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RIFM fragrance ingredient safety assessment, bisabolene, CAS registry number 495-62-5

A.M. Api [a], D. Belsito [b], D. Botelho [a], M. Bruze [c], G.A. Burton Jr. [d], J. Buschmann [e], M.A. Cancellieri [a], M.L. Dagli [f], M. Date [a], W. Dekant [g], C. Deodhar [a], A.D. Fryer [h], L. Jones [a], K. Joshi [a], M. Kumar [a], A. Lapczynski [a], M. Lavelle [a], I. Lee [a], D.C. Liebler [i], H. Moustakas [a], M. Na [a], T.M. Penning [j], G. Ritacco [a], J. Romine [a], N. Sadekar [a], T.W. Schultz [k], D. Selechnik [a], F. Siddiqi [a], I.G. Sipes [l], G. Sullivan [a,*], Y. Thakkar [a], Y. Tokura [m]

[a] *Research Institute for Fragrance Materials, Inc., 50 Tice Boulevard, Woodcliff Lake, NJ, 07677, USA*
[b] *Member Expert Panel for Fragrance Safety, Columbia University Medical Center, Department of Dermatology, 161 Fort Washington Ave., New York, NY, 10032, USA*
[c] *Member Expert Panel for Fragrance Safety, Malmo University Hospital, Department of Occupational & Environmental Dermatology, Sodra Forstadsgatan 101, Entrance 47, Malmo, SE, 20502, Sweden*
[d] *Member Expert Panel for Fragrance Safety, School of Natural Resources & Environment, University of Michigan, Dana Building G110, 440 Church St., Ann Arbor, MI, 58109, USA*
[e] *Member Expert Panel for Fragrance Safety, Fraunhofer Institute for Toxicology and Experimental Medicine, Nikolai-Fuchs-Strasse 1, 30625, Hannover, Germany*
[f] *Member Expert Panel for Fragrance Safety, University of Sao Paulo, School of Veterinary Medicine and Animal Science, Department of Pathology, Av. Prof. dr. Orlando Marques de Paiva, 87, Sao Paulo, CEP 05508-900, Brazil*
[g] *Member Expert Panel for Fragrance Safety, University of Wuerzburg, Department of Toxicology, Versbacher Str. 9, 97078, Würzburg, Germany*
[h] *Member Expert Panel for Fragrance Safety, Oregon Health & Science University, 3181 SW Sam Jackson Park Rd., Portland, OR, 97239, USA*
[i] *Member Expert Panel for Fragrance Safety, Vanderbilt University School of Medicine, Department of Biochemistry, Center in Molecular Toxicology, 638 Robinson Research Building, 2200 Pierce Avenue, Nashville, TN, 37232-0146, USA*
[j] *Member of Expert Panel for Fragrance Safety, University of Pennsylvania, Perelman School of Medicine, Center of Excellence in Environmental Toxicology, 1316 Biomedical Research Building (BRB) II/III, 421 Curie Boulevard, Philadelphia, PA, 19104-3083, USA*
[k] *Member Expert Panel for Fragrance Safety, The University of Tennessee, College of Veterinary Medicine, Department of Comparative Medicine, 2407 River Dr., Knoxville, TN, 37996- 4500, USA*
[l] *Member Expert Panel for Fragrance Safety, Department of Pharmacology, University of Arizona, College of Medicine, 1501 North Campbell Avenue, P.O. Box 245050, Tucson, AZ, 85724-5050, USA*
[m] *Member Expert Panel for Fragrance Safety, The Journal of Dermatological Science (JDS), Editor-in-Chief, Professor and Chairman, Department of Dermatology, Hamamatsu University School of Medicine, 1-20-1 Handayama, Higashi-ku, Hamamatsu, 431-3192, Japan*

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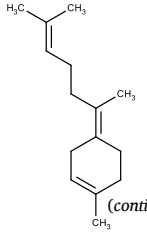
RIFM Fragrance Ingredient Safety Assessments is here: fragrancematerialsafetyresource.elsevier.com.

Name: Bisabolene
CAS Registry Number: 495-62-5
Additional CAS*
502-61-4 α-Farnesene
18794-84-8 β-Farnesene



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* Corresponding author.
E-mail address: gsullivan@rifm.org (G. Sullivan).

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17627-44-0 α-Bisabolene
495-61-4 *l*-β-Bisabolene (No Reported Use)
*Included because the materials are a mixture

Abbreviation/Definition List:

2-Box Model - A RIFM, Inc. proprietary *in silico* tool used to calculate fragrance air exposure concentration
AF - Assessment Factor
BCF - Bioconcentration Factor
CNIH – Confirmation of No Induction in Humans test. A human repeat insult patch test that is performed to confirm an already determined safe use level for fragrance ingredients (Na et al., 2021)
Creme RIFM Model - The Creme RIFM Model uses probabilistic (Monte Carlo) simulations to allow full distributions of data sets, providing a more realistic estimate of aggregate exposure to individuals across a population (Comiskey et al., 2015, 2017; Safford et al., 2015a, 2017) compared to a deterministic aggregate approach
DEREK - Derek Nexus is an *in silico* tool used to identify structural alerts
DRF - Dose Range Finding
DST - Dermal Sensitization Threshold
ECHA - European Chemicals Agency
ECOSAR - Ecological Structure-Activity Relationships Predictive Model
EU - Europe/European Union
GLP - Good Laboratory Practice
IFRA - The International Fragrance Association
LOEL - Lowest Observable Effect Level
MOE - Margin of Exposure
MPPD - Multiple-Path Particle Dosimetry. An *in silico* model for inhaled vapors used to simulate fragrance lung deposition
NA - North America
NESIL - No Expected Sensitization Induction Level
NOAEC - No Observed Adverse Effect Concentration
NOAEL - No Observed Adverse Effect Level
NOEC - No Observed Effect Concentration
NOEL - No Observed Effect Level
OECD - Organisation for Economic Co-operation and Development
OECD TG - Organisation for Economic Co-operation and Development Testing Guidelines
PBT - Persistent, Bioaccumulative, and Toxic
PEC/PNEC - Predicted Environmental Concentration/Predicted No Effect Concentration
Perfumery - In this safety assessment, perfumery refers to fragrances made by a perfumer used in consumer products only. The exposures reported in the safety assessment include consumer product use but do not include occupational exposures
QRA - Quantitative Risk Assessment
QSAR - Quantitative Structure-Activity Relationship
REACH - Registration, Evaluation, Authorisation, and Restriction of Chemicals
RfD - Reference Dose
RIFM - Research Institute for Fragrance Materials
RQ - Risk Quotient
Statistically Significant - Statistically significant difference in reported results as compared to controls with a $p < 0.05$ using appropriate statistical test
TTC - Threshold of Toxicological Concern
UV/Vis spectra - Ultraviolet/Visible spectra
VCF - Volatile Compounds in Food
VoU - Volume of Use
vPvB - (very) Persistent, (very) Bioaccumulative
WoE - Weight of Evidence

The Expert Panel for Fragrance Safety* concludes that this material is safe as described in this safety assessment.

This safety assessment is based on the RIFM Criteria Document (Api, 2015), which should be referred to for clarifications.

Each endpoint discussed in this safety assessment includes the relevant data that were available at the time of writing (version number in the top box is indicative of the date of approval based on a 2-digit month/day/year), both in the RIFM Database (consisting of publicly available and proprietary data) and through publicly

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available information sources (e.g., SciFinder and PubMed). Studies selected for this safety assessment were based on appropriate test criteria, such as acceptable guidelines, sample size, study duration, route of exposure, relevant animal species, most relevant testing endpoints, etc. A key study for each endpoint was selected based on the most conservative endpoint value (e.g., PNEC, NOAEL, LOEL, and NESIL).

*The Expert Panel for Fragrance Safety is an independent body that selects its own members and establishes its own operating procedures. The Expert Panel is comprised of internationally known scientists that provide RIFM with guidance relevant to human health and environmental protection.

