skin irritation study in accordance with Organization for Economic Co-Operation and Development (OECD) test guideline (TG) 439, was predicted to be a non-irritant.[14] An aqueous solution composed of 2.2% Olea Europaea (Olive) Fruit Extract was not irritating to rabbits when tested neat.[86] In further studies in rabbits, Olea Europaea (Olive) Leaf Extract was not a dermal irritant in primary or cumulative skin irritation tests when tested at up to 100%.[14] No irritation was observed with a face cream containing 0.0005% Olea Europaea (Olive) Fruit Extract in a human single-insult occlusive patch test (SIOPT) nor in a 4-d clinical use test.[87,88] No irritation was observed in human dermal irritation studies of formulations containing 0.047% (n = 52)[89] or 1% Olea Europaea (Olive) Leaf Extract (n = 20; 22),[90,91] or in a study with 100% Olea Europaea (Olive) Leaf Extract (n = 46).[14] A body scrub containing 0.025% Olea Europaea (Olive) Seed Powder (tested at 0.5% aq.) elicited a ± response in 1 out of 21 subjects in an SIOPT; no other reactions were observed.[92] No significant clinical changes or subjective discomfort were reported in 1-wk clinical use test of a bar soap containing 1% Olea Europaea (Olive) Seed Powder.[93]

An aqueous solution composed of 2.2% Olea Europaea (Olive) Fruit Extract was not sensitizing in a guinea maximization study when tested neat.[86] In another guinea pig sensitization study, Olea Europaea (Olive) Leaf Extract was negative for sensitization when tested at up to 100% for both induction and challenge phases.[14] In human repeated-insult patch tests (HRIPT), a product containing 0.0025% Olea Europaea (Olive) Fruit Extract and 0.035% Olea Europaea (Olive) Seed Powder (tested as a 0.5% w/v aqueous solution) produced no dermal sensitization in 100 subjects.[94] Dermal sensitization was also not observed in a maximization study of a lip balm containing 5% Olea Europaea (Olive) Leaf Extract (25 subjects), a product containing 20% Olea Europaea (Olive) Leaf Extract (54 subjects), or a product containing 0.3% Olea Europaea (Olive) Leaf Extract (109 subjects).[14,95,96] In an HRIPT with semi-occlusive patches, a product containing 25% Olea Europaea (Olive) Seed Powder was not a dermal sensitizer in 54 subjects.[97] A product containing 0.01% Olea Europaea (Olive) Fruit Extract and a product containing 10% Olea Europaea (Olive) Leaf Extract were not photosensitizers in studies of 27 subjects and 25 subjects, respectively.[98,99]

OCULAR IRRITATION STUDIES

In Vitro

Olea Europaea (Olive) Fruit Extract

In a neutral red release assay, an aqueous solution composed of 2.2% Olea Europaea (Olive) Fruit Extract was diluted in 0.9% sodium chloride to obtain the test concentrations of the solution of 5, 15, 25, 35, and 50% (i.e., test concentrations of the extract were 0.11, 0.33, 0.55, 0.77, and 1.1%, respectively).[86] The test concentrations of the solution were then analyzed through direct application on a monolayer of rabbit cornea fibroblasts. The half-maximal inhibitory concentration (IC_{50}) of the test material was higher than 50%. The precent mortality observed at the dilution of 50% was 10%. The cytotoxicity of the test material was thus negligible. No further details were provided.

Animal

Olea Europaea (Olive) Fruit Extract

The ocular irritation potential of an aqueous solution composed of 2.2% Olea Europaea (Olive) Fruit Extract was assessed in an acute eye irritation/corrosion test in rabbits in accordance with OECD TG 405.[86] A dose of 0.1 ml of the test material was applied neat. The test material was not irritating to rabbit eyes. No further details were provided.

CLINICAL STUDIES

Case Reports

Anaphylaxis was reported in at 21-yr-old woman with a history of allergic rhinitis and asthma following consumption of olives on 3 separate occasions.[100] Symptoms included oropharynx, itchy palms, cough, and dyspnea. No history of food allergy had been reported prior. Skin prick tests were positive to different dust mites and negative for pollens, including olive tree pollen. Prick-by-prick testing with raw olive fruit gave a positive result (25 mm x 20 mm wheal and general skin itching). Five control subjects were negative. Additional testing with a prick-by-prick test of olive oil results in a 6 mm^2 wheal and general itching. Total immunoglobulin E (IgE) was 2524 kU/l and specific IgE was negative for pollens and foods. Immunoblotting suggested an IgE-mediated food allergy to lipoproteins in olive fruit.

In another case report of anaphylaxis, a 19-yr-old woman with a history of childhood atopic dermatitis, persistent mild rhinoconjunctivitis, and intermittent asthma presented with oral itching, generalized urticaria, lip and eyelid edema, dyspnea, sweating, and dizziness with emetic episode 30 min following ingestion of an olive in brine.[101] The patient tolerated spices, garlic, olive oil, and there were no associated co-factors such as medication, alcohol, or exercise. Skin prick tests with an allergen battery were positive for dust mites, cat and dog dander, *Olea europaea* pollen, and grass pollen. Prick-by-prick tests with spices and garlic were negative, but were positive for olive fruit, both fresh and in brine (9 and 8 mm wheals, respectively). An open oral challenge with olive oil had negative results. IgE binding analysis detected reactive proteins (a thaumatin-like protein (Ole e 13) and a storage protein (Conarchin-like protein)) as the probable allergens.

Other Clinical Reports

Olea Europaea (Olive) Fruit Extract

A skin lotion containing olive fruit extract (concentration not reported) and tetramethoxyluteolin was given to 25 mastocytosis patients and an additional 8 patients with acute dermatitis or psoriasis.[102] The patients in the first group were requested to try the lotion on any body part twice per day for at least 2 wk, and were then surveyed regarding any skin symptoms associated with the use of the lotion. The second group were directed to apply the lotion on relevant affected areas twice per day for 1 mo. Eighteen patients in the first group responded to the survey, with none of the patients reporting irritation. No adverse effects to the lotion were reported in the second group of 8 patients.

Olea Europaea (Olive) Leaf Extract

In a study of the oxidative effects of olive leaf extract supplementation, groups of 15 young, healthy adult male and female subjects (total n = 45) were randomized into 3 groups.[103] Two groups received commercial olive leaf extract as a liquid (5 ml) or as a capsule. Concentrations of olive leaf extract in the commercial supplements was not reported. The third group served as a control and received a liquid placebo. In addition to being randomized, the study was a single-center and single-blind. The subjects ingested the test materials 3 times/d for 28 d. Urine samples were taken at baseline and follow-up time periods and measured for creatine, isoprostanes, and 8-hydroxy-2'-deoxyguannosine. All subjects completed the study, but only 36 were compliant with all protocols throughout the test period. No adverse effects were recorded. No significant effects of olive leaf extract on oxidative markers were observed when compared to controls.

In an efficacy study, 36 females with photoaging skin (including wrinkles, skin roughness, dryness, irregular pigmentation, telangiectasia, sallowness, and brown spots) were instructed to apply 0.6 g of a cream lotion containing olive leaf extract to their whole face twice daily for 2 mo.[104] Clinical evaluations were made at baseline, 1 and 2 mo after the start of application, and 1 mo after discontinuation of the cream. No other products were to be applied during the treatment period. No serious adverse events were reported during the study at follow-up visits. However, 16.7% of the subjects were reported to have mild and transient acneiform eruption after the cream treatment started.

SUMMARY

Most of the *Olea europaea* (olive)-derived ingredients detailed in this safety assessment are reported to function in cosmetics as skin-conditioning agents, according to the *Dictionary*. Olea Europaea (Olive) Husk Powder and Olea Europaea (Olive) Seed Powder are reported to only function as abrasives, and Olea Europaea (Olive) Flower Water and Olea Europaea (Olive) Fruit Juice only as antioxidants. Reported function as a skin bleaching agent (for Olea Europaea (Olive) Fruit Extract and Olea Europaea (Olive) Leaf Extract) is not considered a cosmetic function in the US and, therefore, is not addressed in this assessment.

Olea europaea L. is an evergreen tree or shrub native to the Mediterranean region and is one of the earliest domesticated fruit trees in the world, used for its oil, edible fruit, and medicinal properties since antiquity. Composition of constituents of *Olea europaea* (olive)-derived ingredients can vary annually, and is dependent on the cultivar, production area, climate, season and soil characteristics. Oleuropein is the main phenolic component of the unprocessed fruit and leaves of *Olea europaea* L.

According to 2023 VCRP survey data, Olea Europaea (Olive) Leaf Extract is reported to be used in 170 formulations, with a majority of uses in leave-on skin care preparations. Olea Europaea (Olive) Fruit Extract is reported to be used in 124 formulations, also with the majority of uses in leave-on skin care preparations. All other in-use ingredients are reported to be used at much lower numbers. The results of the concentration of use survey conducted by the Council in 2020 indicate Olea Europaea (Olive) Leaf Extract also has the highest concentration of use in a leave-on formulation; it is used at up to 2% in suntan preparations. The highest concentration of use reported for products resulting in rinse-off dermal exposure is 10% in Olea Europaea (Olive) Fruit Unsaponifiables in shaving cream. Fourteen ingredients in this safety assessment have no reported uses.

Different parts of the olive tree have been used for centuries for nutritional properties and protective health effects. Leaves and fruits, extracts, and constituents have been studied for antioxidant, antimicrobial, and anti-inflammatory benefits, as well as for treatments for diabetes, hypertension, and protective effects.

In mouse studies of olive stem bark extract, an aqueous hydrolyzed olive pulp (fruit) extract, and olive leaf extract, the LD_{50} was greater than 2000 mg/kg, the maximum dose tested for each ingredient. In rat studies, an aqueous hydrolyzed olive pulp (fruit) extract had an LD_{50} greater than 5000 mg/kg, and olive leaf extract had an LD_{50} greater than 2000 mg/kg.

No treatment-related mortalities were observed in rats that received olive fruit extract (up to 1381 mg/kg bw/d) or hydrolyzed olive pulp (fruit) extract (aqueous; up to 2000 mg/kg/d) via oral gavage for 90 d. The LOAEL was 1381 mg/kg bw/d and the NOAEL was 691 mg/kg bw/d in the olive fruit extract study; and the NOAEL for the hydrolyzed olive pulp (fruit) extract was 2000 mg/kg/d. In studies of a proprietary olive leaf extract in rats, dose-dependent hyaline droplet nephropathy was observed in males in the 1000 and 2000 mg/kg dose groups, but not in lower dose males or in any females in a 14-d study. No mortality, clinical signs of toxicity, or abnormalities in liver and kidneys were observed in a 28-d study with olive leaf extract (ethanol) at up to 400 mg/kg, but blood concentration of blood urea nitrogen was significantly increased in males in the 100 and 400 mg/kg dose groups when compared to controls. In a 42-d rat study with up to 0.9% olive leaf extract (aq.), livers and kidneys had fatty changes (liver), hepatocellular necrosis, and streaky hemorrhages (kidneys) in all test groups, but the effects were more prominent in the 0.7% and 0.9% dose groups. The NOAEL in a 90-d study was the maximum dose tested of 1000 mg/kg bw/d for a proprietary olive leaf extract.

In male rats treated at up to 450 mg/kg olive fruit extract (hydroalcoholic) for 48 d, a significant decrease in testicle weights (all treatment groups) and seminal vesicle weight (150 mg/kg dose group only) was observed, as were significant decreases in testosterone hormone levels, sperm counts, and sperm motility (all treatment groups for each end point). Hydrolyzed olive pulp (fruit) extract (aqueous; up to 2000 mg/kg/d) produced no treatment-related mortalities in F_0 mature rats or F_1 rat pups and produced no adverse effects in fertility or reproduction. The NOAEL for developmental toxicity in rats was greater than 2000 mg/kg/d when dams received the test material during gestation days 6 through 20.

