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Short Review

RIFM fragrance ingredient safety assessment, methyl 3,3-dimethylbicyclo [2.2.1]heptane-2-carboxylate, CAS Registry Number 52557-97-8

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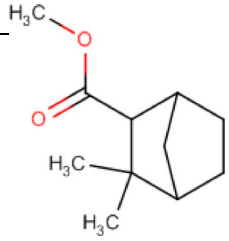
Version: 011419. This version replaces any previous versions.

Name: Methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate
CAS Registry Number: 52557-97-8



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
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
DST - Dermal Sensitization Threshold

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ECHA - European Chemicals Agency
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
QRA - Quantitative Risk Assessment
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 et al., 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 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.

Methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate was evaluated for genotoxicity, repeated dose toxicity, reproductive toxicity, local respiratory toxicity, phototoxicity/photoallergenicity, skin sensitization, and environmental safety. Data show that methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate is not genotoxic. Data on read-across material (2-endo,3-exo)-ethyl 3-(1-methylethyl) bicyclo [2.2.1] hept-5-ene-2-carboxylate (CAS # 116044-44-1) provide a calculated MOE > 100 for the repeated dose toxicity endpoint. The reproductive and local respiratory toxicity endpoints were evaluated using the TTC for a Cramer Class II material, and the exposure to methyl 3,3-dimethylbicyclo [2.2.1]heptane-2-carboxylate is below the TTC (0.009 mg/kg/day and 0.47 mg/day, respectively). The skin sensitization endpoint was completed using the DST for non-reactive materials (900 μg/cm^2); exposure is below the DST. The phototoxicity/photoallergenicity endpoints were evaluated based on data and UV spectra; methyl 3,3-dimethylbicyclo [2.2.1]heptane-2-carboxylate is not expected to be phototoxic/photoallergenic. The environmental endpoints were evaluated; methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate was found not to be PBT as per the IFRA Environmental Standards, and its risk quotients, based on its current volume of use in Europe and North America (i.e., PEC/PNEC), are < 1.

Human Health Safety Assessment

Genotoxicity: Not genotoxic.	(RIFM, 1980a; RIFM, 2018)
Repeated Dose Toxicity: NOAEL = 50 mg/kg/day.	RIFM (1996)
Reproductive Toxicity: No NOAEL available. Exposure is below the TTC.	
Skin Sensitization: No safety concerns at current, declared use levels; Exposure is below the DST.	
Phototoxicity/Photoallergenicity: Not phototoxic/photoallergenic.	(UV Spectra, RIFM Database; RIFM, 1981; RIFM, 1980c)
Local Respiratory Toxicity: No NOAEC available. Exposure is below the TTC.	

Environmental Safety Assessment

Hazard Assessment:	
Persistence:	
Screening-level: 2.7 (BIOWIN 3)	(EPI Suite v4.11; US EPA, 2012a)
Bioaccumulation:	
Screening-level: 81.76 L/kg	(EPI Suite v4.11; US EPA, 2012a)
Ecotoxicity:	
Screening-level: Fish LC50: 14.88 mg/L	(RIFM Framework; Salvito et al., 2002)
Conclusion: Not PBT or vPvB as per IFRA Environmental Standards	
Risk Assessment:	
Screening-level: PEC/PNEC (North America and Europe) < 1	(RIFM Framework; Salvito et al., 2002)
Critical Ecotoxicity Endpoint: Fish LC50: 14.88 mg/L	(RIFM Framework; Salvito et al., 2002)
RIFM PNEC is: 0.01488 μg/L	
•**Revised PEC/PNECs (2015 IFRA VoU):** North America and Europe: not applicable; cleared at screening-level	

1. Identification

1. **Chemical Name:** Methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate
2. **CAS Registry Number:** 52557-97-8
3. **Synonyms:** Bicyclo[2.2.1]heptane-2-carboxylic acid, 3,3-dimethyl-, methyl ester; 2-Norbornanecarboxylic acid, 3,3-dimethyl-, methyl ester; Cistulate; Methyl camphenoate; Methyl kamfenoate; Methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate
4. **Molecular Formula:** $C_{11}H_{18}O_2$

5. **Molecular Weight:** 182.26
6. **RIFM Number:** 5720
7. **Stereochemistry:** Isomer not specified. Three chiral centers and 8 total distereoisomers possible.