Summary: The existing information supports the use of this material as described in this safety assessment.

Bisabolene was evaluated for genotoxicity, repeated dose toxicity, reproductive toxicity, local respiratory toxicity, phototoxicity/photoallergenicity, skin sensitization, and environmental safety. Data from the target material and read-across analog α-farnesene (CAS # 502-61-4) show that bisabolene is not expected to be genotoxic. The repeated dose, reproductive, and local respiratory toxicity endpoints were evaluated using the Threshold of Toxicological Concern (TTC) for a Cramer Class I material, and the exposure to bisabolene is below the TTC (0.03 mg/kg/day, 0.03 mg/kg/day, and 1.4 mg/day, respectively). Data provided bisabolene a No Expected Sensitization Induction Level (NESIL) of 3700 μg/cm^2 for the skin sensitization endpoint. The phototoxicity/photoallergenicity endpoints were evaluated based on ultraviolet/visible (UV/Vis) spectra; bisabolene is not expected to be phototoxic/photoallergenic. The environmental endpoints were evaluated; bisabolene was found not to be Persistent, Bioaccumulative, and Toxic (PBT) as per the International Fragrance Association (IFRA) Environmental Standards, and its risk quotients, based on the current volume of use in Europe and North America and read-across data from farnesane (CAS # 3891-98-3) (i.e., Predicted Environmental Concentration/Predicted No Effect Concentration, PEC/PNEC), are <1.

Human Health Safety Assessment

Genotoxicity: Not expected to be genotoxic. (RIFM, 2003a; RIFM, 2017b)
Repeated Dose Toxicity: No NOAEL available. Exposure is below the TTC.
Reproductive Toxicity: No NOAEL available. Exposure is below the TTC.
Skin Sensitization: NESIL = 3700 μg/cm^2. RIFM (2015)
Phototoxicity/Photoallergenicity: Not expected to be phototoxic/photoallergenic. (UV/Vis Spectra; RIFM Database)
Local Respiratory Toxicity: No NOAEC available. Exposure is below the TTC.

Environmental Safety Assessment

Hazard Assessment:
Persistence:
Critical Measured Value: 67% (OECD 301F; 67 days) for CAS # 495-62-5 RIFM, (2016c)
Bioaccumulation:
Screening-level: 4225 L/kg (EPI Suite v4.11; US EPA, 2012a)
Ecotoxicity:
Critical Measured Value: 21-day *Daphnia magna* NOEC: 0.054 mg/L (read-across to Farnesane [2,6,10-trimethyldodecane; CAS # 3891-98-3]) (ECHA REACH Dossier: 2,6,10-Trimethyldodecane (Farnesane); ECHA, 2013)
Conclusion: Not PBT or vPvB as per IFRA Environmental Standards
Risk Assessment:
Screening-level: PEC/PNEC (North America and Europe) > 1 (RIFM Framework; Salvito, 2002)
Critical Ecotoxicity Endpoint: 21-day *Daphnia magna* NOEC: 0.054 mg/L (read-across to Farnesane [2,6,10-trimethyldodecane; CAS # 3891-98-3]) (ECHA REACH Dossier: 2,6,10-Trimethyldodecane (Farnesane); ECHA, 2013)
RIFM PNEC is: 5.4 μg/L
- Revised PEC/PNECs (2015 IFRA VoU): North America and Europe <1

1. Identification

Chemical Name: Bisabolene	Chemical Name: α-Farnesene	Chemical Name: β-Farnesene	Chemical Name: α-Bisabolene	Chemical Name: *l*-β-Bisabolene
CAS Registry Number: 495-62-5	**CAS Registry Number:** 502-61-4	**CAS Registry Number:** 18794-84-8	**CAS Registry Number:** 17627-44-0	**CAS Registry Number:** 495-61-4
Synonyms: γ-Bisabolene; Cyclohexene, 4-(1,5-dimethyl-4-hexenylidene)-1-methyl-; 6-Methyl-2-(4-methylcyclohex-3-enyl)hept-1,5-diene; １-ﾒﾁﾙ-４-（２-ﾒﾁﾙ-6-ﾍﾌﾟﾃﾆﾘﾃﾞﾝ); 4-(1,5-Dimethylhex-4-en-1-ylidene)-1-methylcyclohexene; Bisabolene	**Synonyms:** (E,E)-α-Farnesene; *trans*-2,6,10-Trimethyl-2,6,9,11-dodecatetraene; *trans-trans*-α-Farnesene; 1,3,6,10-Dodecatetraene, 3,7,11-trimethyl-,(3E,6E)-; 1,3,6,10-Dodecatetraene,3,7,11-trimethyl (α-isomer); 1,6,10-Dodecatriene,7,11-dimethyl-3-methylene; 3,7,11-Trimethyldodeca-1,3,6,10-tetraene; Farnesene; Farnesene, mixture of isomers; α-Farnesene	**Synonyms:** E)-7,11-Dimethyl-3-methylene-1,6,10-dodecatriene; (E)-7,11-Dimethyl-3-methylenedodeca-1,6,10-triene; *trans*-β-Farnesene; 1,6,10-Dodecatriene, 7,11-dimethyl-3-methylene-, (E)-; 7,11-Dimethyl-3-methylenedodeca-1,6,10-triene; Farnesene; Magnolene; ﾄﾗﾝｽ-β-ﾌｧﾙﾈｾﾝ; β-Farnesene	**Synonyms:** １-メチル-４-｛２-メチル-２，５（又は２，６））-ヘプタジエン-６-イル｝シクロヘキセン-１; 2-Methyl-6-(4-methyl-3-cyclohexen-1-yl)-2,5-heptadiene; 4-(1,5-Dimethyl-1,4-hexadienyl)-1-methylcyclohexene; 4-(1,5-Dimethylhexa-1,4-dien-1-yl)-1-methylcyclohexene; 6-Methyl-2-(4-methylcyclohex-3-enyl)hept-2,5-diene; Bisabolen; Cyclohexene, 4-(1,5-dimethyl-1,4-hexadienyl)-1-methyl-; α-Bisabolene	**Synonyms:** Cyclohexene, 1-methyl-4-(5-methyl-1-methylene-4-hexenyl)-, (S)-; *l*-β-Bisabolene
Molecular Formula: $C_{15}H_{24}$	**Molecular Formula:** $C_{15}H_{24}$	**Molecular Formula:** $C_{15}H_{24}$	**Molecular Formula:** $C_{15}H_{24}$	**Molecular Formula:** $C_{15}H_{24}$
Molecular Weight: 204.35 g/mol	**Molecular Weight:** 204.35 g/mol	**Molecular Weight:** 204.35 g/mol	**Molecular Weight:** 204.35 g/mol	**Molecular Weight:** 204.35 g/mol
RIFM Number: 529	**RIFM Number:** 6140	**RIFM Number:** 5033	**RIFM Number:** 5121	**RIFM Number:** No RIFM number
Stereochemistry: One possible stereoisomer	**Stereochemistry:** One possible stereoisomer	**Stereochemistry:** One possible stereoisomer	**Stereochemistry:** One stereocenter and 2 possible stereoisomers	**Stereochemistry:** One stereocenter and 2 possible stereoisomers