Mutagenic activity was observed in a bacterial reverse mutation assay (tested up to 5000 µg/plate) and a chromosome aberration assay (tested up to 1000 µg/ml) of an aqueous hydrolyzed olive pulp (fruit) extract when tested with metabolic activation; however, this test material was not mutagenic in an in vivo micronucleus assay (tested up to 5000 mg/kg/d) in rats. Different olive leaf extracts were not considered genotoxic in a bacterial reverse mutation assay (tested up to 5000 µg/plate), a bacterial Vitrotox™ test (tested up to 5.0 mg/ml), an alkaline comet assay (tested up to 5.0 mg/ml) in human hepatic cells, and a mammalian chromosome aberration test (tested up to 1500 µg/ml) in V79 Chinese hamster lung cells. A proprietary olive leaf extract was not genotoxic in an in vivo micronucleus assay (tested up to 200 mg/ml) in mice.

Olive fruit extract inhibited cell proliferation without cytotoxicity and the restoration of apoptosis in human colon cancer cells. Olive leaf extract (ethanol extract) was not cytotoxic to PMNC.

Olea Europaea (Olive) Leaf Extract, tested at 100% in an in vitro primary skin irritation study, was predicted to be a non-irritant. An aqueous solution composed of 2.2% Olea Europaea (Olive) Fruit Extract was not irritating to rabbits when tested neat. In further rabbit studies, Olea Europaea (Olive) Leaf Extract was not a dermal irritant in primary or cumulative skin irritation tests when tested at up to 100%. No irritation was observed in a face cream containing 0.0005% Olea Europaea (Olive) Fruit Extract in a human SIOPT nor in a 4-d clinical use test. No irritation was observed in human dermal irritation studies of formulations containing 0.047% (n = 52) or 1% Olea Europaea (Olive) Leaf Extract (n = 20; 22), or in a study with 100% Olea Europaea (Olive) Leaf Extract (n = 46). A body scrub containing 0.025% Olea Europaea (Olive) Seed Powder (tested at 0.5% aq.) elicited a + response in 1 out of 21 subjects in an SIOPT; no other reactions were observed. No significant clinical changes or subjective discomfort were reported in a 1-wk clinical use test of a bar soap containing 1% Olea Europaea (Olive) Seed Powder. An aqueous solution composed of 2.2% Olea Europaea (Olive) Fruit Extract was not sensitizing in a guinea maximization study when tested neat. In another guinea pig sensitization study, Olea Europaea (Olive) Leaf Extract was negative for sensitization when tested at up to 100% for both induction and challenge phases. In

human repeated-insult patch tests (HRIPT), a product containing 0.0025% Olea Europaea (Olive) Fruit Extract and 0.035% Olea Europaea (Olive) Seed Powder (0.5% w/v aqueous solution) produced no dermal sensitization in 100 subjects. Dermal sensitization was also not observed in a maximization study of a lip balm containing 5% Olea Europaea (Olive) Leaf Extract (25 subjects), a product containing 20% Olea Europaea (Olive) Leaf Extract (54 subjects), or a product containing 0.3% Olea Europaea (Olive) Leaf Extract (109 subjects). A product containing 25% Olea Europaea (Olive) Seed Powder was not a dermal sensitizer in 54 subjects. A product containing 0.01% Olea Europaea (Olive) Fruit Extract and a product containing 10% Olea Europaea (Olive) Leaf Extract were not photosensitizers in studies of 27 subjects and 25 subjects, respectively.

An aqueous solution composed of 2.2% Olea Europaea (Olive) Fruit Extract was diluted in 0.9% sodium chloride to obtain test concentrations (of the solution) of up to 50% for use in an in vitro ocular neutral red release assay. The IC_{50} of the test material was higher than 50%. The aqueous solution composed of 2.2% Olea Europaea (Olive) Fruit Extract was also not irritating to rabbit eyes when tested neat.

Anaphylaxis has been reported in patients with an IgE-mediated food allergy to lipoproteins in olive fruit. Clinical studies of a skin lotion containing olive fruit extract, an oral supplement containing olive leaf extract, and a skin lotion containing olive leaf extract noted no adverse effects.

No relevant carcinogenicity studies were found in the published literature, and unpublished data were not submitted. No relevant toxicokinetic studies were found in the published literature; however, in general, toxicokinetics data are not expected to be found on botanical ingredients because each botanical ingredient is a complex mixture of constituents.

DISCUSSION

The Panel reviewed the safety of 23 ingredients derived from *Olea europaea* (olive), most of which are reported to function as skin conditioning agents in cosmetic products. The Panel concluded that the available data are sufficent for determining the safety of 16 ingredients, i.e., those derived from the fruit, husk, leaf, and seed, for use in cosmetic products. The Panel noted that the fruit and leaves are consumed as foods and health supplements; composition and other data on the husk and seed denote similarities to both the fruit and the leaf. This information, and the likelihood that these ingredients do not readily absorb, obviate the need for additional toxicological data.

The *Dictionary* indicates that skin bleaching agent is a reported function of extracts of the fruit and leaves of *Olea europaea*; however, no data have been discovered that support or disprove this potential effect. The Panel noted that skin lightening is considered a drug effect in the US, and should not oocur during the use of cosmetic products.

The Panel also expressed concern about heavy metals, pesticide residues, and other plant species that may be present in botanical ingredients. They stressed that the cosmetics industry should continue to minimize impurities in cosmetic formulations according to limits set by the US FDA and EPA.

The Panel discussed the issue of incidental inhalation exposure resulting from these ingredients (e.g., Olea Europaea (Olive) Leaf Extract is used at 0.018% in hair spray and Olea Europaea (Olive) Fruit Extract is used in face powders (no concentration reported)). Inhalation toxicity data were not available. However, the Panel noted that in aerosol products, the majority of droplets/particles would not be respirable to any appreciable amount. Furthermore, droplets/particles deposited in the nasopharyngeal or tracheobronchial regions of the respiratory tract present no toxicological concerns based on the chemical and biological properties of these ingredients. Coupled with the small actual exposure in the breathing zone and the low concentrations at which these ingredients are used (or expected to be used) in potentially inhaled products, the available information indicates that incidental inhalation would not be a significant route of exposure that might lead to local respiratory or systemic effects. A detailed discussion and summary of the Panel's approach to evaluating incidental inhalation exposures to ingredients in cosmetic products is available at https://www.cir-safety.org/cir-findings.

The Panel's respiratory exposure resource document (see link above) notes that airbrush technology presents a potential safety concern, and that no data are available for consumer habits and practices thereof. As a result of deficiencies in these critical data needs, the safety of cosmetic ingredients applied by airbrush delivery systems cannot be assessed by the Panel. Therefore, the Panel has found the data insufficient to support the safe use of cosmetic ingredients applied via an airbrush delivery system.

The Panel also concluded that the data are insufficient for determining safety of the remaining 7 *Olea europaea* (olive)-derived ingredients. For these ingredients, Panel felt that there may be differences in the methods of manufacturing, composition and impurities, and other data points, as compared to the ingredients that had sufficient data; thus, it was unclear if inferences from the fruit, leaf, husk, and seed could be applied to the bark, branch, bud, flower, sap, and wood ingredients. Accordingly, the additional data needed to determine the safety of these ingredients in cosmetics are:

- Method of manufacture for Olea Europaea (Olive) Bark Extract, Olea Europaea (Olive) Branch Extract, Olea Europaea (Olive) Bud Extract, Olea Europaea (Olive) Flower Extract, Olea Europaea (Olive) Sap Extract, and Olea Europaea (Olive) Wood Extract
- Composition and impurities data for Olea Europaea (Olive) Branch Extract and Olea Europaea (Olive) Flower Water

- 28-day dermal toxicity data for Olea Europaea (Olive) Bark Extract, Olea Europaea (Olive) Branch Extract, Olea Europaea (Olive) Bud Extract, Olea Europaea (Olive) Flower Extract, Olea Europaea (Olive) Sap Extract, and Olea Europaea (Olive) Wood Extract
    - If positive, additional data (e.g., DART and genotoxicity data) may be needed
- Dermal irritation and sensitization data for Olea Europaea (Olive) Bark Extract, Olea Europaea (Olive) Branch Extract, Olea Europaea (Olive) Bud Extract, Olea Europaea (Olive) Flower Extract, Olea Europaea (Olive) Sap Extract, and Olea Europaea (Olive) Wood Extract

CONCLUSION

The Expert Panel for Cosmetic Ingredient Safety concluded that the following 16 *Olea europaea* (olive)-derived ingredients are safe in cosmetics in the present practice of use and concentration described in this safety assessment:

Hydrolyzed Olive Fruit*
Hydrolyzed Olive Fruit Extract*
Hydrolyzed Olive Leaf Extract*
Olea Europaea (Olive) Fruit
Olea Europaea (Olive) Fruit Extract
Olea Europaea (Olive) Fruit Juice*
Olea Europaea (Olive) Fruit Juice Extract*
Olea Europaea (Olive) Fruit Unsaponifiables
Olea Europaea (Olive) Fruit Water*
Olea Europaea (Olive) Husk Powder*
Olea Europaea (Olive) Leaf*
Olea Europaea (Olive) Leaf Extract
Olea Europaea (Olive) Leaf Powder
Olea Europaea (Olive) Leaf Water
Olea Europaea (Olive) Seed*
Olea Europaea (Olive) Seed Powder

**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.*

Additionally, the Panel also concluded that the available data are insufficient to make a determination of safety for the following 7 *Olea europaea* (olive)-derived ingredients under the intended conditions of use in cosmetic formulations:

Olea Europaea (Olive) Bark Extract**
Olea Europaea (Olive) Branch Extract**
Olea Europaea (Olive) Bud Extract
Olea Europaea (Olive) Flower Extract**
Olea Europaea (Olive) Flower Water**
Olea Europaea (Olive) Sap Extract
Olea Europaea (Olive) Wood Extract**