2. Physical data

1. **Boiling Point:** 216.96 °C (EPI Suite)
2. **Flash Point:** 82 °C (GHS)
3. **Log K_{OW}:** 3.4 (EPI Suite)
4. **Melting Point:** 21.85 °C (EPI Suite)
5. **Water Solubility:** 66.57 mg/L (EPI Suite)
6. **Specific Gravity:** Not Available
7. **Vapor Pressure:** 0.101 mm Hg @ 20 °C (EPI Suite v4.0), 0.153 mm Hg @ 25 °C (EPI Suite)
8. **UV Spectra:** No significant absorbance between 290 and 700 nm; molar absorption coefficient is below the benchmark (1000 L $mol^{-1} \cdot cm^{-1}$)
9. **Appearance/Organoleptic:** Not Available

3. Exposure to fragrance ingredient

1. **Volume of Use (Worldwide Band):** 0.1–1 metric ton per year (IFRA, 2015)
2. **95**th **Percentile Concentration in Hydroalcoholics:** 0.15% (RIFM, 2017)
3. **Inhalation Exposure*:** 0.00019 mg/kg/day or 0.014 mg/day (RIFM, 2017)
4. **Total Systemic Exposure**:** 0.0017 mg/kg/day (RIFM, 2017)

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

**95th percentile calculated exposure; assumes 100% absorption unless modified by dermal absorption data as reported in Section IV. 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 et al., 2015, 2017; Safford et al., 2015a, 2017).

4. Derivation of systemic absorption

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

5. Computational toxicology evaluation

1. **Cramer Classification:** Class II, Intermediate* (Expert Judgment)

Expert Judgment	Toxtree v 2.6	OECD QSAR Toolbox v 3.2
II	I	I

*Due to potential discrepancies between the current *in silico* tools (Bhatia et al., 2015), the Cramer Class of the target material was determined using expert judgment, based on the Cramer decision tree. See the Appendix for explanation.

2. Analogs Selected:
   a. **Genotoxicity:** None
   b. **Repeated Dose Toxicity:** (2-endo,3-exo)-Ethyl 3-(1-methylethyl) bicyclo[2.2.1] hept-5-ene-2-carboxylate (CAS # 116044-44-1)
   c. **Reproductive Toxicity:** None
   d. **Skin Sensitization:** None
   e. **Phototoxicity/Photoallergenicity:** None
   f. **Local Respiratory Toxicity:** None
   g. **Environmental Toxicity:** None
3. Read-across Justification: See Appendix below

6. Metabolism

No relevant data available for inclusion in this safety assessment.

6.1. Additional References

None.

7. Natural occurrence (discrete chemical) or composition (NCS)

Methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate is not reported to occur in foods by the VCF*.

*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.

8. IFRA standard

None.

9. REACH dossier

Pre-registered for 2010; no dossier available as of 01/14/19.

10. Summary

10.1. Human health endpoint summaries

10.1.1. Genotoxicity

Based on the current existing data, methyl 3,3-dimethylbicyclo [2.2.1]heptane-2-carboxylate does not present a concern for genotoxicity.

10.1.1.1. Risk assessment. The mutagenic activity of methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate has been evaluated in a bacterial reverse mutation assay conducted equivalent to OECD TG 471 using the standard plate incorporation method. *Salmonella typhimurium* strains TA100, TA1535, TA1536, TA1537, and TA1538 were treated with methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate in dimethyl sulfoxide (DMSO) at concentrations up to 10 μL/plate. No increases in the mean number of revertant colonies were observed at any tested concentration in the presence or absence of S9 (RIFM, 1980a). Under the conditions of the study, methyl 3,3-dimethylbicyclo [2.2.1]heptane-2-carboxylate was not mutagenic in the Ames test.