2. Physical data

CAS # 495-62-5	CAS # 502-61-4	CAS # 18794-84-8	CAS # 17627-44-0	CAS # 495-61-4
Boiling Point: 271.84 °C (EPI Suite)	**Boiling Point:** 261.11 °C (EPI Suite)	**Boiling Point:** 254.57 °C (EPI Suite)	**Boiling Point:** 269.95 °C (EPI Suite)	**Boiling Point:** 263.59 °C (EPI Suite)
Flash Point: >93 °C (Globally Harmonized System), >200 °F; CC (Fragrance Materials Association [FMA])	**Flash Point:** Not available	**Flash Point:** >93 °C (EPI Suite)	**Flash Point:** >93 °C (EPI Suite)	**Flash Point:** >93 °C (EPI Suite)
Log K_{OW}: log Pow = >6.0 (RIFM, 2004b), 7.18 (EPI Suite)	**Log K_{OW}:** 7.1 (EPI Suite)	**Log K_{OW}:** 7.17 (EPI Suite)	**Log K_{OW}:** 7.05 (EPI Suite)	**Log K_{OW}:** 7.12 (EPI Suite)
Melting Point: 16.49 °C (EPI Suite)	**Melting Point:** -17.22 °C (EPI Suite)	**Melting Point:** -17.46 °C (EPI Suite)	**Melting Point:** 6.1 °C (EPI Suite)	**Melting Point:** 5.9 °C (EPI Suite)
Water Solubility: 0.008994 mg/L (EPI Suite)	**Water Solubility:** 0.642 ± 0.30 mg/L in laboratory freshwater (RIFM, 2000a); 0.01053 mg/L (EPI Suite)	**Water Solubility:** 0.009022 mg/L (EPI Suite)	**Water Solubility:** 0.01161 mg/L (EPI Suite)	**Water Solubility:** 0.009945 mg/L (EPI Suite)
Specific Gravity: 8712 (EOA, 1974 Sample 74–167), 0.860 (FMA)	**Specific Gravity:** Not available	**Specific Gravity:** Not available	**Specific Gravity:** Not available	**Specific Gravity:** Not available

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Vapor Pressure: 0.00897 mm Hg at 20 °C (EPI Suite v4.0), 0.03 mm Hg at 20 °C (FMA), 0.0143 mm Hg at 25 °C (EPI Suite)	**Vapor Pressure:** 0.0159 mm Hg at 20 °C, (EPI Suite v4.0), 0.025 mm Hg at 25 °C (EPI Suite)	**Vapor Pressure:** 0.0224 mm Hg at 20 °C (EPI Suite v4.0), 0.008 mm Hg at 20 °C (FMA), 0.0349 mm Hg at 25 °C (EPI Suite), 0.009022 mg/L (EPI Suite)	**Vapor Pressure:** 0.00993 mm Hg at 20 °C (EPI Suite v4.0), 0.0158 mm Hg at 25 °C (EPI Suite)	**Vapor Pressure:** 0.022 mm Hg at 25 °C (EPI Suite)
UV Spectra: Minor absorbance between 290 and 700 nm; molar absorption coefficient is below the benchmark (1000 L mol^{-1} • cm^{-1})	**UV Spectra:** Minor absorbance between 290 and 700 nm; molar absorption coefficient is below the benchmark (1000 L mol^{-1} • cm^{-1})	**UV Spectra:** Minor absorbance between 290 and 700 nm; molar absorption coefficient is below the benchmark (1000 L mol^{-1} • cm^{-1})	**UV Spectra:** Minor absorbance between 290 and 700 nm; molar absorption coefficient is below the benchmark (1000 L mol^{-1} • cm^{-1})	**UV Spectra:** Not Available
Appearance/ Organoleptic: A colorless, slightly viscous oil. Pleasant, warm, sweet-spicy-balsamic odor, inevitably reminding the perfumer of opopanax.	**Appearance/ Organoleptic:** Not available	**Appearance/ Organoleptic:** Not available	**Appearance/ Organoleptic:** Not available	**Appearance/ Organoleptic:** Not available

3. Volume of use (Worldwide band)

1. 10–100 metric tons per year (IFRA, 2015)

4. Exposure to fragrance ingredient (Creme RIFM aggregate exposure model v1.0)

1. **95**th **Percentile Concentration in Hydroalcoholics:** 0.026% (RIFM, 2017e)
2. **Inhalation Exposure*:** 0.000060 mg/kg/day or 0.0044 mg/day (RIFM, 2017e)
3. **Total Systemic Exposure**:** 0.00054 mg/kg/day (RIFM, 2017e)

*95th percentile calculated exposure derived from concentration survey data in the Creme RIFM Aggregate Exposure Model (Comiskey, 2015, 2017; Safford, 2015, 2017).

**95th percentile calculated exposure; assumes 100% absorption unless modified by dermal absorption data as reported in Section V. It is derived from concentration survey data in the Creme RIFM Aggregate Exposure Model and includes exposure via dermal, oral, and inhalation routes whenever the fragrance ingredient is used in products that include these routes of exposure (Comiskey, 2015, 2017; Safford, 2015, 2017).

5. Derivation of systemic absorption

1. **Dermal:** Assumed 100%
2. **Oral:** Assumed 100%
3. **Inhalation:** Assumed 100%

6. Computational toxicology evaluation

1. **Cramer Classification:** Class I, Low

Expert Judgment	Toxtree v3.1	OECD QSAR Toolbox v4.2
I	I	I

2. Analogs Selected:
   a. **Genotoxicity:** α-Farnesene (CAS # 502-61-4)
   b. **Repeated Dose Toxicity:** None
   c. **Reproductive Toxicity:** None
   d. **Skin Sensitization:** None
   e. **Phototoxicity/Photoallergenicity:** None
   f. **Local Respiratory Toxicity:** None
   g. **Environmental Toxicity:** Farnesane (CAS # 3891-98-3)
3. Read-across Justification: See Appendix below

7. Metabolism

No relevant data available for inclusion in this safety assessment.
Additional References: None.

8. Natural occurrence

Bisabolene is reported to occur in the following foods by the VCF*:

Carrot (*Daucus carota* L.)
Ginger (*Zingiber* species)

α-Farnesene is reported to occur in the following foods by the VCF*:

Apple brandy (Calvados)
Apple fresh (*Malus* species)
Apple processed (*Malus* species)
Beli, bael (*Aegle marmelos Correa*)
Camomile

β-Farnesene is reported to occur in the following foods by the VCF*:

Alpinia species
Angelica (*Angelica archangelica* L.)
Ashanti pepper (*Piper guineense* Schum and Thom)
Camomile
Capsicum species

α-Bisabolene is reported to occur in the following foods by the VCF*:

Citrus fruits
Hop (*Humulus lupulus*)
Laurel (*Laurus nobilis* L.)
Mace (*Myristica fragrans Houttuyn*)
Mushroom

l-β-Bisabolene is reported to occur in the following foods by the VCF*:

Mastic (*Pistacia lentiscus*)

*VCF (Volatile Compounds in Food): Database/Nijssen, L.M.; Ingen-Visscher, C.A. van; Donders, J.J.H. (eds). – Version 15.1 – Zeist (The Netherlands): TNO Triskelion, 1963–2014. A continually updated database containing information on published volatile compounds that have been found in natural (processed) food products. Includes FEMA GRAS and EU-Flavis data.

9. REACH dossier

Dossier available for CAS # 18794-84-8 (ECHA, 2015); accessed on 09/29/21; no dossiers available for the other materials.

10. Conclusion

The maximum acceptable concentrations[a] in finished products for bisabolene are detailed below.

IFRA Category[b]	Description of Product Type	Maximum Acceptable Concentrations[a] in Finished Products (%)
1	Products applied to the lips (lipstick)	0.28
2	Products applied to the axillae	0.085
3	Products applied to the face/body using fingertips	1.7
4	Products related to fine fragrances	1.6
5A	Body lotion products applied to the face and body using the hands (palms), primarily leave-on	0.40
5B	Face moisturizer products applied to the face and body using the hands (palms), primarily leave-on	0.40
5C	Hand cream products applied to the face and body using the hands (palms), primarily leave-on	0.40
5D	Baby cream, oil, talc	0.40
6	Products with oral and lip exposure	0.93
7	Products applied to the hair with some hand contact	3.2
8	Products with significant ano-genital exposure (tampon)	0.17
9	Products with body and hand exposure, primarily rinse-off (bar soap)	3.1
10A	Household care products with mostly hand contact (hand dishwashing detergent)	11

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IFRA Category[b]	Description of Product Type	Maximum Acceptable Concentrations[a] in Finished Products (%)
10B	Aerosol air freshener	11
11	Products with intended skin contact but minimal transfer of fragrance to skin from inert substrate (feminine hygiene pad)	6.2
12	Other air care products not intended for direct skin contact, minimal or insignificant transfer to skin	No Restriction

Note: [a]Maximum acceptable concentrations for each product category are based on the lowest maximum acceptable concentrations (based on systemic toxicity, skin sensitization, or any other endpoint evaluated in this safety assessment). For bisabolene, the basis was a skin sensitization NESIL of 3700 μg/cm^2.
[b]For a description of the categories, refer to the IFRA RIFM Information Booklet (https://www.rifm.org/downloads/RIFM-IFRA%20Guidance-for-the-use-of-IFRA-Standards.pdf; December 2019).