***There are currently no uses reported for these ingredients.*

TABLES

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

Ingredient & CAS No.	Definition	Function(s)
Hydrolyzed Olive Fruit	Hydrolyzed Olive Fruit is the hydrolysate of Olea Europaea (Olive) Fruit derived by acid, enzyme, or other method of hydrolysis.	Antioxidant; light stabilizer; skin protectant; skin-conditioning agent – emollient
Hydrolyzed Olive Fruit Extract	Hydrolyzed Olive Fruit Extract is the hydrolysate of Olea Europaea (Olive) Fruit Extract derived by acid, enzyme, or other method of hydrolysis.	Antioxidant
Hydrolyzed Olive Leaf Extract	Hydrolyzed Olive Leaf Extract is the hydrolysate of Olea Europaea (Olive) Leaf Extract derived by acid, enzyme, or other method of hydrolysis.	Skin-conditioning agent – misc.
Olea Europaea (Olive) Bark Extract 84012-27-1 (generic)	Olea Europaea (Olive) Bark Extract is the extract of the bark of *Olea europaea.*	Skin-conditioning agent – misc.
Olea Europaea (Olive) Branch Extract 84012-27-1 (generic)	Olea Europaea (Olive) Branch Extract is the extract of the branches of *Olea europaea.*	Skin-conditioning agent – misc.
Olea Europaea (Olive) Bud Extract 84012.27-1 (generic)	Olea Europaea (Olive) Bud Extract is the extract of the buds of the *Olea europaea.*	Antioxidant; skin-conditioning agent - emollient
Olea Europaea (Olive) Flower Extract 84012-27-1 (generic)	Olea Europaea (Olive) Flower Extract is the extract of the flowers of *Olea europaea.*	Skin-conditioning agent – misc.
Olea Europaea (Olive) Flower Water 84012-27-1 (generic)	Olea Europaea (Olive) Flower Water is an aqueous solution of the steam distillate obtained from the flowers of *Olea europaea*.	Antioxidant
Olea Europaea (Olive) Fruit	Olea Europaea (Olive) Fruit is the fruit obtained from *Olea europaea*.	Abrasive; skin-conditioning agent – misc.
Olea Europaea (Olive) Fruit Extract 84012-27-1	Olea Europaea (Olive) Fruit Extract is the extract of the fruit of *Olea europaea.*	Skin bleaching agent; skin-conditioning agent – misc.
Olea Europaea (Olive) Fruit Juice	Olea Europaea (Olive) Fruit Juice is the juice expressed from the fruit of *Olea europaea*.	Antioxidant
Olea Europaea (Olive) Fruit Juice Extract	Olea Europaea (Olive) Fruit Juice Extract is the extract of Olea Europaea (Olive) Fruit Juice.	Skin-conditioning agent – humectant
Olea Europaea (Olive) Fruit Unsaponifiables	Olea Europaea (Olive) Fruit Unsaponifiables is the fraction of olive fruit remaining after fractional distillation.	Antioxidant; binder; emulsion stabilizer; hair conditioning agent; skin conditioning agent – emollient
Olea Europaea (Olive) Fruit Water	Olea Europaea (Olive) Fruit Water is an aqueous solution of the steam distillate obtained from the fruit of *Olea europaea*.	Skin-conditioning agent – misc.
Olea Europaea (Olive) Husk Powder	Olea Europaea (Olive) Husk Powder is the powder obtained from the dried, ground husks of *Olea europaea.*	Abrasive
Olea Europaea (Olive) Leaf	Olea Europaea (Olive) Leaf is the leaf of *Olea europaea*.	Skin-conditioning agent – misc.
Olea Europaea (Olive) Leaf Extract 84012-27-1 (generic); 8060-29-5 (generic)	Olea Europaea (Olive) Leaf Extract is the extract of leaves of *Olea europaea*.	Skin bleaching agent; skin-conditioning agent – misc.
Olea Europaea (Olive) Leaf Powder 84012-27-1 (generic)	Olea Europaea (Olive) Leaf Powder is the powder obtained from the dried, ground leaves of *Olea europaea*.	Abrasive; skin-conditioning agent – misc.
Olea Europaea (Olive) Leaf Water 84012-27-1 (generic)	Olea Europaea (Olive) Leaf Water is an aqueous solution of the steam distillates obtained from the leaves of *Olea europaea* (olive).	Skin-conditioning agent – misc.
Olea Europaea (Olive) Sap Extract	Olea Europaea (Olive) Sap Extract is the sap obtained from the stems of *Olea europaea*.	Skin-conditioning agent – misc.
Olea Europaea (Olive) Seed	Olea Europaea (Olive) Seed is the seed of *Olea europaea*.	Abrasive; skin-conditioning agent – misc.
Olea Europaea (Olive) Seed Powder 84012-27-1 (generic)	Olea Europaea (Olive) Seed Powder is the powder obtained from the dried, ground seeds of *Olea europaea*.	Abrasive
Olea Europaea (Olive) Wood Extract	Olea Europaea (Olive) Wood Extract is the extract of the wood of *Olea europaea*.	Skin-conditioning agent – misc.

Table 2. Generic plant part definitions as they apply to *Olea europaea* (olive)-derived ingredients.[1]

Plant Part	Definition
Bark	Tough protective covering of the woody stems and roots of trees and other woody perennial plants, consisting of cells produced by a cork cambium
Bud	A not yet developed shoot in the axil of a leaf, often covered with scales; a young flower that has not yet opened
Flower	The reproductive shoot in flowering plants, usually with sepals, petals, stamens and pistil(s)
Fruit	Mature, ripened ovary of flowering plant, containing seeds
Husk	A dry outer covering of a fruit or seed
Juice	The liquid contained in the vegetative parts or fruits
Leaf	Flattened photosynthetic organs, attached to stems
Sap	The fluid transported through the vascular system of a plant
Seed	A propagating sexual structure resulting from the fertilization of an ovule, formed by embryo, endosperm, or seed coat
Wood	Parts of woody stems or branches formed by lignification of cells

Table 3. Chemical properties.

Property	Value	Reference
Olea Europaea (Olive) Fruit Extract (prepared in butylene glycol and water)		
Physical Form	Colorless to light yellow liquid	[9]
Odor	Characteristic	[9]
Specific Gravity (@ 25 ºC)	1.02 (range 1.00 - 1.04)	[9]
Water Solubility	Soluble in any proportion in water	[9]
Olea Europaea (Olive) Leaf Extract (prepared in water)		
Physical Form	Colorless to light yellow liquid	[10]
Odor	Characteristic	[10]
Specific Gravity (@ 25 ºC)	1.00 (range 0.99 - 1.01)	[10]
Water Solubility	Soluble in any proportion of water	[10]

Table 4. Secondary constituents for powdered olive bark (mg/g).[22]

Solvents	Total polyphenols	Total flavonoids	Total polysaccharides	Total glycosaponins
n-hexane	28.49	38.09	33.06	1.06
chloroform	35.61	64.33	156.235	74.06
methanol	28.33	14.71	195.66	78.01
ethanol	26.15	11.13	268.75	76.93
water	27.04	8.11	30.25	72.02

Table 5. Comparison of constituent levels in ethyl acetate extract of different olive fruit cultivars from Italy and Algeria (mg/kg dw, except where noted). [31]

Cultivar	total polyphenol content*	total tannin content**	*p*-hydroxy-benzoic acid	vanillic acid	caffeic acid	syringic acid	*p*-coumaric acid	ferulic acid	sinapic acid	tyrosol	hydroxy-tyrosol	verbascoside	oleuropein	luteolin	chrysoeriol
Italian cultivars															
Coratina	290.21	52.92	NR	134.66	80.65	32.84	6.57	37.78	30.64	134.75	1927.57	319.78	126.92	221.74	11.68
Frantoio	223.81	63.95	309.36	203.46	142.17	81.65	35.74	25.22	25.95	200.84	2338.45	693.77	2562.63	585.64	135.57
Leccino	224.92	86.86	66.43	NR	129.32	63.24	21.95	31.75	24.22	194.13	1876.23	643.09	1074.28	2828.86	303.14
Maiatica	182.35	66.27	308.87	493.94	96.46	120.68	19.33	156.54	44.67	17.96	3683.44	718.68	1361.47	513.24	549.25
Ogliarola	226.89	57.51	116.42	37.53	83.42	39.12	28.74	31.36	31.85	115.74	2974.14	335.34	804.56	1362.51	158.74
Algerian cultivars															
Chemlal	272.83	81.28	NR	34.84	8.72	6.64	17.65	103.09	23.46	100.21	2024.63	21.37	109.86	201.70	21.73
Sigoise	147.13	20.08	3.22	200.93	29.64	13.64	66.37	63.38	26.34	34.34	245.23	52.72	216.70	109.54	36.37

NR=not reported
*mg of gallic acid equivalents/g extract
**mg of tannic acid equivalents/g extract

Table 6. Constituent levels in leaf extracts of different olive cultivars from Italy and Tunisia (g/kg dw).

Cultivar	quinic acid	hydroxytyrosol	luteolin 7-*O*-glucoside	2-methoxy oleuropein	oleuropein	luteolin	verbascoside	tyrosol	4-hydroxybenzoic acid	rutin	apigenin
Ethanolic extracts of Italian olive cultivars[38]											
Apollo	21.31	8.17*	39.78	10.51	24.28	2.66	0.16	NR	NR	NR	NR
Ascolanatenera	12.71	10.96*	32.75	7.80	22.06	0.15	0.18	NR	NR	NR	NR
Carolea	13.93	17.34*	35.05	12.71	28.30	0.10	0.13	NR	NR	NR	NR
Cellina di Nardo	11.25	57.75*	23.31	22.14	9.69	2.62	0.20	NR	NR	NR	NR
Cipressino	13.31	3.58*	29.13	9.42	25.52	0.21	0.22	NR	NR	NR	NR
Itrana	25.19	1.13*	31.56	8.42	30.46	1.54	0.11	NR	NR	NR	NR
Maurino	14.81	2.05*	27.88	4.08	18.53	3.02	0.10	NR	NR	NR	NR
Minerva	6.05	2.42*	15.95	3.32	17.38	1.06	0.18	NR	NR	NR	NR
Moraiolo	9.20	11.88*	20.12	5.56	14.61	1.41	0.14	NR	NR	NR	NR
Nociara	10.22	7.14*	35.13	3.92	9.89	0.18	0.10	NR	NR	NR	NR
Ogliarola	6.24	7.90*	8.69	8.82	7.49	0.21	0.14	NR	NR	NR	NR
Pendolino	12.55	1.69*	17.84	2.55	12.58	0.88	0.15	NR	NR	NR	NR
Ravece	13.02	3.72*	15.85	3.07	18.12	0.09	0.13	NR	NR	NR	NR
Sant Agostino	16.50	3.48*	21.57	5.28	23.55	0.16	0.11	NR	NR	NR	NR
Taggiasca	12.54	4.58*	18.14	4.14	21.74	0.95	0.12	NR	NR	NR	NR
Methanolic extracts of Tunisian olive cultivars[3]											
Chetoui	NR	0.0913	0.176	NR	0.428	NR	NR	0.141	0.0838	0.156	0.0343
Meski	NR	0.0896	0.116	NR	0.520	NR	NR	0.114	0.0663	0.210	0.0292
Jarbouii	NR	0.0893	0.217	NR	0.259	NR	NR	0.0862	0.0811	0.249	0.0433
Ouslati	NR	0.0757	0.113	NR	0.246	NR	NR	0.0835	0.0548	0.146	0.0217

NR = not reported
*Reported as hydroxytyrosol glucoside

Table 7. Frequency (2023) and concentration (2020/2023) of use according to likely duration and exposure and by product category[47-49]

	# of Uses	Max Conc of Use (%)	# of Uses	Max Conc of Use (%)	# of Uses	Max Conc of Use (%)
	Olea Europaea (Olive) Bud Extract		Olea Europaea (Olive) Fruit		Olea Europaea (Olive) Fruit Extract[d]	
Totals	**1**	**NR**	**14**	**0.6**	**124**	**0.0002-0.5**
summarized by likely duration and exposure*						
Duration of Use						
Leave-On	*1*	*NR*	*9*	*0.6*	*96*	*0.00025-0.45*
Rinse-Off	*NR*	*NR*	*5*	*NR*	*28*	*0.0002-0.5*
Diluted for (Bath) Use	*NR*	*NR*	*NR*	*NR*	*NR*	*NR*
Exposure Type*						
Eye Area	NR	NR	NR	NR	3	NR
Incidental Ingestion	NR	NR	NR	NR	16	0.24
Incidental Inhalation-Spray	1[a]	NR	5[b]	NR	2; 30[a]; 29[b]	0.0008
Incidental Inhalation-Powder	NR	NR	5[b]; 4[c]	NR	3; 29[b]	0.23-0.45[c]
Dermal Contact	1	NR	12	0.6	95	0.00025-0.5
Deodorant (underarm)	NR	NR	NR	NR	NR	0.0008-0.005
Hair - Non-Coloring	NR	NR	2	NR	13	0.0002-0.069
Hair-Coloring	NR	NR	NR	NR	NR	NR
Nail	NR	NR	NR	NR	NR	NR
Mucous Membrane	NR	NR	3	NR	223	0.00025-0.24
Baby Products	NR	NR	NR	NR	NR	NR
as reported by product category						
Baby Products						
Baby Shampoos						
Baby Lotions/Oils/Powders/Creams						
Other Baby Products						
Eye Makeup Preparations						
Eye Lotion					1	NR
Other Eye Makeup Preparations					2	NR
Fragrance Preparations						
Cologne and Toilet Water					1	NR
Other Fragrance Preparation					1	NR
Hair Preparations (non-coloring)						
Hair Conditioner					6	0.0002
Hair Spray (aerosol fixatives)						
Rinses (non-coloring)						
Shampoos (non-coloring)			1	NR	4	0.0098
Tonics, Dressings, and Other Hair Grooming Aids			1	NR	NR	0.069 (not spray)
Other Hair Preparations					3	NR
Makeup Preparations						
Face Powders					3	NR
Foundations						
Lipstick					16	0.24
Makeup Bases					1	NR
Other Makeup Preparations					1	NR
Manicuring Preparations (Nail)						
Other Manicuring Preparations						
Personal Cleanliness Products						
Bath Soaps and Detergents			3	NR	2	0.00025-0.11
Deodorants (underarm)					NR	0.005 (not spray) 0.0008 (aerosol)
Feminine Deodorants					1	NR
Other Personal Cleanliness Products					4	NR
Shaving Preparations						
Beard Softeners						
Shaving Cream					NR	0.5
Skin Care Preparations						
Cleansing			1	NR	11	0.01
Depilatories						
Face and Neck (exc shave)			2	NR	14	0.4-0.45 (not spray)
Body and Hand (exc shave)			2	NR	14	0.23 (not spray)
Moisturizing			4	0.6 (not spray)	28	0.00025 (not spray)
Night	1	NR			2	0.00025 (not spray)
Paste Masks (mud packs)					1	NR
Other Skin Care Preparations					8	0.01
Suntan Preparations						
Suntan Gels, Creams, and Liquids						