The clastogenic activity of methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate 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 methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate in DMSO at concentrations up to 1820 μg/mL in a dose range finding (DRF) study. Micronuclei analysis was conducted at 125 μg/mL in the presence and absence of metabolic activation (S9) for 4 h and in the absence of metabolic activation for 24 h. Methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate did not induce binucleated cells with micronuclei when tested up to cytotoxic levels in either the presence or absence of an S9 activation system (RIFM, 2018). Under the conditions of the study, methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate was considered to be non-clastogenic in the *in vitro* micronucleus test.

Table 1
Maximum acceptable concentrations for methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate that present no appreciable risk for skin sensitization based on non-reactive DST.

IFRA Category[a]	Description of Product Type	Maximum Acceptable Concentrations in Finished Products Based on Non-reactive DST	Reported 95th Percentile Use Concentrations in Finished Products
1	Products applied to the lips	0.069%	NRU[b]
2	Products applied to the axillae	0.021%	0.0030%
3	Products applied to the face using fingertips	0.41%	4.5×10^{-4}%
4	Fine fragrance products	0.39%	0.15%
5	Products applied to the face and body using the hands (palms), primarily leave-on	0.10%	0.010%
6	Products with oral and lip exposure	0.23%	NRU[b]
7	Products applied to the hair with some hand contact	0.79%	NRU[b]
8	Products with significant ano-genital exposure	0.041%	No Data[c]
9	Products with body and hand exposure, primarily rinse-off	0.75%	0.0072%
10	Household care products with mostly hand contact	2.7%	0.042%
11	Products with intended skin contact but minimal transfer of fragrance to skin from inert substrate	1.5%	No Data[c]
12	Products not intended for direct skin contact, minimal or insignificant transfer to skin	Not Restricted	0.36%

Note:
[a] For a description of the categories, refer to the IFRA/RIFM Information Booklet.
[b] No reported use.
[c] Fragrance exposure from these products is very low. These products are not currently in the Creme RIFM Aggregate Exposure Model.

Additional References: None.
Literature Search and Risk Assessment Completed On: 12/28/18.

10.1.2. Repeated dose toxicity

The margin of exposure (MOE) for 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate is adequate for repeated dose toxicity endpoint.

10.1.2.1. Risk assessment. There are no repeated dose toxicity data on methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate. Read-across material (2-endo,3-exo)-ethyl 3-(1-methylethyl)bicyclo[2.2.1]hept-5-ene-2-carboxylate (CAS # 116044-44-1; see Section V) has sufficient repeated dose toxicity data. In a GLP-compliant (equivalent to EU Method B.7) study, 5 Sprague Dawley rats/sex/dose were administered the treatment material at doses of 0 (vehicle: corn oil), 50, 150, and 1000 mg/kg/day through oral gavage for 28 days. No treatment-related adverse effects were reported for mortality, clinical signs, body weight, food consumption, hematology, clinical chemistry, and organ weights at any dose level. The absolute liver weight in females and relative liver weight in both sexes were significantly increased at 1000 mg/kg/day. In addition, relative kidney weight was significantly increased in male rats along with pale kidney appearance in 2 male rats of 1000 mg/kg/day group. A dose-dependent increase in the presence of intra-epithelial eosinophilic droplets in proximal tubules of male rats was reported along with accumulation of α-2u-globulin protein in the tubules of treated male rats. Kidney changes in males at ≥50 mg/kg/day were consistent with documented changes of α-2u-globulin nephropathy, which is species-specific to male rats. Thus, this effect is not considered a hazard to human health (Lehman-McKeeman and Caudill, 1992; Lehman-McKeeman et al., 1990). In males, brown pigmentation in intrahepatic bile ducts and portal regions of rats accompanied by inflammation of the portal triads were reported at the highest dose. However, this finding was less pronounced in their female counterparts. At the highest dose, degeneration of bile duct epithelium was observed in all males (and 1 female) along with bile ducts hypertrophy. Based on degenerative changes in bile duct epithelium and inflammatory cell reactions of the portal triad, the NOAEL was considered to be 150 mg/kg/day (RIFM, 1996).