11. Summary

11.1. Human health endpoint summaries

11.1.1. Genotoxicity

Based on the current existing data and use levels, bisabolene does not present a concern for genetic toxicity.

11.1.1.1. Risk assessment. The mutagenic activity of bisabolene has been evaluated in a bacterial reverse mutation assay conducted in compliance with GLP regulations and in accordance with OECD TG 471 using the standard plate incorporation and preincubation methods. *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537, and TA102 were treated with bisabolene in ethanol at concentrations up to 5000 μg/plate. No increases in the mean number of revertant colonies were observed at any tested concentration in the presence or absence of S9 (RIFM, 2003a). Under the conditions of the study, bisabolene was not mutagenic in the Ames test.

There are no studies assessing the clastogenic activity of bisabolene; however, read-across can be made to α-farnesene (CAS # 502-61-4; see Section VI).

The clastogenic activity of α-farnesene was evaluated in an *in vitro* micronucleus test conducted in compliance with GLP regulations and in accordance with OECD TG 487. Human peripheral blood lymphocytes were treated with α-farnesene in dimethylformamide (DMF) at concentrations up to 1000 μg/mL in a dose range finding (DRF) study; micronuclei analysis was conducted at concentrations up to 200 μg/mL in the presence and absence of metabolic activation. α-Farnesene did not induce binucleated cells with micronuclei when tested up to the cytotoxic level concentration in either the presence or absence of an S9 activation system (RIFM, 2017b). Under the conditions of the study, α-farnesene was considered to be non-clastogenic in the *in vitro* micronucleus test, and this can be extended to bisabolene.

Based on the data available, α-farnesene does not present a concern for genotoxic potential, and this can be extended to bisabolene.

Additional References: None.

Literature Search and Risk Assessment Completed On: 09/24/21.

11.1.2. Repeated dose toxicity

There are insufficient repeated dose toxicity data on bisabolene or any read-across materials. The total systemic exposure to bisabolene is below the TTC for the repeated dose toxicity endpoint of a Cramer Class I material at the current level of use.

11.1.2.1. Risk assessment. There are no repeated dose toxicity data on bisabolene or any read-across materials that can be used to support the repeated dose toxicity endpoint. The total systemic exposure (0.54 μg/kg/day) is below the TTC for bisabolene (30 μg/kg/day; Kroes, 2007).

Additional References: None.

Literature Search and Risk Assessment Completed On: 09/17/21.

11.1.3. Reproductive toxicity

There are insufficient reproductive toxicity data on bisabolene or any read-across materials. The total systemic exposure to bisabolene is below the TTC for the reproductive toxicity endpoint of a Cramer Class I material at the current level of use.

11.1.3.1. Risk assessment. There are no fertility or developmental toxicity data on bisabolene or any read-across materials that can be used to support the reproductive toxicity endpoint. The total systemic exposure (0.54 μg/kg/day) is below the TTC for bisabolene (30 μg/kg/day; Kroes, 2007; Laufersweiler, 2012).

Additional References: None.

Literature Search and Risk Assessment Completed On: 09/17/21.

11.1.4. Skin sensitization

Based on the existing data, bisabolene is considered a sensitizer with a defined NESIL of 3700 μg/cm^2.

11.1.4.1. Risk assessment. Based on the existing data, bisabolene is considered a skin sensitizer. The chemical structure of this material indicates that it would not be expected to react with skin proteins directly, while its metabolites and autoxidation products are expected to be reactive (Roberts, 2007; Toxtree v3.1.0; OECD Toolbox v4.2). Bisabolene was found to be negative in an *in vitro* direct peptide reactivity assay (DPRA), while oxidized bisabolene was found to be positive (Natsch, 2007). Additionally, β-farnesene, a component chemical of bisabolene, was found to be negative in an *in vitro* DPRA, but positive in KeratinoSens, and human cell line activation test (h-CLAT) (RIFM, 2016a; RIFM, 2016b; RIFM, 2017a). In a murine local lymph node assay (LLNA), bisabolene was found to be sensitizing with an EC3 value of 8.6% (2150 μg/cm^2) (RIFM, 2002), while β-farnesene was found not to be sensitizing up to 30% (7500 μg/cm^2). In a guinea pig maximization test, β-farnesene led to skin sensitization reactions (RIFM, 1982c). In a series of modified Buehler sensitization studies with β-farnesene, skin sensitization reactions were observed (RIFM, 1981a; RIFM, 1981b). In a human maximization test with bisabolene, no skin sensitization reactions were observed (RIFM, 1974). In a Confirmation of No Induction in Humans test (CNIH) with 2500 μg/cm^2 of bisabolene in an unidentified vehicle, no reactions indicative of sensitization were observed in any of the 47 volunteers (RIFM, 2003b). Additionally, in a CNIH with 3779 μg/cm^2 of β-Farnesene in 1:3 ethanol:diethyl phthalate, no

Table 1
Data Summary for bisabolene.

LLNA Weighted Mean EC3 Value μg/cm^2 (No. Studies)	Potency Classification Based on Animal Data[a]	Human Data			
		NOEL-CNIH (Induction) μg/cm^2	NOEL-HMT (Induction) μg/cm^2	LOEL[b] (Induction) μg/cm^2	WoE NESIL[c] μg/cm^2
2150 [1]	Moderate	3779	6900	6350	3700

NOEL = No observed effect level; CNIH = Confirmation of No Induction in Humans test; HMT = Human Maximization test; LOEL = lowest observed effect level; NA = Not Available.
[a] Based on animal data using classification defined in ECETOC, Technical Report No. 87, 2003.
[b] Data derived from CNIH or HMT.
[c] WoE NESIL limited to 2 significant figures.

reactions indicative of sensitization were observed in any of the 110 volunteers (RIFM, 2015). However, in another CNIH with 6350 μg/cm^2 of β-farnesene in alcohol SD39C, sensitization reactions were observed in 1/51 volunteers (RIFM, 1982a; RIFM, 1982b).

Based on the weight of evidence (WoE) from structural analysis, animal, and human studies, bisabolene is a moderate sensitizer with a WoE NESIL of 3700 μg/cm^2 (Table 1). Section X provides the maximum acceptable concentrations in finished products, which take into account skin sensitization and application of the Quantitative Risk Assessment (QRA2) described by Api et al. (RIFM, 2020b).

Additional References: RIFM, 1984; ECHA, 2015.

Literature Search and Risk Assessment Completed On: 09/27/21.

11.1.5. Phototoxicity/photoallergenicity

Based on the available UV/Vis spectra, bisabolene would not be expected to present a concern for phototoxicity or photoallergenicity.

11.1.5.1. Risk assessment. There are no phototoxicity studies available for bisabolene in experimental models. UV/Vis absorption spectra indicate minor absorption between 290 and 700 nm. The corresponding molar absorption coefficient is below the benchmark of concern for phototoxicity and photoallergenicity (Henry, 2009). Based on the lack of significant absorbance in the critical range, bisabolene does not present a concern for phototoxicity or photoallergenicity.

11.1.5.2. UV spectra analysis. UV/Vis absorption spectra (OECD TG 101) were obtained. The spectra indicate minor absorbance in the range of 290–700 nm. The molar absorption coefficient is below the benchmark of concern for phototoxic effects, 1000 L mol^{-1} • cm^{-1} (Henry, 2009).