Table 7. Frequency (2023) and concentration (2020/2023) of use according to likely duration and exposure and by product category[47-49]

	# of Uses	Max Conc of Use (%)	# of Uses	Max Conc of Use (%)	# of Uses	Max Conc of Use (%)
	Olea Europaea (Olive) Fruit Unsaponifiables		Olea Europaea (Olive) Leaf Extract		Olea Europaea (Olive) Leaf Powder	
Totals	**10**	**10**	**170**	**0.0002-2**	**1**	**0.1**
summarized by likely duration and exposure*						
Duration of Use						
Leave-On	*10*	*NR*	*125*	*0.0002-2*	*1*	*0.1*
Rinse-Off	*NR*	*10*	*45*	*0.0002-0.3*	*NR*	*NR*
Diluted for (Bath) Use	*NR*	*NR*	*NR*	*NR*	*NR*	*NR*
Exposure Type*						
Eye Area	NR	NR	8	NR	NR	NR
Incidental Ingestion	NR	NR	1	0.002	NR	NR
Incidental Inhalation-Spray	2[a]; 7[b]	NR	42[a]; 53[b]	0.018	1[a]	NR
Incidental Inhalation-Powder	7[b]	NR	1; 53[b]	0.0014-0.4[c]	NR	0.1[c]
Dermal Contact	10	10	157	0.0002-2	1	0.1
Deodorant (underarm)	NR	NR	NR	0.095 (not spray) 0.0002 (spray)	NR	NR
Hair - Non-Coloring	NR	NR	11	0.0005-0.018	NR	NR
Hair-Coloring	NR	NR	NR	NR	NR	NR
Nail	NR	NR	1	NR	NR	NR
Mucous Membrane	NR	NR	28	0.0003-0.002	NR	NR
Baby Products	NR	NR	2	0.002-0.013	NR	NR
as reported by product category						
Baby Products						
Baby Shampoos			2	0.0065		
Baby Lotions/Oils/Powders/Creams			NR	0.013		
Other Baby Products			NR	0.002		
Eye Makeup Preparations						
Eye Lotion			3	NR		
Other Eye Makeup Preparations			5	NR		
Fragrance Preparations						
Cologne and Toilet Water						
Other Fragrance Preparation						
Hair Preparations (non-coloring)						
Hair Conditioner			2	0.003-0.018		
Hair Spray (aerosol fixatives)			NR	0.018 (pump spray)		
Rinses (non-coloring)			NR	0.0005		
Shampoos (non-coloring)			3	0.001-0.018		
Tonics, Dressings, and Other Hair Grooming Aids						
Other Hair Preparations			4	NR		
Makeup Preparations						
Face Powders			1	NR		
Foundations			NR	0.1		
Lipstick			1	0.002		
Makeup Bases						
Other Makeup Preparations			1	NR		
Manicuring Preparations (Nail)						
Other Manicuring Preparations			1	NR		
Personal Cleanliness Products						
Bath Soaps and Detergents			24	0.0003		
Deodorants (underarm)			NR	0.095 (not spray) 0.0002 (aerosol)		
Feminine Deodorants						
Other Personal Cleanliness Products			3	NR		
Shaving Preparations						
Beard Softeners			1	NR		
Shaving Cream	NR	10				
Skin Care Preparations						
Cleansing			7	0.0002-0.3		
Depilatories			1	NR		
Face and Neck (exc shave)	6	NR	30	0.0014-0.4 (not spray)		
Body and Hand (exc shave)	1	NR	23	NR	NR	0.1 (not spray)
Moisturizing	2	NR	41	0.0065 (not spray)	1	NR
Night			1	0.4 (not spray)		
Paste Masks (mud packs)			3	NR		
Other Skin Care Preparations	1	NR	13	0.002		
Suntan Preparations						
Suntan Gels, Creams, and Liquids			NR	2 (not spray)		

Table 7. Frequency (2023) and concentration (2020/2023) of use according to likely duration and exposure and by product category[47-49]

	# of Uses	Max Conc of Use (%)	# of Uses	Max Conc of Use (%)	# of Uses	Max Conc of Use (%)
	Olea Europaea (Olive) Leaf Water		Olea Europaea (Olive) Sap Extract		Olea Europaea (Olive) Seed Powder	
Totals	**1**	**NR**	**NR**	**0.005**	**6**	**NR**
summarized by likely duration and exposure*						
Duration of Use						
Leave-On	*1*	*NR*	*NR*	*0.005*	*2*	*NR*
Rinse-Off	*NR*	*NR*	*NR*	*0.005*	*4*	*NR*
Diluted for (Bath) Use	*NR*	*NR*	*NR*	*NR*	*NR*	*NR*
Exposure Type*						
Eye Area	NR	NR	NR	NR	NR	NR
Incidental Ingestion	NR	NR	NR	NR	NR	NR
Incidental Inhalation-Spray	1[b]	NR	NR	NR	1[b]	NR
Incidental Inhalation-Powder	1[b]	NR	NR	NR	1[b]	NR
Dermal Contact	1	NR	NR	NR	6	NR
Deodorant (underarm)	NR	NR	NR	NR	NR	NR
Hair - Non-Coloring	NR	NR	NR	0.005	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
as reported by product category						
Baby Products						
Baby Shampoos						
Baby Lotions/Oils/Powders/Creams						
Other Baby Products						
Eye Makeup Preparations						
Eye Lotion						
Other Eye Makeup Preparations						
Fragrance Preparations						
Cologne and Toilet Water						
Other Fragrance Preparation						
Hair Preparations (non-coloring)						
Hair Conditioner			NR	0.005		
Hair Spray (aerosol fixatives)						
Rinses (non-coloring)						
Shampoos (non-coloring)			NR	0.005		
Tonics, Dressings, and Other Hair Grooming Aids						
Other Hair Preparations			NR	0.005		
Makeup Preparations						
Face Powders						
Foundations						
Lipstick						
Makeup Bases						
Other Makeup Preparations						
Manicuring Preparations (Nail)						
Other Manicuring Preparations						
Personal Cleanliness Products						
Bath Soaps and Detergents						
Deodorants (underarm)						
Feminine Deodorants						
Other Personal Cleanliness Products						
Shaving Preparations						
Beard Softeners						
Shaving Cream						
Skin Care Preparations						
Cleansing					4	NR
Depilatories						
Face and Neck (exc shave)	1	NR				
Body and Hand (exc shave)					1	NR
Moisturizing						
Night						
Paste Masks (mud packs)						
Other Skin Care Preparations					1	NR
Suntan Preparations						
Suntan Gels, Creams, and Liquids						

NR – not reported

*Likely duration and exposure is derived based on product category (see Use Categorization https://www.cir-safety.org/cir-findings)

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

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

[b] Not specified whether a spray or a powder, but it is possible the use can be as a spray or a powder, therefore the information is captured in both categories.

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

[d] Includes 2 uses described as Olive Extract in the VCRP.

Table 8. Ingredients not reported to be in use, according to VCRP and Council data.[47-49]

Hydrolyzed Olive Fruit	Olea Europaea (Olive) Fruit Juice
Hydrolyzed Olive Fruit Extract	Olea Europaea (Olive) Fruit Juice Extract
Hydrolyzed Olive Leaf Extract	Olea Europaea (Olive) Fruit Water
Olea Europaea (Olive) Bark Extract	Olea Europaea (Olive) Husk Powder
Olea Europaea (Olive) Branch Extract	Olea Europaea (Olive) Leaf
Olea Europaea (Olive) Flower Extract	Olea Europaea (Olive) Seed
Olea Europaea (Olive) Flower Water	Olea Europaea (Olive) Wood Extract

Table 9. **Acute toxicity studies.**

Test Article	Animals	No./Group	Vehicle	Concentration/Dose/Protocol	LD_{50}/Results	Reference
			ORAL			
olive stem bark extract; tested as a crude 80% methanol extract and as solvent fractions (80% methanol followed by fractionating with butanol, water, or chloroform)	Female Swiss albino mice	5	distilled water	Single 2000 mg/kg oral dose (total volume 10 ml/kg bw) in accordance with OECD TG 425; observations made for 14 d	> 2000 mg/kg for the 80% methanol extract and the solvent fractions; no gross physical or behavioral changes or mortality observed	[75]
hydrolyzed olive pulp (fruit) extract (aqueous)	Male and female CD-1 mice	5 per sex	deionized water	Single limit dose of 2000 mg/kg via gavage followed by a 14-d recovery period	> 2000 mg/kg; no mortalities or morbidities observed and no abnormal clinical signs or gross morphologic changes were noted	[79]
hydrolyzed olive pulp (fruit) extract (aqueous)	Male and female Crl: CD(SD)IGS BR VAF/Plus rats	5 per sex	0.5% methylcellulose	0, 1000, 1500, 2000, or 5000 mg/kg via gavage	> 5000 mg/kg; no mortalities or morbidities observed and no abnormal clinical signs or gross changes were observed at necropsy	[79]
olive leaf extract; tested as a crude 80% methanol extract and as solvent fractions (80% methanol followed by fractionating with butanol, water, or chloroform)	Female Swiss albino mice	5	distilled water	Single 2000 mg/kg oral dose (total volume 10 ml/kg bw) in accordance with OECD TG 425; observations made for 14 d	> 2000 mg/kg for the 80% methanol extract and the solvent fractions; no gross physical or behavioral changes or mortality observed	[57]
Olea Europaea (Olive) Leaf Extract (ethanol extract)	mice (strain not reported)	10/sex	not reported	Acute toxicity test, no further details provided	> 2000 mg/kg; no further details provided	[14]
olive leaf extract (ethanolic)	Wistar rats	3 per sex	as supplied	Single 2000 mg/kg dose via gavage; control group received 10 ml/kg ethanol solution (51%); observations made for 14 d; blood collected at observation end for hematological and biochemical study; liver and kidneys examined microscopically	> 2000 mg/kg; no mortality, clinical signs of toxicity, or significant changes to body weight gain observed in treated rats; significant differences in hematological parameters, including red blood cells, hemoglobin, mean corpuscular volume, mean cell corpuscular hemoglobin concentration, and platelets (details not provided); blood concentration of creatinine significantly decreased ($p < 0.05$) in treated females as compared to the control group, while cholesterol was significantly decrease in treated males; authors determined hematological and biochemical parameters with significant differences may be due to experimental variations and were not treatment-related; no abnormalities were observed in the liver and kidneys	[80]

Table 10. Repeated dose toxicity studies.