A default safety factor of 3 was used when deriving a NOAEL from the 28-day study. The safety factor has been approved by Independent Expert Panel for Fragrance Safety*.

The derived NOAEL for the repeated dose toxicity data is 150/3 or 50 mg/kg/day. Therefore, the MOE can be calculated by dividing the NOAEL in mg/kg/day for (2-endo,3-exo)-ethyl 3-(1-methylethyl)bicyclo[2.2.1]hept-5-ene-2-carboxylate by the total systemic exposure (mg/kg/day) of methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate, 50/0.0017 or 29412.

In addition, the total systemic exposure to 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate (1.7 μg/kg bw/day) is below the TTC (9 μg/kg bw/day; Kroes et al., 2007) for the repeated dose toxicity endpoint of a Cramer Class II material at the current level of use.

*The Expert Panel for Fragrance Safety is composed of scientific and technical experts in the respective fields. This group provides advice and guidance.

Additional References: None.
Literature Search and Risk Assessment Completed On: 01/04/19.

10.1.3. Reproductive toxicity

There are no reproductive toxicity data on methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate or on any read-across materials. The total systemic exposure to methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate is below the TTC for the reproductive toxicity endpoint of a Cramer Class II material at the current level of use.

10.1.3.1. Risk assessment. There are no reproductive toxicity data on methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate or on any read-across materials that can be used to support the reproductive toxicity endpoint. The total systemic exposure to methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate (1.7 μg/kg bw/day) is below the TTC (9 μg/kg bw/day; Kroes et al., 2007; Laufersweiler et al., 2012) for the reproductive toxicity endpoint of a Cramer Class II material at the current level of use.

Additional References: None.
Literature Search and Risk Assessment Completed On: 12/18/18.

10.1.4. Skin sensitization

Based on the existing data and the application of DST, methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate does not present a safety concern for skin sensitization under the current, declared levels of use.

10.1.4.1. Risk assessment. The chemical structure of this material indicates that it would not be expected to react with skin proteins (Roberts et al., 2007; Toxtree 3.1.0; OECD Toolbox v4.2). No *in chemico*

or in *vitro* predictive skin sensitization studies are available for methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate. However, in a guinea pig maximization test using 12 test group animals and 10 control group animals, no skin sensitization reactions were observed (RIFM, 1980b). Acting conservatively, due to the insufficient data, the reported exposure was benchmarked utilizing the non-reactive DST of 900 μg/cm^2 (Safford, 2008, 2011, 2015b; Roberts et al., 2015). The current exposure from the 95th percentile concentration is below the DST for non-reactive materials when evaluated in all QRA categories. Table 1 provides the maximum acceptable concentrations for methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate that present no appreciable risk for skin sensitization based on the non-reactive DST. These levels represent maximum acceptable concentrations based on the DST approach. However, additional studies may show it could be used at higher levels.

Additional References: None.

Literature Search and Risk Assessment Completed On: 12/06/18.

10.1.5. Phototoxicity/photoallergenicity

Based on the available data and UV/Vis spectra, methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate would not be expected to present a concern for phototoxicity or photoallergenicity.

10.1.5.1. Risk assessment. UV/Vis absorption spectra indicate no significant absorption between 290 and 700 nm. The corresponding molar absorption coefficient is well below the benchmark of concern for phototoxicity and photoallergenicity (Henry et al., 2009). In studies conducted with rabbits and guinea pigs, application of methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate did not result in phototoxic or photoallergenic reactions, respectively, (RIFM, 1981; RIFM, 1980c). Based on the lack of absorbance and available study data, methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate does not present a concern for phototoxicity or photoallergenicity.