Additional References: None.

Literature Search and Risk Assessment Completed On: 09/17/21.

11.1.6. Local Respiratory Toxicity

The margin of exposure could not be calculated due to a lack of appropriate data. The exposure level for bisabolene is below the Cramer Class I TTC value for inhalation exposure local effects.

11.1.6.1. Risk assessment. There are insufficient inhalation data available on bisabolene. Based on the Creme RIFM Model, the inhalation exposure is 0.0044 mg/day. This exposure is 318.2 times lower than the Cramer Class I TTC value of 1.4 mg/day (based on human lung weight of 650 g; Carthew, 2009); therefore, the exposure at the current level of use is deemed safe.

Additional References: None.

Literature Search and Risk Assessment Completed On: 09/17/21.

11.2. Environmental endpoint summary

11.2.1. Screening-level assessment. A screening-level risk assessment of bisabolene was performed following the RIFM Environmental Framework (Salvito, 2002), which provides 3 tiered levels of screening for aquatic risk. In Tier 1, only the material's regional VoU, its log K_{OW}, and its molecular weight are needed to estimate a conservative risk quotient (RQ), expressed as the ratio Predicted Environmental Concentration/Predicted No Effect Concentration (PEC/PNEC). A general QSAR with a high uncertainty factor applied is used to predict fish toxicity, as discussed in Salvito et al. (2002). In Tier 2, the RQ is refined by applying a lower uncertainty factor to the PNEC using the ECOSAR model (US EPA, 2012b), which provides chemical class-specific ecotoxicity estimates. Finally, if necessary, Tier 3 is conducted using measured biodegradation and ecotoxicity data to refine the RQ, thus allowing for lower PNEC uncertainty factors. The data for calculating the PEC and PNEC for this safety assessment are provided in the table below. For the PEC, the range from the most recent IFRA Volume of Use Survey is reviewed. The PEC is then calculated using the actual regional tonnage, not the extremes of the range. Following the RIFM Environmental Framework, bisabolene was identified as a fragrance material with the potential to present a possible risk to the aquatic environment (i.e., its screening-level PEC/PNEC >1).

A screening-level hazard assessment using EPI Suite v4.1 (US EPA, 2012a) identified bisabolene as not persistent but possibly bioaccumulative based on its structure and physical–chemical properties. This screening-level hazard assessment considers the potential for a material to be persistent *and* bioaccumulative *and* toxic, or very persistent *and* very bioaccumulative as defined in the Criteria Document (Api, 2015). As noted in the Criteria Document, the screening criteria applied are the same as those used in the EU for REACH (ECHA, 2012). For persistence, if the EPI Suite model BIOWIN 3 predicts a value < 2.2 and either BIOWIN 2 or BIOWIN 6 predicts a value < 0.5, then the material is considered potentially persistent. A material would be considered potentially bioaccumulative if the EPI Suite model BCFBAF predicts a fish BCF ≥2000 L/kg. Ecotoxicity is determined in the above screening-level risk assessment. If, based on these model outputs (Step 1), additional assessment is required, a WoE-based review is then performed (Step 2). This review considers available data on the material's physical–chemical properties, environmental fate (e.g., OECD Guideline biodegradation studies or die-away studies), fish bioaccumulation, and higher-tier model outputs (e.g., US EPA's BIOWIN and BCFBAF found in EPI Suite v4.1). Data on persistence and bioaccumulation are reported below and summarized in the Environmental Safety Assessment section prior to Section 1.

11.2.2. Risk assessment. Based on the current Volume of Use (2015), bisabolene presents a risk to the aquatic compartment in the screening-level assessment.

11.2.2.1. Key studies

Biodegradation

CAS # 495-62-5.

RIFM, 2004a: The inherent biodegradability of the test material was evaluated by the manometric respirometry test according to the OECD 302C method. The nominal concentration of the test material was 30 mg/L. Under the conditions of the study, biodegradation of 46% was observed by day 28.

RIFM, 2010: The inherent biodegradability of the test material was determined by the modified manometric respirometry rest according to the OECD 302C method. Under the conditions of the study, the test material underwent 46% biodegradation by day 61.

RIFM, 2016c: The ready biodegradability of the test material was evaluated by the manometric respirometry test according to the OECD 301F method. Under the conditions of the study, biodegradation of 58% was observed after 28 days, 65% after 42 days, and 67% after 67 days.

Ecotoxicity

CAS # 495-62-5.

RIFM, 2017c: An algae (*Pseudokirchneriella subcapitata*) growth inhibition study was conducted according to the OECD 201 method in a closed system without headspace. Due to the low solubility and complex nature of the test materials, the test concentrations were prepared as water accommodation fractions (WAFs). Six WAFs were prepared with a nominal loading concentration of 0.316, 1.00, 3.16, 10.0, 31.6, and 100 mg/L. The concentrations of the test material in all test loadings were analytically verified by GC-FID analysis at the start and the end of the exposure. The measured concentrations ranged from 44 to 77% of the

initial measured concentrations. Based on the nominal test material loadings, the 0–72 h EL50 was reported to be > 100 mg/L for growth rate and 4.13 mg/L for yield. The NOEL was reported to be less than 0.316 mg/L for both growth rate and yield inhibition.

RIFM, 2017d: A 48-h *Daphnia magna* acute immobilization test was conducted according to the OECD 202 method under semi-static conditions in a closed system without headspace**.** Due to the low solubility and complex nature of the test materials, the test concentrations were prepared as a WAF. Five WAFs were prepared at loading levels of 0.111, 0.333, 1.00, 3.00, and 9.00 mg/L in a geometric series. The concentrations of the test material were analytically verified by GC-FID in freshwater media at the start of the exposure and renewal (0 and 24 h) and in old media at renewal and the end of the test (24 and 48 h) at all loading levels. The measured concentrations were in the range of 30%–83% of the initial measured concentrations. The geometric mean measured concentrations were reported to be 0.0513, 0.162, 0.560, 2.17, and 5.24 mg/L. Based on the nominal loadings of the test material, the 48-h EL10 and EL50 were reported to be 2.79 mg/L and 4.08 mg/L, respectively.

CAS # 502-61-4.

RIFM, 2000b: A 48-h *Daphnia magna* acute immobilization test was conducted according to the OECD 202 (Part I) method under static conditions. Due to the low solubility and complex nature of the test materials, the test concentrations were prepared as a WAF. The mean measured concentrations were 11.7, 26.3, 40.7, 89.6, and 189 μg/L and ranged between 62 and 66% of the 0-h measured concentrations. The calculated 48-h EC50 based on mean measured test concentrations were 110 μg/L.

Other available data

Bisabolene has been registered under REACH with no additional data available at this time.

Farnesane (2,6,10-trimethyldodecane; CAS # 3891-98-3) has been identified as a read-across material for bisabolene and the following data is available under REACH (ECHA, 2013):

A fish early life stage toxicity test was conducted according to the OECD 210 method under flow-through conditions and a solvent, N,N-dimethylformamide due to the poor water solubility of the test material. The fathead minnows were exposed to the test material at mean measured concentrations of 11–66 μg/L for 32 days (4-day hatching period followed by a 28-day post-hatch period). The 32-day no effect concentration (NOEC) for hatching success, survival, and growth was reported to be 66 μg/L (arithmetic mean of the measured test material concentration).

A *Daphnia magna* reproduction test was conducted according to the OECD 211 method under flow-through conditions and a solvent, N,N-dimethylformamide due to the poor water solubility of the test material. *Daphnia magna* specimens were exposed to the test material at mean measured concentrations of 12–77 μg/L for 21 days. The NOEC, based on growth and reproduction was reported to be 54 μg/L.

11.2.3. Risk assessment refinement. Ecotoxicological data and PNEC derivation (all endpoints reported in mg/L; PNECs in μg/L).

Endpoints used to calculate PNEC are underlined.