Test Article	Animals/Group	Study Duration	Vehicle	Dose/Concentration/Protocol	Results	Reference
ORAL						
olive fruit extract containing 35% hydroxytyrosol	Groups of 10 male and 10 female Wistar rats	90 d	Reverse osmosis water	0, 345, 691, or 1381 mg/kg bw/d via gavage; an additional 2 recovery groups included a vehicle control and a high dose group that were followed for 28 d after the completion of the 90-d treatment to assess recovery; study performed in accordance with OECD TG 408; animals observed twice daily for mortality and clinical signs; body weight and feed consumption measured weekly; ophthalmological examination performed prior to treatment and at treatment and recovery end; blood samples collected during weeks 4, 8, 13 and 15 (recovery) from the control and high dose groups; urinalysis samples collected from all rats at the end of the main study and recovery study; vaginal smears and sperm collection were made; gross pathological exams and absolute organ weights determinations in all animals; histopathological exams performed in control and high-dose groups	LOAEL = 1381 mg/kg bw/d and the NOAEL = 691 mg/kg bw/d; no mortality or morbidity were observed during the study period; no treatment-related clinical signs observed in the low dose groups, while the mid- and high-dose groups had mild to moderate intermittent salivation – observation was considered non-adverse; reduction in terminal body weight and statistically significant reduction in body weight gain observed at week 13 in high-dose males; statistically significant increase in relative weights of the liver, heart, and kidneys observed in high-dose males and females	[81]
hydrolyzed olive pulp (fruit) extract (aqueous)	Groups of 20 male and 20 female Crl: CD(SD)IGS BR VAF/Plus rats	90 d	0.5% methylcellulose	0, 1000, 1500, or 2000 mg/kg/d via gavage; physical and ophthalmic examinations conducted before and near the end of study; clinical signs were recorded daily, body weights and feed consumption were recorded weekly, and hematology and serum chemistry determinations were made at necropsy	NOAEL = 2000 mg/kg/d; small decreases in body weight gains observed in 2000 mg/kg/d males and in all groups of females; feed consumption comparable to controls; no adverse clinical, hematologic, biochemical, organ weight or gross necropsy effects; focal, minimal, or mild hyperplasia of the mucosal squamous epithelium of the limiting ridge of the forestomach occurred in some 2000 mg/kg rats, but this was attributed to local irritation from gavage procedures	[79]
olive leaf extract; proprietary product with a standardized olive polyphenol content of 40%	Male and female CRL: (WI)BR Wistar SPF rats; no further details provided	14 d	1% Tween 80 prepared in distilled water	0, 360, 600, 1000, or 2000 mg/kg bw/d oral dose study in accordance with OECD TG 407; no further details provided	Male rats in the 1000 and 2000 mg/kg bw/d groups had hyaline droplet nephropathy in a dose-dependent manner; this effect was not observed in 300 or 600 mg/kg dose group males or in females at any dose level; no other treatment-related significant findings noted; no further details provided	[82]
olive leaf extract (ethanol)	Groups of 5 male and 5 female Wistar rats	28 d	as supplied	100, 200, or 400 mg/kg oral dose; negative control group received 10 ml/kg ethanol solution (51%); body weight gain measured at the end of dosing, blood collected and hematological parameters measured; rats killed and liver and kidneys examined microscopically	No mortality or clinical signs of toxicity observed; body weight gains normal in all dose groups; hematological parameters in treated rats comparable to the controls; blood urea nitrogen significantly increased ($p < 0.05$) in males in the 100 and 400 mg/kg dose groups when compared to the controls, but no other biochemical parameters exhibited any differences; no abnormalities found in the liver and kidneys	[80]

Table 10. Repeated dose toxicity studies.

Test Article	Animals/Group	Study Duration	Vehicle	Dose/Concentration/Protocol	Results	Reference
olive leaf extract (aq.)	Groups of 6 male Wistar albino rats	42 d	Dietary feed	0, 0.2%, 0.4%, 0.7%, or 0.9%; rats observed daily for clinical signs; hematological and biochemical parameters, including concentration of alkaline phosphatase (ALP), lactate dehydrogenase (LDH), total bilirubin, cholesterol, glucose, and triglycerides measured at the end of dosing; rats were killed and histological examination performed on livers, kidneys, and spleens	No clinical signs of toxicity observed; when compared to control group, a significant increase ($p < 0.001$) in serum ALP observed in all treated groups; a significant increase of total bilirubin observed in the 0.4%, 0.7%, and 0.9% dose groups; a significant decrease in serum triglycerides, glucose, and cholesterol observed in all test groups when compared to the control group; a significant decrease ($p < 0.05$) in values of red blood cell counts, hemoglobin, and packed cell volume observed in the 0.9% dose group; a significant decrease ($p < 0.05$) in hemoglobin and packed cell volume observed in the 0.2% dose group, and mean corpuscular volume was significantly higher in the 0.4%, 0.7%, and the 0.9% dose groups, when compared to the control group; a marked reduction in white blood cells in all treated groups compared to the control group; no pathological changes in the spleen observed in the control or the treated groups; livers in the 0.7% and 0.9% dose groups had fatty changes and hepatocellular necrosis; these changes were observed in a lesser degree in the 0.2% and 0.4% dose groups; kidneys in treated groups had streaky hemorrhages and congestion in the cortical region, with more severe hemorrhage in the two higher dose groups	[4]
olive leaf extract; proprietary product with a standardized olive polyphenol content of 40%	Male and female CRL: (WI)BR Wistar SPF rats; 10 per sex in main group and 5 per sex in satellite groups	90 d	1% Tween 80	0, 360, 600, or 1000 mg/kg bw/d at a dose volume of 10 ml/kg via gavage; toxicity study performed in accordance with OECD TG 408; animals observed twice daily for mortality; clinical signs observed once daily; body weight measured prior to treatment, twice weekly during weeks 1-4, once weekly during weeks 5-13, and immediately after rats were killed; ophthalmological examination performed prior to treatment in all animals and in control and high-dose animals at the end of treatment; blood samples collected at study end; gross pathological exams and absolute organ weights determinations in all animals; histopathological exams performed in control and high-dose groups; 28-d satellite study performed to determine whether the findings of the above 14-d study were repeatable	NOAEL = 1000 mg/kg bw/d in both sexes; 1 female in the 1000 mg/kg bw/d group died on day 2 and 1 male in the 1000 mg/kg bw/d group died on day 60 due to treatment procedure; no toxicologically relevant treatment-related clinical signs or effects on body weight or feed consumption observed compared to controls; no ophthalmological alterations observed; no toxicologically-relevant changes in hematology, blood coagulation, or clinical chemistry parameters observed; no test article-induced gross pathological lesions or organ weight difference observed in any organs or tissues in any dose groups compared to controls; histopathological exams did not reveal any treatment-related findings that were considered toxicologically significant; satellite study for nephropathy was negative	[82]

Table 11. DART studies.

Test Article	Animals/Group	Vehicle	Dose/Concentration	Procedure	Results	Reference
			ORAL			
olive fruit extract (hydro-alcoholic)	groups of 8 male Sprague-Dawley rats	saline	0, 50, 150, or 450 mg/kg	Test material administered via gavage for 48 d; body weight measured and blood samples taken prior to initial dosing and 24 h after final dosing; rats killed at treatment end and weights of left prostate, left testis, epididymis, and seminal vesicle taken; sperm count and sperm motility measured	A significant decrease ($p = 0.03$) observed in weights of the left testicle in all treatment groups and in weights of the seminal vesicle in the 150 mg/kg dose group; significant decreases in testosterone hormone levels ($p \leq 0.04$), sperm counts ($p \leq 0.001$), and sperm motility ($p \leq 0.04$) in all treatment groups; no significant effects observed in body, prostate, or epididymis weights or in estradiol hormone levels	[83]
hydrolyzed olive pulp (fruit) extract (aqueous)	groups of male and female Crl: CD(SD)IGS BR VAF/Plus rats	0.5% methylcellulose	0, 500, 1000, 1500, or 2000 mg/kg	Dosage-range reproduction study; rats received test material for 14 d before cohabitation and up until the day before necropsy (49 total doses for males; for females, after day 22 post-partum); clinical signs, body weights of males and females, feed consumption, estrous cycling, female maternal behavior, litter sizes, pup viability, pup body weights, and necropsy observations were records; pups from the F_1 generation weaned 21-d post-partum; 2 pups/sex/litter (80 rats/sex total) selected for a week of daily gavage treatments and recordings of clinical signs, body weights, and viability before being necropsied on post-partum day 28; remaining pups subjected to gross necropsy on post-partum day 21	No treatment-related mortality observed in F_0 males and females; only adverse clinical sign for F_0 rats was dose-dependent excess salivation; absolute and relative feed intake and feed consumption values comparable between groups; in treated F_0 males, non-dose-dependent increased body weight gains; all mating and fertility parameters, terminal body weights, and paired epididymal and testicular weights comparable among the groups; in treated F_0 females, body weight gains were increased during the pre-cohabitation period, were comparable during gestation, and were decreased in the 1500 and 2000 mg/kg/d dose groups compared to controls; no adverse effects in treated groups for number of estrous stages, in mating, fertility, gestation, delivery or litter parameters, or in parturition, lactation, or necropsy parameters; slight reductions in pup weight/litter on lactation days 14 and 21 were not statistically significant; no treatment-related deaths, clinical signs, or gross necropsy findings were observed in the F_1 generation pups; pups (2/sex/litter) treated for 7 d after weaning with all treatment levels had comparable body weights on post-partum day 28	[79]
hydrolyzed olive pulp (fruit) extract (aqueous)	groups of 25 mated female Crl: CD(SD)IGS BR VAF/Plus rats	0.5% methylcellulose	0, 500, 1000, 1500, or 2000 mg/kg	Developmental toxicity study; dams received test material on gestation days 6 – 20, and observed daily for viability and clinical signs, resorptions, and premature delivery; body weights recorded on gestation day 0 through necropsy; feed consumption values recorded on gestation days 0, 6, 9, 12, 15, 18, and 21	NOAEL > 2000 mg/kg/d; no mortalities observed during treatment period; one 2000 mg/kg/d dam killed due to premature labor, but no abnormalities observed with dam or litter; no adverse clinical or necropsy findings; no differences in maternal body weight, body weight gains, gravid uterine weights, corrected maternal body weights or body weight gains, or absolute or relative feed consumption in any dose group; litter parameters unaffected by test material; significantly increased mean number of corpora lutea in the high dose group within historical control ranges; all gross external, soft tissue, and skeletal fetal alternations comparable in type, incidence, and distribution to controls	[11,79]

Table 12. Genotoxicity studies.