10.1.5.2. UV spectra analysis. UV/Vis absorption spectra (OECD TG 101) were obtained. The spectra indicate no significant 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 et al., 2009).

Additional References: None.

Literature Search and Risk Assessment Completed On: 12/04/18.

10.1.6. Local respiratory toxicity

The MOE could not be calculated due to lack of appropriate data. The exposure level for methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate is below the Cramer Class III* TTC value for inhalation exposure local effects.

10.1.6.1. Risk assessment. There are no inhalation data available on methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate. Based on the Creme RIFM Model, the inhalation exposure is 0.014 mg/day. This exposure is 33.6 times lower than the Cramer Class III* TTC value of 0.47 mg/day (based on human lung weight of 650 g; Carthew et al., 2009); therefore, the exposure at the current level of use is deemed safe.

*As per Carthew et al. (2009), Cramer Class II materials default to Cramer Class III for the local respiratory toxicity endpoint.

Additional References: None.

Literature Search and Risk Assessment Completed On: 12/11/18.

10.2. Environmental endpoint summary

10.2.1. Screening-level assessment

A screening-level risk assessment of methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate was performed following the RIFM Environmental Framework (Salvito et al., 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, methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate was identified as a fragrance material with the potential to present no possible risk to the aquatic environment (i.e., its screening-level PEC/PNEC < 1).

A screening-level hazard assessment using EPI Suite v4.11 (US EPA, 2012a) did not identify methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate as possibly being persistent or 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 et al., 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 $\geq$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 v4.11).

10.2.2. Risk assessment

Based on the current Volume of Use (2015), methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate presents no risk to the aquatic compartment in the screening-level assessment.

10.2.3. Key studies

10.2.3.1. Biodegradation. No data available.

10.2.3.2. Ecotoxicity. No data available.

10.2.4. Other available data

Methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate has been pre-registered for REACH with no additional data at this time.

10.2.5. 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)**	14.88			1000000	0.01488	

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

Exposure	Europe (EU)	North America (NA)
Log K_{ow} Used	3.4	3.4
Biodegradation Factor Used	0	0
Dilution Factor	3	3
Regional Volume of Use Tonnage Band	< 1	< 1
Risk Characterization: PEC/PNEC	**< 1**	**< 1**

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

The RIFM PNEC is 0.01488 μg/L. The revised PEC/PNECs for EU and NA are: not applicable. The material was cleared at screening-level and therefore does not present a risk to the aquatic environment at the current reported volumes of use.

Literature Search and Risk Assessment Completed On: 01/02/19.

11. 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**
- **SciFinder:** https://scifinder.cas.org/scifinder/view/scifinder/scifinderExplore.jsf
- **PubMed:** https://www.ncbi.nlm.nih.gov/pubmed
- **TOXNET:** 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 05/31/19.

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. We wish to confirm that there are no known conflicts of interest associated with this publication and there has been no significant financial support for this work that could have influenced its outcome. RIFM staff are employees of the Research Institute for Fragrance Materials, Inc. (RIFM). The Expert Panel receives a small honorarium for time spent reviewing the subject work.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fct.2020.111194.

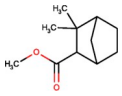
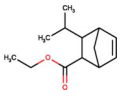
Appendix

Read-across Justification

Methods

The read-across analog was identified following the strategy for structuring and reporting a read-across prediction of toxicity as described in Schultz et al. (2015). The strategy is also 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, 2016).

- 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, and oncologic classification predictions were generated using OECD QSAR Toolbox v4.2 (OECD, 2018).
- ER binding and repeat dose categorization 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).