	LC50 (Fish) (mg/L)	EC50 (*Daphnia*) (mg/L)	EC50 (Algae) (mg/L)	AF	PNEC (μg/L)	Chemical Class
RIFM Framework Screening-level **(Tier 1)**	0.09128			1000000	9.13E-05	
ECOSAR Acute Endpoints **(Tier 2)** ***Ver 1.11***	0.043	0.035	0.121	10000	0.0035	Neutral Organic

Tier 3: Measured Data including Read-across

	LC50	EC50	NOEC	AF	PNEC
Fish			0.066		
Daphnia		0.11	0.054	10	5.4
Algae		> 100 (EL50)	4.13		

Exposure information and PEC calculation (following RIFM Environmental Framework: Salvito, 2002).

Exposure	Europe	North America
Log K_{ow} Used	>6.0	>6.0
Biodegradation Factor Used	1	1
Dilution Factor	3	3
Regional Volume of Use Tonnage Band*	1–10	1–10
Risk Characterization: PEC/PNEC	**<1**	**<1**

*Combined Regional Volume of Use Tonnage for all CAS #s.

Based on available data, the RQ for this material is < 1. No further assessment is necessary.

The RIFM PNEC is 5.4 μg/L. The revised PEC/PNECs for EU and NA are <1; therefore, the material presents no risk to the aquatic environment at the current reported volumes of use.

Literature Search and Risk Assessment Completed On: 09/17/21.

Literature Search*

- **RIFM Database:** Target, Fragrance Structure-Activity Group materials, other references, JECFA, CIR, SIDS
- **ECHA:** https://echa.europa.eu/
- **NTP:** https://ntp.niehs.nih.gov/
- **OECD Toolbox:** https://www.oecd.org/chemicalsafety/risk-assessment/oecd-qsar-toolbox.htm
- **SciFinder:** https://scifinder.cas.org/scifinder/view/scifinder/scifinderExplore.jsf
- **PubMed:** https://www.ncbi.nlm.nih.gov/pubmed
- **National Library of Medicine's Toxicology Information Services:** https://toxnet.nlm.nih.gov/
- **IARC:** https://monographs.iarc.fr
- **OECD SIDS:** https://hpvchemicals.oecd.org/ui/Default.aspx
- **EPA ACToR:** https://actor.epa.gov/actor/home.xhtml
- **US EPA HPVIS:** https://ofmpub.epa.gov/oppthpv/public_search.publicdetails?submission_id=24959241&ShowComments=Yes&sqlstr=null&recordcount=0&User_title=DetailQuery%20Results&EndPointRpt=Y#submission
- **Japanese NITE:** https://www.nite.go.jp/en/chem/chrip/chrip_search/systemTop
- **Japan Existing Chemical Data Base (JECDB):** http://dra4.nihs.go.jp/mhlw_data/jsp/SearchPageENG.jsp
- **Google:** https://www.google.com
- **ChemIDplus:** https://chem.nlm.nih.gov/chemidplus/

Search keywords: CAS number and/or material names.

*Information sources outside of RIFM's database are noted as appropriate in the safety assessment. This is not an exhaustive list. The links listed above were active as of 09/29/21.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fct.2022.112953.

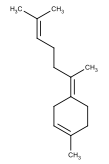
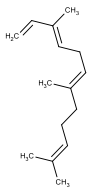
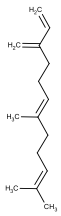
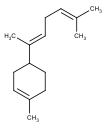
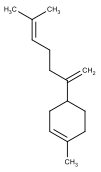
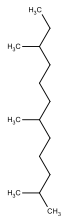
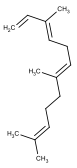
Appendix

Read-across Justification

Methods

The read-across analogs were identified using RIFM fragrance materials chemical inventory clustering and read-across search criteria (RIFM, 2020a). These criteria follow the strategy for structuring and reporting a read-across prediction of toxicity as described in Schultz et al. (2015) and are consistent with the guidance provided by OECD within Integrated Approaches for Testing and Assessment (OECD, 2015) and the European Chemical Agency read-across assessment framework (ECHA, 2017).

- First, materials were clustered based on their structural similarity. Second, data availability and data quality on the selected cluster were examined. Third, appropriate read-across analogs from the cluster were confirmed by expert judgment.
- Tanimoto structure similarity scores were calculated using FCFC4 fingerprints (Rogers and Hahn, 2010).
- The physical–chemical properties of the target material and the read-across analogs were calculated using EPI Suite v4.11 (US EPA, 2012a).
- J_{max} values were calculated using RIFM's Skin Absorption Model (SAM). The parameters were calculated using the consensus model (Shen et al., 2014).
- DNA binding, mutagenicity, genotoxicity alerts, oncologic classification, ER binding, and repeat dose categorization predictions were generated using OECD QSAR Toolbox v4.2 (OECD, 2018).
- Developmental toxicity was predicted using CAESAR v2.1.7 (Cassano et al., 2010).
- Protein binding was predicted using OECD QSAR Toolbox v4.2 (OECD, 2018), and skin sensitization was predicted using Toxtree.
- The major metabolites for the target material and read-across analogs were determined and evaluated using OECD QSAR Toolbox v4.2 (OECD, 2018).
- To keep continuity and compatibility with *in silico* alerts, OECD QSAR Toolbox v4.2 was selected as the alert system.

	Target Material (mixture)					Read-across Material	Read-across Material
Principal Name	Bisabolene	α-Farnesene	β-Farnesene	α-Bisabolene	*l*-β-Bisabolene	Farnesane	α-Farnesene
CAS No.	495-62-5	502-61-4	18794-84-8	17627-44-0	495-61-4	3891-98-3	502-61-4
Structure							
Similarity (Tanimoto Score)						N/A	N/A
Endpoint						• Environmental	• Genotoxicity
Molecular Formula	$C_{15}H_{24}$	$C_{15}H_{24}$	C15H24	$C_{15}H_{24}$	$C_{15}H_{24}$	$C_{15}H_{32}$	$C_{15}H_{24}$
Molecular Weight (g/mol)	204.357	204.357	204.357	204.357	204.357	212.42	204.357
Melting Point (°C, EPI Suite)	16.49	−17.22	−17.46	6.10	5.90	−19.80	−17.22
Boiling Point (°C, EPI Suite)	271.84	261.11	254.57	269.95	263.59	228.48	261.11
Vapor Pressure (Pa @ 25°C, EPI Suite)	1.91E+00	3.33E+00	4.65E+00	2.11E+00	2.93E+00	17.13	3.33E+00
Water Solubility (mg/L, @ 25°C, WSKOW v1.42 in EPI Suite)	8.99E-03	1.05E-02	9.02E-03	1.16E-02	9.95E-03	4.421E-03	1.05E-02
Log KOW	7.18	7.1	7.17	7.05	7.12	7.49	7.1
J_{max} (μg/cm^2/h, SAM)	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Henry's Law (Pa·m^3/mol, Bond Method, EPI Suite)	2.30E+05	2.67E+05	2.27E+05	1.95E+05	1.65E+05	2.22E+06	2.67E+05
Genotoxicity							
DNA Binding (OASIS v1.4, QSAR Toolbox v4.2)	No alert found	No alert found	No alert found	No alert found	No alert found		No alert found
DNA Binding (OECD QSAR Toolbox v4.2)	No alert found	No alert found	No alert found	No alert found	No alert found		No alert found
Carcinogenicity (ISS)	No alert found	No alert found	No alert found	No alert found	No alert found		No alert found
DNA Binding (Ames, MN, CA, OASIS v1.1)	No alert found	No alert found	No alert found	No alert found	No alert found		No alert found
***In Vitro* Mutagenicity (Ames, ISS)**	No alert found	No alert found	No alert found	No alert found	No alert found		No alert found
***In Vivo* Mutagenicity (Micronucleus, ISS)**	No alert found	No alert found	No alert found	No alert found	No alert found		No alert found
Oncologic Classification	Not classified	Not classified	Not classified	Not classified	Not classified		Not classified
Environmental toxicity							
BIOWIN 3	2.75	2.75	2.75	2.75	2.75	2.75	
ECOSAR (96-h Fish LC50) for Neutral Organics in mg/L	0.004	0.004	0.004	0.005	0.004	0.002	
ECOSAR (48-h *Daphnia* LC50) for Neutral Organics in mg/L	0.003	0.004	0.003	0.004	0.004	0.00194	
ECOSAR (96-h Algae LC50) Neutral Organics in mg/L	0.019	0.021	0.019	0.023	0.020	0.012	
Metabolism							
Rat Liver S9 Metabolism Simulator and Structural Alerts for Metabolites (OECD QSAR Toolbox v4.2)	See Supplemental Data 1	See Supplemental Data 2	See Supplemental Data 3	See Supplemental Data 4	See Supplemental Data 5	N/A	See Supplemental Data 6