Test Article	Concentration/Dose	Vehicle	Test System	Procedure	Results	Reference
IN VITRO						
hydrolyzed olive pulp (fruit) extract (aqueous)	5 - 5000 µg/plate	0.5% carboxymethylcellulose solution or dimethyl sulfoxide	*Salmonella typhimurium* TA97a, TA98, TA100, TA1335 or *Escherichia coli* WP2 uvrA	Bacterial reverse mutation assay, with and without metabolic activation	Mutagenic activity detected in strains TA98 and TA100 at 100 and 2500 µg/plate with metabolic activation; however, inconsistencies between regular and repeat trials, antibacterial properties of the test material, and observation of positive findings in only 2 concentrations (with precipitates and toxicity also present) complicated interpretation of findings	[79]
hydrolyzed olive pulp (fruit) extract (aqueous)	10-1000 µg/ml	dimethyl sulfoxide	Chinese hamster ovary cells	Chromosome aberration assay, with and without metabolic activation	A significant increase in the percentage of aberrant cells observed at 1000 µg/ml, with activation	[79]
olive leaf extract; proprietary product with a standardized olive polyphenol content of 40%	51.2, 128, 320, 800, 2000, and 5000 µg/plate	Ultrapure water	*S. typhimurium* TA98, TA100, TA1335, TA1537 or *E. coli* WP2 uvrA	Bacterial reverse mutation assay in accordance with OECD TG 471, with and without S9 metabolic activation	Not genotoxic; no substantial increases in revertant colony numbers observed in any of the strains, with or without metabolic activation, at any concentration level; sporadic increases in revertant colony numbers compared to vehicle control observed, however no dose-related increase beyond generally acknowledged border of biological relevance observed and mutation rates were well below threshold of being considered positive	[82]
4 different olive leaf extracts from different regions of Tunisia	Up to 5000 µg/ml	Aqueous, no further details	2 *S. typhimurium* TA 104 constructs	Bacterial Vitotox™ test, with and without S9 metabolic activation	Negative in 3 extracts, with or without metabolic activation; 4th extract had borderline genotoxicity with metabolic activation; antigenotoxic properties were not observed	[84]
4 different olive leaf extracts from different regions of Tunisia	Up to 5000 µg/ml	Aqueous, no further details	Human C3A hepatic cells	Alkaline comet assay; cells were incubated with test materials for 24 h without metabolic activation and lysed in alkaline solution before analysis for DNA damage	Not genotoxic in 3 extracts; an increase in DNA damage was observed in the 4th extract that had borderline genotoxicity in the bacterial study described above	[84]
olive leaf extract; proprietary product with a standardized olive polyphenol content of 40%	3 h exposure Without S9: 250, 500, 750, 1000, or 1250 µg/ml With S9: 250, 500, 750, or 1000 µg/ml 20 h exposure Without S9: 62.5, 125, 250, or 500 µg/ml With S9: 500, 750, 1000, 1250, or 1500 µg/ml	Dulbecco's Modified Eagle medium	V79 male Chinese hamster lung cells	Mammalian chromosome aberration test in accordance with OECD TG 473, with and without S9 metabolic activation; positive and negative controls used	Not clastogenic; test material did not induce an increase in the number of cells with aberrations or rates of polyploidy or endoreduplicated metaphases at any concentration during either period of exposure, with or without metabolic activation; no statistically significant differences between treatment and solvent control groups, and no dose-response relationships were observed; controls yielded expected results	[82]

Table 12. Genotoxicity studies.

Test Article	Concentration/Dose	Vehicle	Test System	Procedure	Results	Reference
IN VIVO						
hydrolyzed olive pulp (fruit) extract (aqueous)	0, 1000, 1500, 2000, or 5000 mg/kg/d	0.5% methylcellulose	groups of 5-7 male and 5-7 female Crl: CD(SD) IGS BR VAF/Plus rats	Micronucleus assay; rats given single or 28 consecutive daily doses (1000-2000 mg/kg/d) or 29 consecutive daily doses (5000 mg/kg/d); via gavage	Not mutagenic; numbers of micronucleated polychromatic erythrocytes not significantly increased in any group treated with test article when compared to negative controls	[79]
Olive leaf extract; proprietary product with a standardized olive polyphenol content of 40%	50, 100, or 200 mg/ml in dose volume of 10 ml/kg bw	sterile water	Groups of male SPF Crl: NMRI BR mice; negative control and high dose group had 10 mice each, remaining groups had 5 mice each	Micronucleus assay in accordance with OECD TG 474; mice received single dose via gavage; positive control (cyclophosphamide), low-, and mid-dose group mice were killed at 24 h post treatment, 5 mice each in the positive control and high-dose were killed at 24 h or 48 h	Not genotoxic; no mortality, clinical signs of toxicity, or adverse reactions were observed in the controls or the 500 or 1000 mg/kg bw dose groups; a slight decrease in activity and piloerection was observed in 4 out of 10 mice treated with 2000 mg/kg; no significant differences observed in frequency of micronucleated polychromatic erythrocytes between the 3 dose groups compared to negative control; in the 2000 mg/kg dose group, the number of polychromatic erythrocytes was slightly decreased compared to negative control at 48 h sampling time; positive control yielded expected results	[82]

Table 13. Dermal irritation and sensitization studies.

Test Article	Vehicle	Concentration/Dose	Test Population	Procedure	Results	Reference
			IRRITATION			
			IN VITRO			
Olea Europaea (Olive) Leaf Extract	none	100%	not reported	OECD TG 439 primary skin irritation method; no further details provided	Not irritating	[14]
			ANIMAL			
Aqueous solution composed of 2.2% Olea Europaea (Olive) Fruit Extract	not reported	Neat; 0.5 ml dose	rabbits; no further details provided	Acute dermal irritation/corrosion test in accordance with OECD TG 404; no further details provided	Unclassified among the chemicals irritating to skin; no further details provided	[86]
Olea Europaea (Olive) Leaf Extract	not reported	10% and 100%	3 rabbits; no further details provided	Primary skin irritation test; no further details provided	No irritation; no further details provided	[14]
Olea Europaea (Olive) Leaf Extract	not reported	12.5%, 25%, 50%, 100%	3 rabbits; no further details provided	Cumulative skin irritation test; no further details provided	No irritation; no further details provided	[14]
			HUMAN			
Face cream containing 0.0005% Olea Europaea (Olive) Fruit Extract	none	As supplied	19 subjects	SIOPT	No irritation; primary irritation index = 0.0	[88]
Face cream containing 0.0005% Olea Europaea (Olive) Fruit Extract	none	As supplied	14 subjects	4-d clinical use test; test material applied twice daily to face	No significant clinical changes; no reports subjective discomfort	[87]
Moisturizer lotion containing 0.047% Olea Europaea (Olive) Leaf Extract	none	As supplied	52 subjects; at least 50% considered to have sensitive skin	4-wk clinical use test; monadic design; subjects instructed to use test material twice daily; dermatological exams conducted at baseline, wk 2 and wk 4	Test material did not elicit any significant objective or subjective irritation; test material did not elicit significant dryness	[89]
Liquid lip color containing 1% Olea Europaea (Olive) Leaf Extract	none	As supplied	20 subjects	SIOPT	No irritation; primary irritation index = 0.0	[90]
Lip product containing 1% Olea Europaea (Olive) Leaf Extract	none	As supplied	22 subjects	5-d clinical use test; test material applied twice daily to upper and lower lips	No significant clinical changes; no reported subjective discomfort	[91]
Olea Europaea (Olive) Leaf Extract	none	100%	46 subjects	Irritation study; occlusive patch; no further details provided	No irritation; no further details provided	[14]
Body scrub containing 0.025% Olea Europaea (Olive) Seed Powder	none	aqueous 0.5%	21 subjects	SIOPT; 24-h	One subject had a $\pm$ response, no other reactions observed; primary irritation index = 0.02	[92]
Bar soap containing 1% Olea Europaea (Olive) Seed Powder	none	As supplied	12 subjects	1-wk clinical use test; test material applied twice daily to whole body	No significant clinical changes; no reported subjective discomfort	[93]
			SENSITIZATION			
			ANIMAL			
Aqueous solution composed of 2.2% Olea Europaea (Olive) Fruit Extract	not reported	As supplied	guinea pigs; no further details provided	Guinea pig maximization study with intradermal injection and topical application in accordance with OECD TG 406; challenge patch was applied neat; no further details provided	Not a sensitizer; no further details provided	[86]
Olea Europaea (Olive) Leaf Extract	not reported	25% for 1st induction; 100% for 2nd induction; 10% and 100% for challenge	5 guinea pigs/group; no further details provided	Skin sensitization study; no further details provided	Negative for sensitization; no further details provided	[14]
			HUMAN			
Product containing 0.0025% Olea Europaea (Olive) Fruit Extract and 0.035% Olea Europaea (Olive) Seed Powder	Not reported	0.5% w/v aqueous solution; 0.2 ml applied	100 subjects	HRIPT under occlusive patches; induction patch applied on the back for 9 total applications; 10-15 d non-treatment period followed by challenge patch applied to naïve site and scored at 48 h and 72 h post-application; Webril patch was 2 cm^2	No dermal sensitization	[94]

Table 13. Dermal irritation and sensitization studies.

Test Article	Vehicle	Concentration/Dose	Test Population	Procedure	Results	Reference
Lip balm containing 5% Olea Europaea (Olive) Leaf Extract	As supplied	0.05 ml	25 subjects	Maximization study under occlusive patches; induction and challenge sites pretreated with 0.25% sodium lauryl sulfate (0.05 ml); induction patch applied on upper outer arm for five 48-h total applications, application site allowed to air dry for 30 min prior to patching; 7-10 d non-treatment period followed by challenge patch applied to naïve site and scored at ~48 and 72 h post-application; patch was 13 mm Webril disc	No dermal sensitization; no adverse events reported	[95]
Olea Europaea (Olive) Leaf Extract	Not reported	20%	54 subjects	HRIPT using modified Shelanski method; no further details provided	No contact sensitization; no further details provided	[14]
Product containing 0.3% Olea Europaea (Olive) Leaf Extract	As supplied	0.02 ml	109 subjects	HRIPT under occlusive patches; induction patch applied on back for total of 9 applications; 13 d non-treatment period followed by challenge patch applied to naïve site and scored at 48 h post-application; patches were 50 mm^2 Finn chambers	No primary or cumulative dermal irritation, mean irritation index = 0.01; no dermal sensitization	[96]
Product containing 25% Olea Europaea (Olive) Seed Powder	water	0.02 ml	54 subjects	HRIPT under semi-occlusive patches; induction patch applied on back for total of 9 applications; 2-wk non-treatment period followed by challenge patch applied to naïve site and scored at 48 and 96 h post-application; Webril patch was 1 cm^2	No dermal sensitization	[97]
PHOTOSENSITIZATION						
HUMAN						
Product containing 0.01% Olea Europaea (Olive) Fruit Extract	Neat	40 mg	27 subjects	Photosensitization study under occlusive patch; repeat insult patch test with ultraviolet radiation (solar simulated); test material administered to same test site on mid or lower back area for 6 induction exposures over a 3 wk period; induction patches in place for 24 h, after which the sites were wiped off with dry gauze and exposed to 2 minimal erythema doses from a xenon arc solar simulator; after a 10 d non-treatment period, challenge patch applied to naïve site for 24 h in duplicate, one set removed after 24 h and irradiated with ½ minimal erythema dose plus 4 J/cm^2 UV; unirradiated patches served as control sites; test sites examined for reactions at 48 and 72 h post-irradiation; patch was 2 x 2 cm^2 Webril pad	Not a photosensitizer; no adverse events reported	[98]
Product containing 10% Olea Europaea (Olive) Leaf Extract	Neat	40 mg	25 subjects	Photosensitization study under occlusive patch; repeat insult patch test with ultraviolet radiation (solar simulated); test material administered to same test site on mid or lower back area for 6 induction exposures over a 3 wk period, application site allowed to air dry for 30 min prior to patching; induction patches in place for 24 h, after which the sites were wiped off with dry gauze and exposed to 3 minimal erythema doses from a xenon arc solar simulator; after a 11 d non-treatment period, challenge patch applied to naïve site for 24 h in duplicate, one set removed after 24 h and irradiated with ½ minimal erythema dose plus 4 J/cm^2 UV; unirradiated patches served as control sites; test sites examined for reactions at 48 and 72 h post-irradiation; patch was 2 x 2 cm^2 Webril pad	Not a photosensitizer; no adverse events reported	[99]

REFERENCES

1. Nikitakis J, Kowcz A. Web-Based International Cosmetic Ingredient Dictionary and Handbook. https://incipedia.personalcarecouncil.org/winci/. Washington, DC: Personal Care Products Council. Accessed 05/01/2023.