	Target Material	Read-across Material
Principal Name	Methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate	(2-endo,3-exo)-Ethyl 3-(1-methylethyl)bicyclo[2.2.1]hept-5-ene-2-carboxylate
CAS No.	52557-97-8	116044-44-1
Structure		
Similarity (Tanimoto Score)		0.52
Read-across Endpoint		• Repeated Dose Toxicity
Molecular Formula	$C_{11}H_{18}O_2$	$C_{13}H_{20}O_2$
Molecular Weight	182.26	208.30
Melting Point (°C, EPI Suite)	21.85	24.97
Boiling Point (°C, EPI Suite)	216.96	254.37
Vapor Pressure (Pa @ 25 °C, EPI Suite)	20.4	2.87
Log K_{OW} (KOWWIN v1.68 in EPI Suite)	3.40	4.13
Water Solubility (mg/L, @ 25 °C, WSKOW v1.42 in EPI Suite)	66.57	11.67
J_{max} (μg/cm^2/h, SAM)	23.50	19.09
Henry's Law (Pa·m^3/mol, Bond Method, EPI Suite)	3.33E+001	5.16E+001
Repeated Dose Toxicity		
Repeated Dose (HESS)	• Not categorized	• Not categorized
Metabolism		
Rat Liver S9 Metabolism Simulator and Structural Alerts for Metabolites (OECD QSAR Toolbox v4.2)	See Supplemental Data 1	See Supplemental Data 2

Summary

There are insufficient toxicity data on methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate (CAS # 52557-97-8). 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, (2-endo,3-exo)-Ethyl 3-(1-methylethyl)bicyclo[2.2.1]hept-5-ene-2-carboxylate (CAS # 116044-44-1) was identified as a read-across analog with sufficient data for toxicological evaluation.

Conclusions

- (2-endo,3-exo)-Ethyl 3-(1-methylethyl)bicyclo[2.2.1]hept-5-ene-2-carboxylate (CAS # 116044-44-1) was used as a read-across analog for the target material methyl 3,3-dimethylbicyclo[2.2.1]heptane-2-carboxylate (CAS # 52557-97-8) for the repeated dose toxicity endpoint.
  - o The target material and the read-across analog are structurally similar and belong to a class of bridged bicyclic alkyl esters.
  - o The target material and the read-across analog share a bridged bicyclic ester moiety.
  - o The key difference between the target material and the read-across analog is that the target material has a saturated bridged bicyclic acid branch while the read-across analog has an unsaturated vinylene bridged bicyclic acid branch. The target material has a dimethyl substitution in the bridged bicycle moiety while the read-across analog has an isopropyl group in the same position. The target material has a methanol branch while the read-across analog has an ethanol branch. This structural difference is toxicologically insignificant.
  - o 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 physical–chemical properties of the target material and the read-across analog are sufficiently similar to enable comparison of 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 Data are consistent with *in silico* alerts.
  - 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.

Explanation of Cramer Classification

Due to potential discrepancies between the current *in silico* tools (Bhatia et al., 2015), the Cramer Class of the target material was determined using expert judgment, based on the Cramer decision tree.

Q1. Normal constituent of the body? No
Q2. Contains functional groups associated with enhanced toxicity? No
Q3. Contains elements other than C, H, O, N, and divalent S? No
Q5. Simply branched aliphatic hydrocarbon or a common carbohydrate? No
Q6. Benzene derivative with certain substituents? No
Q7. Heterocyclic? No
Q16. Common terpene? (see Cramer et al., 1978 for detailed explanation)? No
Q17. Readily hydrolyzed to a common terpene? No A terpene ester, but no reference found for natural occurrence. So N to 17
Q19. Open chain? No

Q23. Aromatic? No
Q24. Monocarbocyclic with simple substituents? No
Q25. Cyclopropane (see explanation in Cramer et al., 1978)? No
Q26. Monocycloalkanone or a bicyclo compound? Yes, Class II (Class moderate)

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