Summary

There are insufficient toxicity data on bisabolene (CAS # 495-62-5). Hence, *in silico* evaluation was conducted to determine read-across analogs for this material. Based on structural similarity, reactivity, physical–chemical properties, and expert judgment, farnesane (CAS # 3891-98-3) and α-farnesene (CAS # 502-61-4) were identified as read-across analogs with sufficient data for toxicological evaluation.

Conclusions

- α-Farnesene (CAS # 502-61-4) was used as a read-across analog for the target material bisabolene (CAS # 495-62-5) for the genotoxicity endpoint.
  - o The target material and the read-across analog are structurally similar and belong to the structural class of alkenes.
  - o The target material and the read-across analog share a branched, alkyl hydrocarbon skeleton.
  - o The critical difference between the target material and the read-across analog is that the target material is a mixture of 5 components that are unsaturated, branched, and cyclic sesquiterpenes, while the read-across analog is one of the components of the target mixture, which is an unsaturated, branched sesquiterpene. Due to the availability of vinyl bonds in the read-across analog, it represents similar to more reactivity towards nucleic acids and proteins via epoxidation MOA.
  - o The physical–chemical properties of the target material and the read-across analog are sufficiently similar to compare their toxicological properties.

- o According to the OECD QSAR Toolbox v4.2, structural alerts for toxicological endpoints are consistent between the target material and the read-across analog.
- o The target material and the read-across analog are expected to be metabolized similarly, as shown by the metabolism simulator.
- o The structural alerts for the endpoints evaluated are consistent between the metabolites of the read-across analog and the target material.

- Farnesane (CAS # 3891-98-3) was used as a read-across analog for the target material bisabolene (CAS # 495-62-5) (mixture) for the environmental endpoint.
  - o The target material and the read-across analog are structurally similar and belong to the class of sesquiterpenes.
  - o The similarity between the target material and the read-across analog is indicated by the Tanimoto score. Differences between the structures that affect the Tanimoto score are toxicologically insignificant.
  - o The critical difference between the target material and the read-across analog is that the target material is a mixture of 5 components, which are unsaturated, branched, and cyclic sesquiterpenes alkenes. On the other hand, the read-across analog is a saturated, branched sesquiterpene alkane.
  - o In general, alkenes are more reactive compared to alkanes due to the availability of pi electrons. But for high log K_{ow} chemicals (low solubility, typically log K_{ow} >6), and towards aquatic toxicity, log K_{ow} drives the toxicity compared to the substructure features of the hydrocarbon skeleton. Also, the target material and the read-across analog belong to a class of hydrocarbons. The lack of electrophilic hetero atom (oxygen, nitrogen, or sulfur) in conjugation with pi electrons further reduces the reactivity of the target material and the read-across analog.
  - o The physical–chemical properties of the target material and the read-across analog are sufficiently similar to enable a comparison of their toxicological properties.
  - o According to the EPI Suite (BIOWIN v4.1 and ECOSAR v1.11), predictions of biodegradation and LC50 for fish, *Daphnia*, and algae within environmental toxicological endpoint are consistent between the target material and the read-across analog.
  - o Therefore, based on the above facts, farnesane presents an appropriate choice of the read-across analog for the environmental endpoint.

References

Api, A.M., Belsito, D., Bruze, M., Cadby, P., Calow, P., Dagli, M.L., Dekant, W., Ellis, G., Fryer, A.D., Fukayama, M., Griem, P., Hickey, C., Kromidas, L., Lalko, J.F., Liebler, D.C., Miyachi, Y., Politano, V.T., Renskers, K., Ritacco, G., Salvito, D., Schultz, T.W., Sipes, I.G., Smith, B., Vitale, D., Wilcox, D.K., 2015. Criteria for the Research Institute for fragrance materials, Inc. (RIFM) safety evaluation process for fragrance ingredients. Food Chem. Toxicol. 82, S1–S19.

Carthew, P., Clapp, C., Gutsell, S., 2009. Exposure based waiving: the application of the toxicological threshold of concern (TTC) to inhalation exposure for aerosol ingredients in consumer products. Food Chem. Toxicol. 47 (6), 1287–1295.

Cassano, A., Manganaro, A., Martin, T., Young, D., Piclin, N., Pintore, M., Bigoni, D., Benfenati, E., 2010. CAESAR models for developmental toxicity. Chem. Cent. J. (4 Suppl. 1), S4.

Comiskey, D., Api, A.M., Barratt, C., Daly, E.J., Ellis, G., McNamara, C., O'Mahony, C., Robison, S.H., Safford, B., Smith, B., Tozer, S., 2015. Novel database for exposure to fragrance ingredients in cosmetics and personal care products. Regul. Toxicol. Pharmacol. 72 (3), 660–672.

Comiskey, D., Api, A.M., Barrett, C., Ellis, G., McNamara, C., O'Mahony, C., Robison, S. H., Rose, J., Safford, B., Smith, B., Tozer, S., 2017. Integrating habits and practices data for soaps, cosmetics and air care products into an existing aggregate exposure model. Regul. Toxicol. Pharmacol. 88, 144–156.

ECHA, 2012. Guidance on information requirements and chemical safety assessment. November 2012 v2.1. http://echa.europa.eu/.

ECHA, 2013. 2,6,10-Trimethyldodecane (farnesane) Registration Dossier. Retrieved from.

ECHA, 2015. (E)-7,11-Dimethyl-3-methylenedodeca-1,6,10-triene registration dossier. Retrieved from. https://www.echa.europa.eu/web/guest/registration-dossier/-/registered-dossier/10936.

ECHA, 2017. Read-across Assessment Framework (RAAF). Retrieved from. www.echa.europa.eu/documents/10162/13628/raaf_en.pdf.

Henry, B., Foti, C., Alsante, K., 2009. Can light absorption and photostability data be used to assess the photosafety risks in patients for a new drug molecule? J. Photochem. Photobiol. B Biol. 96 (1), 57–62.

IFRA (International Fragrance Association), 2015. Volume of Use Survey. February 2015.

Kroes, R., Renwick, A.G., Feron, V., Galli, C.L., Gibney, M., Greim, H., Guy, R.H., Lhuguenot, J.C., van de Sandt, J.J.M., 2007. Application of the threshold of toxicological concern (TTC) to the safety evaluation of cosmetic ingredients. Food Chem. Toxicol. 45 (12), 2533–2562.

Laufersweiler, M.C., Gadagbui, B., Baskerville-Abraham, I.M., Maier, A., Willis, A., et al., 2012. Correlation of chemical structure with reproductive and developmental toxicity as it relates to the use of the threshold of toxicological concern. Regul. Toxicol. Pharmacol. 62 (1), 160–182.

Natsch, A., Gfeller, H., Rothaupt, M., Ellis, G., 2007. Utility and limitations of a peptide reactivity assay to predict fragrance allergens in vitro. Toxicol. Vitro 21 (7), 1220–1226.

OECD, 2015. Guidance Document on the Reporting of Integrated Approaches to Testing and Assessment (IATA). ENV/JM/HA, p. 7, 2015, Retrieved from. http://www.oecd.org/.