2. Burnett CL, Fiume MM, Bergfeld WF, et al. Safety Assessment of Plant-Derived Fatty Acid Oils. *Int J Toxicol.* 2017;36(Suppl):51S-129S.

3. Edziri H, Jaziri R, Chehab H, et al. A comparative study on chemical composition, antibiofilm and biological activities of leaves extracts of four Tunisian olive cultivars. *Heliyon.* 2019;5(5):e01604.

4. Omer SA, Elobeid MA, Elamin MH, et al. Toxicity of olive leaves (*Olea europaea* L.) in Wistar albino rats. *Asian J Anim Vet Adv.* 2012;7(11):1175-1182.

5. Liphschitz N, Gophna R, Hartman M, Biger G. The beginning of olive (*Olea europaea*) cultivation in the Old World: A reassessment. *J Archaeol Sci.* 1991;18(4):441-453.

6. Bracci T, Busconi M, Fogher C, Sebastiani L. Molecular studies in olive (*Olea europaea* L.): Overview on DNA markers applications and recent advances in genome analysis. *Plant Cell Rep.* 2011;30(4):449-462.

7. Hashmi MA, Khan A, Hanif M, Farooq U, Perveen S. Traditional uses, phytochemistry, and pharmacology of *Olea europaea* (olive). *Evid Based Complement Alternat Med.* 2015;2015:541591.

8. D'Angeli S, Falasca G, Matteucci M, Altamura MM. Cold perception and gene expression differ in *Olea europaea* seed coat and embryo during drupe cold acclimation. *New Phytol.* 2013;197(1):123-138.

9. Anonymous. 2022. Olea Europaea (Olive) Fruit Extract - Summary information. Unpublished data submitted by the Personal Care Products Council on September 1, 2022.

10. Anonymous. 2022. Olea Europaea (Olive) Leaf Extract - Summary information. Unpublished data submitted by the Personal Care Products Council on September 1, 2022.

11. Soni MG, Burdock GA, Christian MS, Bitler CM, Crea R. Safety assessment of aqueous olive pulp extract as an antioxidant or antimicrobial agent in foods. *Food Chem Toxicol.* 2006;44(7):903-915.

12. The Innovation Company. 2022. Olea Europaea (Olive) Fruit Extract Method of Manufacture. Unpublished data submitted by the Personal Care Products Council on August 15, 2022.

13. Medouni-Haroune L, Zaidi F, Medouni-Adrar S, Kecha M. Olive pomace: From an olive mill waste to a resource, an overview of the new treatments. *J Crit Rev.* 2018;5(6):1-6.

14. Anonymous. 2022. Summary information Olea Europaea (Olive) Fruit Juice Extract and Olea Europaea (Olive) Leaf Extract. Unpublished data submitted by the Personal Care Products Council on October 11, 2022.

15. Anonymous. 2022. Method of Manufacture - Olea Europaea (Olive) Leaf Extract and Olea Europaea (Olive) Leaf Water. Unpublished data submitted by the Personal Care Products Council on August 9, 2022.

16. Martiny TR, Raghavan V, de Moraes CC, da Rosa GS, GL D. Bio-based active packaging: Carrageenan film with olive leaf extract for lamb meat preservation. *Foods.* 2020;9(12):1759.

17. Anonymous. 2022. Method of Manufacture - Olea Europaea (Olive) Leaf Powder. Unpublished data submitted by the Personal Care Products Council on August 9, 2022.

18. Nediani C, Ruzzolini J, Romani A, Calorini L. Oleuropein, a bioactive compound from *Olea europaea* L., as a potential preventive and therapeutic agent in non-communicable diseases. *Antioxidants (Basel).* 2019;8(12):578.

19. Sofo A, Fausto C, Mininni AN, Dichio B, Lucini L. Soil management type differentially modulates the metabolomic profile of olive xylem sap. *Plant Physiol Biochem.* 2019;139:707-714.

20. Breakspear I, Guillaume C. A quantitative phytochemical comparison of olive leaf extracts on the Australian market. *Molecules.* 2020;25(18):4099.

21. Goldsmith CD, Vuong QV, Sadeqzadeh E, Stathopoulos CE, Roach PD, Scarlett CJ. Phytochemical properties and anti-proliferative activity of *Olea europaea* L. leaf extracts against pancreatic cancer cells. *Molecules.* 2015;20(7):12992-13004.

22. Liaqat S, Islam M, Saeed H, Iqtedar M, Mehmood A. Investigation of *Olea ferruginea* Roylebark extracts for potential in vitro antidiabetic and anticancer effects. *Turk J Chem.* 2021;45(1):92-103.

23. Mehmood A, Murtaza G. Phenolic contents, antimicrobial and antioxidant activity of *Olea ferruginea* Royle (Oleaceae). *BMC Complement Altern Med.* 2018;18(1):173.

24. Cheriyot KR, Olila D, Kateregga J. In-vitro antibacterial activity of seleted medicinal plants from Longisa region of Bomet district, Kenya. *Afr Health Sci.* 2009;9(S1):42-46.

25. Taamalli A, Abaza L, Roman DA, et al. Characterisation of phenolic compounds by HPLC-TO/IT/MS in buds and open flowers of "Chemlali" olive cultivar. *Phytochem Anal.* 2013;24(5):504-512.

26. Rhouma HE, Trabelsi N, Chimento A, et al. *Olea europaea* L. flowers as a new promising anticancer natural product: Phenolic composition, antiproliferative activity and apoptosis induction. *Nat Prod Res.* 2021;35(11):1836-1839.

27. Mahdavi FS, Mardi P, Mahdavi SS, et al. Therapeutic and preventive effects of *Olea europaea* extract on indomethacin-induced small intestinal injury model in rats. . *Evid Based Complement Alternat Med.* 2020;2020:6669813.

28. Guinda A, Rada M, Delgado T, Gutierrez-Adanez P, Castellano JM. Pentacyclic triterpenoids from olic fruit and leaf. *J Agric Food Chem.* 2010;58(17):9685-9691.

29. Drakou M, Birmpa A, Koutelidakis AE, Komaitis M, Panagou EZ, Kapsokefalou M. Total antioxidant capacity, total phenolic content and iron and zinc dialyzability in selected Greek varieties of table olives, tomatoes and legumes from conventional and organic farming. *Int J Food Sci Nutr.* 2015;66(2):197-202.

30. Beltran G, Bejaoui MA, Jimenez A, Sanchez-Ortiz A. Ethanol in olive fruit. Changes during ripening. *J Agric Food Chem.* 2015;63(22):5309-5312.

31. Dekdouk N, Malafronte N, Russo D, et al. Phenolic compounds from *Olea europaea* L. possess antioxidant activity and inhibit carbohydrate metabolizing enzymes in vitro. *Evid Based Complement Alternat Med.* 2015;2015:684925.

32. Kritikou E, Kalogiouri NP, Kolyvira L, Thomaidis NS. Target and suspect HRMS metabolomics for the determination of functional ingredients in 13 varieties of olive leaves and drupes from Greece. *Molecules.* 2020;25(21):4889.

33. Martinez L, Castillo J, Ros G, Nieto G. Antioxidant and antimicrobial activity of rosemary, pomegranate and olive extracts in fish patties. *Antioxidants (Basel).* 2019;8(4):86.

34. Tamasi G, Baratto MC, Bonechi C, et al. Chemical characterization and antioxidant properties of products and by-products from *Olea europaea* L. *Food Sci Nutr.* 2019;7(9):2907-2920.

35. Omar SH, Kerr PG, Scott CJ, Hamlin AS, Obied HK. Olive (*Olea europaea* L.) biophenols: A nutriceutical against oxidative stress in SH-SY5Y cells. *Molecules.* 2017;22(11):1858.

36. Qabaha K, Al-Rimawi F, Qasem A, Naser SA. Oleuropein is responsible for the major anti-inflammatory effects of olive leaf extract. *J Med Food.* 2018;21(3):302-305.

37. Giacometti J, Zauhar G, Zuvic M. Optimization of ultrasonic-assisted extraction of major phenolic compounds from olive leaves (*Olea europaea* L.) using response surface methodology. *Foods.* 2018;7(9):149.

38. Nicoli F, Negro C, Vergine M, et al. Evaluation of phytochemical and antioxidant properties of 15 Italian *Olea europaea* L. cultivar leaves. *Molecules.* 2019;24(10):1998.

39. Zairi A, Nouir S, Zarrouk A, Haddad H, Khelifa A, Achour L. Phytochemical profile, cytotoxic, antioxidant, and allelopathic potentials of aqueous leaf extracts of *Olea europaea*. *Food Sci Nutr.* 2020;8:4805-4813.

40. Sarikurkcu C, Locatelli M, Tartaglia A, et al. Enzyme and biological activities of the water extracts from the plants *Aesculus hippocastanum, Olea europaea* and *Hypericum perforatum* that are used as folk remedies in Turkey. *Molecules.* 2020;25(5):1202.

41. Yu M, Gouvinhas I, Rocha J, Barros AIRNA. Phytochemical and antioxidant analysis of medicinal and food plants towards bioactive food and pharmaceutical resources. *Sci Rep.* 2021;11(1):10041.

42. Cataldi TRI, Margiotta G, Iasi L, Di Chio B, Xiloyannis C, Bufo SA. Determination of sugar compounds in olive plant extract by anion-exchange chromatography with pulsed amperometric detection. *Anal Chem.* 2000;72(16):3902-3907.

43. Saad AB, Tiss M, Keskes H, et al. Antihyperlipidemic, antihyperglycemic, and liver function protection of *Olea europaea* var. Meski stone and seed extracts: LC-ESI-HRMS-based composition analysis. *J Diabetes Res.* 2021;2021:6659415.

44. Reis R, Sipahi H, Zeybekoglu G, et al. Hydroxytyrosol: The phytochemical responsible for bioactivity of tradtionally used olive pits. *Euroasian J Hepatogastroenterol.* 2018;8(2):126-132.

45. Perez-Bonilla M, Salido S, van Beek TA, et al. Isolation and identification of radical scavengers in olive tree (*Olea europaea*) wood. *J Chromat A.* 2006;1112(1-2):311-318.

46. Perez-Bonilla M, Salido S, van Beek TA, et al. Isolation of antioxidative secoiridoids from olive wood (*Olea europaea* L.) guided by on-line HPLC-DAD-radical scavenging detection. *Food Chem.* 2011;124(1):36-41.

47. U.S. Food and Drug Administration Center for Food Safety & Applied Nutrition (CFSAN). Voluntary Cosmetic Registration Program - Frequency of Use of Cosmetic Ingredients. College Park, MD. 2023. (Obtained under the Freedom of Information Act from CFSAN; requested as "Frequency of Use Data" January 4, 2023; received February 2, 2023.)

48. Personal Care Products Council. 2020. Concentration of Use Information by FDA Product Category: Olive-Derived Ingredients. Unpublished data submitted by the Personal Care Products Council on February 28, 2020.

49. Personal Care Products Council. 2023. Concentration of Use by FDA Product Category: Hydrolyzed Olive Fruit, Hydrolyzed Olive Fruit Extract, and Hydrolyzed Olive Leaf Extract. Unpublished data submitted by the Personal Care Products Council on February 22, 2023.

50. EUR-Lex. Regulation (EC) No 1223/2009 of the European Parliament and of the Council of 30 November 2009 on cosmetic products. https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX%3A02009R1223-20221217&qid=1681317694221. Accessed 04/28/2023. .

51. Flemmig J, Kuchta K, Arnhold J, Rauwald HW. *Olea europaea* leaf (Ph.Eur.) extract as well as several of its isolated phenolics inhibit the gout-related enzyme xanthine oxidase. *Phytomedicine.* 2010;18(7):561-566.