OECD, 2018. The OECD QSAR Toolbox, v3.2–4.2. Retrieved from. http://www.qsartoolbox.org/.

RIFM (Research Institute for Fragrance Materials, Inc), 1974. Report on Human Maximization Studies. Report to RIFM. RIFM Report Number 1801. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc), 1981a. Delayed Contact Hypersensitivity Study with Beta-Farnesene (Magnolene) in guinea Pigs. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from International Flavors and Fragrances. RIFM report number 53508.

RIFM (Research Institute for Fragrance Materials, Inc), 1981b. Delayed Contact Hypersensitivity Study with Beta-Farnesene (Magnolene) in guinea Pigs. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from International Flavors and Fragrances. RIFM report number 53509.

RIFM (Research Institute for Fragrance Materials, Inc), 1982a. Evaluation of Fragrance Materials by Dermal Contact in Human Subjects. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from International Flavors and Fragrances. RIFM report number 51299.

RIFM (Research Institute for Fragrance Materials, Inc), 1982b. Human Repeated Insult Patch Test with Beta-Farnesene (Magnolene). RIFM, Woodcliff Lake, NJ, USA. Unpublished report from International Flavors and Fragrances. RIFM report number 53504.

RIFM (Research Institute for Fragrance Materials, Inc), 1982c. Guinea Pig Sensitization Maximization Test (Magnusson-Kligman) with Beta-Farnesene (Magnolene). RIFM, Woodcliff Lake, NJ, USA. Unpublished report from International Flavors and Fragrances. RIFM report number 53507.

RIFM (Research Institute for Fragrance Materials, Inc), 1984. Klecak Open Epicutaneous Test: Determination of the Contact Sensitization Potential of Alpha-Bisabolene (Bisabolen) in the guinea Pig. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from Symrise. RIFM report number 62816.

RIFM (Research Institute for Fragrance Materials, Inc), 2000a. Determination of Water Solubility by the Shake Flask Method on Fragrance Materials. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from Firmenich SA. RIFM report number 39486.

RIFM (Research Institute for Fragrance Materials, Inc), 2000b. Alpha-Farnesene: Acute Toxicity of the Water Accommodated Fraction (WAF) to the Water Flea. RIFM, Woodcliff Lake, NJ, USA. Daphnia magna, determined under static test conditions. [Amendment Attached] Unpublished report from Firmenich SA. RIFM report number 42133.

RIFM (Research Institute for Fragrance Materials, Inc), 2002. Bisabolene: Local Lymph Node Assay (LLNA) in Mice (Identification of Contact Allergens). RIFM, Woodcliff Lake, NJ, USA. Unpublished report from Givaudan. RIFM report number 41935.

RIFM (Research Institute for Fragrance Materials, Inc), 2003a. Salmonella typhimurium Reverse Mutation Assay with Bisabolene. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from Givaudan. RIFM report number 56032.

RIFM (Research Institute for Fragrance Materials, Inc), 2003b. Repeated Insult Patch Test with Bisabolene. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from Givaudan. RIFM report number 56034.

RIFM (Research Institute for Fragrance Materials, Inc), 2004a. Inherent Biodegradability of Bisabolene. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from Givaudan. RIFM report number 56031.

RIFM (Research Institute for Fragrance Materials, Inc), 2004b. Partition Coefficient N-Octanol/water of Bisabolene. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from Givaudan. RIFM report number 56033.

RIFM (Research Institute for Fragrance Materials, Inc), 2010. Inherent Biodegradability of Bisabolene. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from Givaudan. RIFM report number 59956.

RIFM (Research Institute for Fragrance Materials, Inc), 2015. Beta-Farnesene: Repeated Insult Patch Test. RIFM Report Number 68614. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc), 2016a. Direct Peptide Reactivity Assay (DPRA) in Fragrance Materials. RIFM Report Number 72227. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc), 2016b. Induction of Antioxidant-Response-Element Dependent Gene Activity and Cytotoxicity (Using MTT) in the Keratinocyte ARE-Reporter Cell Line KeratinoSens. RIFM Report Number 72235. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc), 2016c. Bisabolene: Ready Biodegradability. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from Givaudan. RIFM report number 73109.
RIFM (Research Institute for Fragrance Materials, Inc), 2017a. Beta-Farnesene: in Vitro Sensitization: Dendritic Cell Line Activation Assay Human Cell Line Activation Test (H-CLAT). RIFM Report Number 72758. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc), 2017b. Alpha-Farnesene: in Vitro Human Lymphocyte Micronucleus Assay. RIFM Report Number 72856. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc), 2017c. Bisabolene: Alga, Growth Inhibition Test with Pseudokirchneriella Subcapitata, 72 Hours. RIFM, Woodcliff Lake, NJ, USA. Unpublished report from Givaudan. RIFM report number 73098.
RIFM (Research Institute for Fragrance Materials, Inc), 2017d. Bisabolene: acute immobilization test to Daphnia magna, semi-static, 48 hours. In: A Closed System without Headspace. Unpublished Report from Givaudan. RIFM Report Number 73099. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc), 2017e. Exposure Survey 16. May 2017.
RIFM (Research Institute for Fragrance Materials, Inc), 2020a. Clustering a Chemical Inventory for Safety Assessment of Fragrance Ingredients: Identifying Read-Across Analogs to Address Data Gaps. RIFM Report Number 76272. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc), 2020b. Updating Exposure Assessment for Skin Sensitization Quantitative Risk Assessment for Fragrance Materials. RIFM, Woodcliff Lake, NJ, USA. RIFM report number 76775.
Roberts, D.W., Patlewicz, G., Kern, P.S., Gerberick, F., Kimber, I., Dearman, R.J., Ryan, C. A., Basketter, D.A., Aptula, A.O., 2007. Mechanistic applicability domain classification of a local lymph node assay dataset for skin sensitization. Chem. Res. Toxicol. 20 (7), 1019–1030.
Rogers, D., Hahn, M., 2010. Extended-connectivity fingerprints. J. Chem. Inf. Model. 50 (5), 742–754.
Safford, B., Api, A.M., Barratt, C., Comiskey, D., Daly, E.J., Ellis, G., McNamara, C., O'Mahony, C., Robison, S., Smith, B., Thomas, R., Tozer, S., 2015. Use of an aggregate exposure model to estimate consumer exposure to fragrance ingredients in personal care and cosmetic products. Regul. Toxicol. Pharmacol. 72, 673–682.
Safford, B., Api, A.M., Barratt, C., Comiskey, D., Ellis, G., McNamara, C., O'Mahony, C., Robison, S., Rose, J., Smith, B., Tozer, S., 2017. Application of the expanded Creme RIFM consumer exposure model to fragrance ingredients in cosmetic, personal care and air care products. Regul. Toxicol. Pharmacol. 86, 148–156.
Salvito, D.T., Senna, R.J., Federle, T.W., 2002. A Framework for prioritizing fragrance materials for aquatic risk assessment. Environ. Toxicol. Chem. 21 (6), 1301–1308.
Schultz, T.W., Amcoff, P., Berggren, E., Gautier, F., Klaric, M., Knight, D.J., Mahony, C., Schwarz, M., White, A., Cronin, M.T., 2015. A strategy for structuring and reporting a read-across prediction of toxicity. Regul. Toxicol. Pharmacol. 72 (3), 586–601.
Shen, J., Kromidas, L., Schultz, T., Bhatia, S., 2014. An *in silico* skin absorption model for fragrance materials. Food Chem. Toxicol. 74, 164–176.
US EPA, 2012a. Estimation Programs Interface Suite for Microsoft Windows, v4.0–v4.11. United States Environmental Protection Agency, Washington, DC, USA.
US EPA, 2012b. The ECOSAR (ECOlogical Structure Activity Relationship) Class Program for Microsoft Windows, v1.11. United States Environmental Protection Agency, Washington, DC, USA.