52. Bouzabata A. Traditional treatment of high blood pressure and diabetes in Souk Ahras District. *J Pharmacogn Phytotherapy.* 2013;5(1):12-20.

53. Mehraein F, Sarbishegi M, Golipoor Z. Different effects of olive leaf extract on antioxidant enzyme activities in midbrain and dopaminergic neurons of Substantia Nigra in young and old rats. *Histol Histopathol.* 2016;31(4):425-431.

54. Karygianni L, Cecere M, Skaltsounis AL, et al. High-level antimicrobial efficacy of representative Mediterranean natural plant extracts against oral microorganisms. *Biomed Res Int.* 2014;2014:839019.

55. Markin D, Duek L, Berdicevsky I. In vitro antimicrobial activity of olive leaves. *Mycoses.* 2003;46(3-4):132-136.

56. Ali NH, Faizi S, Kazmi SU. Antibacterial activity in spices and local medicinal plants against clinical isolates of Karachi, Pakistan. *Pharm Biol.* 2011;49(8):833-839.

57. Misganaw D, Engidawork E, Nedi T. Evaluation of the anti-malarial activity of crude extract and solvent fractions of the leaves of *Olea europaea* (*Oleaceae*) in mice. *BMC Complement Altern Med.* 2019;19(1):171.

58. De Cicco P, Maisto M, Tenore GC, Ianaro A. Olive leaf extract, from *Olea europaea* L., reduces palmitate-induced inflammation via regulation of murine macrophages polarization. *Nutrients.* 2020;12(12):3663.

59. Fukumitsu S, Villareal MO, Aida K, et al. Maslnic acid in olive fruit alleviates mild knee joint pain and improves quality of life by promoting weight loss in the elderly. *J Clin Biochem Nutr.* 2016;59(3):220-225.

60. Lee-Huang S, Zhang L, Huang PL, Chang Y-T, Huang PL. Anti-HIV activity of olive leaf extract (OLE) and modulation of host cell gene expression by HIV-1 infection and OLE treatment. *Biochem Biophys Res Commun.* 2003;307(4):1029-1037.

61. Pais P, Villar A, Rull S. Impact of a proprietary standardized olive fruit extract (SOFE) on Cardio-Ankle Vascular Index, visual analog scale and C-reactive protein assessments in subjects with arterial stiffness risk. *Drugs R D.* 2016;16(4):355-368.

62. Jemai H, Mahmoudi A, Feryeni A, et al. Hepatoprotective effect of oleuropein-rich extract from olive leaves against cadmium-induced toxicity in mice. *Biomed Res Int.* 2020;2020(4398924):1-9.

63. Mohagheghi F, Bigdeli MR, Rasoulian B, Hashemi P, Pour MR. The neuroprotective effect of olive leaf extract is related to improved blood-brain barrier permeability and brain edema in rat with experimental focal cerebral ischemia. *Phytomedicine.* 2011;18(2-3):170-175.

64. Rabiei Z, Bigdeli MR, Rasoulian B, Ghassempour A, Mirzajani F. The neuroprotection effect of pretreatment with olive leaf extract on brain lipidomics in rat stroke model. *Phytomedicine.* 2012;19(10):940-946.

65. Benot-Dominguez R, Tupone MG, Castelli V, et al. Olive leaf extract impairs mitochondria by pro-oxidant activity in MDA-MB-231 and OVCAR-3 cancer cells. *Biomed Pharmacother.* 2021;134:111139.

66. Skalli S, Hassikou R, Arahou M. An ethnobotanical survey of medicinal plants used for diabetes treatment in Rabat, Morocco. *Heliyon.* 2019;5(3):e01421.

67. Cumaoglu A, Rackova L, Stefek M, Kartal M, Maechler P, Karasu C. Effects of olive leaf polyphenols against H_2O_2 toxicity in insulin secreting β-cells. *Acta Biochim Pol.* 2011;58(1):45-50.

68. Wainstein J, Ganz T, Boaz M, et al. Olive leaf extract as a hypoglycemic agent in both human diabetic subjects and in rats. *J Med Food.* 2012;15(7):1-6.

69. Elkafrawy N, Younes K, Naguib A, et al. Antihypertensive efficacy and safety of a standardized herbal medicinal product of *Hibiscus sabdariffa* and *Olea europaea* extracts (NW Roselle): A phase-II, randomized, double-blind, captopril-controlled clinical trial. *Phytother Res.* 2020;34(12):3379-3387.

70. Ismail MA, Norhayati MN, Mohamad N. Olive leaf extract effect on cardiometabolic profile among adults with prehypertension and hypertension: A systematic review and meta-analysis. *PeerJ.* 2021;9:e11173.

71. Abugomaa A, Elbadawy M. Olive leaf extract modulates glycerol-induced kidney and liver damage in rats. *Environ Sci Pollut Res Int.* 2020;27(17):22100-22111.

72. Koca U, Suntar I, Akkol EK, Yilmazer D, Alper M. Wound repair potential of *Olea europaea* L. leaf extracts revealed by in vivo experimental models and comparative evaluation of the extracts' antioxidant activity. *J Med Food.* 2011;24(1-2):140-146.

73. Kang H, Koppula S. *Olea europaea* Linn. fruit pulp extract protects against carbon tetrachloride-induced hepatic damage in mice. *Indian J Pharm Sci.* 2014;76(4):274-280.

74. Altarejos J, Salido S, Perez-Bonilla M, et al. Preliminary assay on the radical scavenging activity of olive wood extracts. *Fitoterapia.* 2005;76(3-4):348-351.

75. Hailesilase GG, Rajeshwar Y, Hailu GS, Sibhat GG, Bitew H. In vico antimalarial evaluation of crude extract, solvent fractions, and TLC-isolated compounds from *Olea europaea* Linn Subsp. *cuspidata* (Oleaceae). *Evid Based Complement Alternat Med.* 2020;2020:12.

76. Garcia AV, Alvarez-Perez OB, Rojas R, Aguilar CN, Garrigos MC. Impact of olive extract addition on corn starch-based active edible films properties for food packaging applications. *Foods.* 2020;9(9):1339.

77. Thielmann J, Kohnen S, Hauser C. Antimicrobial activity of *Olea europaea* Linne extracts and their applicability as natural food preservative agents. *Int J Food Microbiol.* 2017;251:48-66.

78. Cohen SM, Flikushima S, Gooderham NJ, et al. GRAS 27 Flavoring Substances. *Food Technol.* 2015;69(1):41-59.

79. Christian MS, Sharper VA, Hoberman AM, et al. The toxicity profile of hydrolyzed aqueous olive pulp extract. *Drug Chem Toxicol.* 2004;27(4):309-330.

80. Gaube Guex C, Reginato FZ, Figuerdeo KC, et al. Safety assessment of ethanolic extract of *Olea europaea* L. leaves after acute and subacute administration to Wistar rats. *Regul Toxicol Pharmacol.* 2018;95:395-399.

81. Heilman J, Anyangwe N, Tran N, Edwards J, Beilstein P, Lopez J. Toxicological evaluation of an olive extract, H35: Subchronic toxicity in the rat. *Food Chem Toxicol.* 2015;84:18-28.

82. Clewell AE, Beres E, Vertesi A, et al. A comprehensive toxicological safety assessment of an extract of *Olea europaea* L. leaves (Bonolive™). *Int J Toxicol.* 2016;35(2):208-221.

83. Najafizadeh P, Dehghani F, Shahin MP, Taj SH. The effect of a hydro-alcoholic extract of olive fruit on reproductive argons in male Sprague-Dalwy rat. *Iran J Reprod Med.* 2013;11(4):293-300.

84. Verschaeve L, Edziri H, Anthonissen R, et al. In vitro toxicity and genotoxic activity of aqueous leaf extracts from four varieties of *Olea europea* (L.). *Pharmacogn Mag.* 2017;13(Suppl 1):S63-S68.

85. Juan ME, Wenzel U, Ruiz-Gutierrez V, Daniel H, Planas JM. Olive fruit extracts inhibit proliferation and induce apoptosis in HT-29 human colon cancer cells. *J Nutr.* 2006;136(10):2553-2557.

86. Anonymous. 2023. Olea Europaea (Olive) Fruit Extract - Summary Information. Unpublished data submitted by the Personal Care Products Council on March 8, 2023.

87. Anonymous. 2013. 4-Day face use test (face cream contains 0.0005% Olea Europaea (Olive) Fruit Extract). Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

88. Anonymous. 2013. Human patch test (face cream contains 0.0005% Olea Europaea (Olive) Fruit Extract). Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

89. Anonymous. 2002. Clinical safety in use test - moisturizer containing 0.047% Olea Europaea (Olive) Leaf Extract. Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

90. Anonymous. 2008. Human patch test - liquid lip color containing 1% Olea Europaea (Olive) Leaf Extract. Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

91. Anonymous. 2008. 5-Day use test (lips) - product containing 1% Olea Europaea (Olive) Leaf Extract. Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

92. Anonymous. 2013. Human patch test (scrub contains 0.025% Olea Europaea (Olive) Seed Powder). Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

93. Anonymous. 2013. 1-Week home use test of a bar soap containing 1% Olea Europaea (Olive) Seed Powder. Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

94. Anonymous. 2014. Repeated insult patch test (product contains 0.0025% Olea Europaea (Olive) Fruit Extract and 0.035% Olea Europaea (Olive) Seed Powder). Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

95. Anonymous. 2008. Evaluation of the contact-sensitization potential of a topical coded product in human skin by means fo the maximization assay (product contains 5% Olea Europaea (Olive) Leaf Extract). Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

96. Anonymous. 2010. Verification of the absence of sensitizing potential and of the good cutaneous compatibility of a cosmetic investigational product, by repeated epicutaneous applications under occlusive patch, in 110 (or 109) healthy adult subjects (product contains 0.3% Olea Europaea (Olive) Leaf Extract). Unpublished data submitted by the Personal Care Products Council on August 17, 2022.

97. Anonymous. 2007. Human repeat insult patch test with challenge (product contains 25% Olea Europaea (Olive) Seed Powder). Unpublished data submitted by the Personal Care Products Council on August 17, 2022.

98. Anonymous. 2011. An assessment of the photosensitization potential of three topical coded test products using a human photocontact allergenicity assay (blend contains 0.01% Olea Europaea (Olive) Fruit Extract). Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

99. Anonymous. 2010. An assessment of the photosensitization potential of two topical coded test products using a human photocontact allergenicity assay (product contains 10% Olea Europaea (Olive) Leaf Extract). Unpublished data submitted by the Personal Care Products Council on August 16, 2022.

100. Alvarez-Eire MG, Pineda de la Losa F, Varela Losada S, Gonzalex de la Cuesta C, Ricard Palacios R. Anaphylasix to olive fruit due to lipoprotein sensitization. *Allergol Immunopathol (Madr).* 2011;40(3):198-200.

101. Prados-Castaño M, Reguero-Capilla M, Bartolome B, Ochando Diez-Canseco M, Quiralte J. Allergens responsible of olive fruit ingestion anaphylaxis. *J Investig Allergol Clin Immunol.* 2022;33(5):8 pages.

102. Theoharides TC, Stewart JM, Tsilioni I. Tolerability and benefit of a tetramethoxyluteolin-containing skin lotion. *Int J Immunopathol Pharmacol.* 2017;30(2):146-151.

103. Kendall M, Batterham M, Obied H, Prenzler P, Ryan D, Robards K. Zero effect of multiple dosage of olive leaf supplements on urinary biomarkers of oxidative stress in healthy humans. *Nutrition.* 2009;25(3):270-280.

104. Wanitphakdeedecha R, Ng JNC, Junsuwan N, et al. Efficacy of olive leaf extract-containing cream for facial rejuvention: A pilot study. *J Cosmet Dermatol.* 2020;19(7):1662-1666.