RIFM fragrance ingredient safety assessment, nopol, CAS Registry Number 128-50-7


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

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Version: 111721. Initial publication. All fragrance materials are evaluated on a five-year rotating basis. Revised safety assessments are published if new relevant data become available. Open access to all RIFM Fragrance Ingredient Safety Assessments is here: fragrancematerialssafetyresource.elsevier.com.

(continued on next page)
LOEL
IFRA
GLP
Nopol was evaluated for genotoxicity, repeated dose toxicity, reproductive toxicity, local respiratory toxicity, phototoxicity/photoallergenicity, skin sensitization, and environmental
Summary: The existing information supports the use of this material as described in this safety assessment.

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

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; Safford et al., 2015a, 2017; Comiskey et al., 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 Observed 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 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, Authorization, and Restriction of Chemicals

RD - 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., 2015a), 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.

Nopol was evaluated for genotoxicity, repeated dose toxicity, reproductive toxicity, and skin sensitization, and environmental safety. Data from read-across analog myrtenol (CAS # 515-00-4) provide nopol a No Expected Sensitization Induction Level (NESIL) of 3800 μg/cm² for the skin sensitization endpoint. The phototoxicity/photoallergenicity endpoints were evaluated based on ultraviolet/visible (UV/Vis) spectra; nopol is not expected to be phototoxic/photoallergenic. The local respiratory toxicity endpoint was completed using the Threshold of Toxicological Concern (TTC) for a Cramer Class III material; exposure is below the TTC (0.47 mg/day). The environmental endpoints were evaluated; nopol 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 its current volume of use in Europe and North America (i.e., Predicted Environmental Concentration/Predicted No Effect Concentration (PEC/PNEC)), are <1.

Human Health Safety Assessment

Genotoxicity: Not expected to be genotoxic. (RIFM, 2015c; RIFM, 2015b)
Repeated Dose Toxicity: NOAEL ~ 60 mg/kg/day.
Reproductive Toxicity: NOAEL = 478.5 mg/kg/day.

Skin Sensitization: NESIL = 3800 µg/cm².

Phototoxicity/Photoallergenicity: Not expected to be phototoxic/photallergenic.

Local Respiratory Toxicity: No NOAEC available. Exposure is below the TTC.

Environmental Safety Assessment

Hazard Assessment:

Persistence: Critical Measured Value: 5% (OECD 301D) for CAS # 35836-73-8

Bioaccumulation: Screening-level: 69.3 L/kg

Ecotoxicity: Screening-level: Fish LC50: 16.92 mg/L

Conclusion: Not PBT or vPvB as per IFRA Environmental Standards

Risk Assessment:

Screening-level: PEC/PNEC (North America and Europe) < 1

Critical Ecotoxicity Endpoint: Fish LC50: 16.92 mg/L

RIFM PNEC is: 0.01662 µg/L
* Revised PEC/PNECs (2015 IFRA VolU): North America and Europe: Not applicable; cleared at screening-level

1. Identification

Chemical Name: Nopol
CAS Registry Number: 128-50-7
Molecular Formula: C₁₃H₂₆O
Molecular Weight: 166.26
RIFM Number: 9178
Stereochemistry: Two stereocenters and 4 possible stereoisomers.

Chemical Name: (1R)-Nopol
CAS Registry Number: 35836-73-8
Synonyms: (1R)-6,6-dimethylbicyclo[3.1.1]hept-2-ene-2-ethanol
Molecular Formula: C₁₃H₂₆O
Molecular Weight: 166.26
RIFM Number: 7366
Stereochemistry: Two stereocenters and 4 possible stereoisomers.

2. Physical data

1. Boiling Point: 230 °C (Fragrance Materials Association [FMA]), 250.35 °C (EPI Suite)
2. Flash Point: >93 °C (Global Harmonized System [GHS]), >200 °F; CC (FMA)
3. Log Kow: 3.29 (EPI Suite)
4. Melting Point: 49.6 °C (EPI Suite)
5. Water Solubility: 318.1 mg/L (EPI Suite)
6. Specific Gravity: 0.973 (FMA)
7. Vapor Pressure: 0.002 mm Hg at 20 °C (FMA), 0.00252 mm Hg at 20 °C (EPI Suite v4.0), 0.00474 mm Hg at 25 °C (EPI Suite)
8. UV Spectra: No absorbance between 290 and 700 nm; molar absorption coefficient is below the benchmark (1000 L mol⁻¹ cm⁻³)
9. Appearance/Organoleptic: Colorless, slightly viscous liquid with a mild woody camphoraceous odor

3. Volume of use (Worldwide band)

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

4. Exposure to fragrance ingredient (Creme RIFM aggregate exposure model v3.1.4)*

1. 95th Percentile Concentration in Fine Fragrance: 0.20% (RIFM, 2020b)
2. Inhalation Exposure**: 0.00080 mg/kg/day or 0.050 mg/day (RIFM, 2020b)
3. Total Systemic Exposure**: 0.010 mg/kg/day (RIFM, 2020b)

*When a safety assessment includes multiple materials, the highest exposure out of all included materials will be recorded here for the 95th Percentile Concentration in fine fragrance, inhalation exposure, and total exposure.

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

3800 µg/cm²

1. Analogs Selected: (ECHA REACH Dossier: Bicyclo[3.1.1]hept-2-ene-2-ethanol, 6,6-dimethyl-, 2-acetate, (1R,SS); ECHA, 2013b)
2. Analogs Selected: (ECHA REACH Dossier: Bicyclo[3.1.1]hept-2-ene-2-ethanol, 6,6-dimethyl-, 2-acetate, (1R,SS); ECHA, 2013b)
3. Analogs Selected: RIFM (2005c) (UV/Vis Spectra; RIFM Database)
4. Analogs Selected: RIFM (Framework; Salvito et al., 2002)
5. Analogs Selected: RIFM (Framework; Salvito et al., 2002)
6. Analogs Selected: RIFM (Framework; Salvito et al., 2002)

5. Derivation of systemic absorption

1. Dermal: 80%
2. Oral: Assumed 100%
3. Inhalation: Assumed 100%

6. Computational toxicology evaluation

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

**See the Appendix below for further details.

1. Analogs Selected:
a. Genotoxicity: Myrtenol (CAS # 515-00-4)
b. Repeated Dose Toxicity: (1R)-Nopyl acetate (CAS # 35836-72-7)
c. Reproductive Toxicity: (1R)-Nopyl acetate (CAS # 35836-72-7)
d. Skin Sensitization: 2,4,6-Trimethyl-3-cyclohexene-1-methanol (CAS # 68527-77-5)
e. Phototoxicity/Photoallergenicity: None
f. Local Respiratory Toxicity: None
g. Environmental Toxicity: None
2. Read-across Justification: See Appendix below

7. Metabolism

Not considered for this risk assessment and therefore not reviewed except where it may pertain in specific endpoint sections as discussed below.

7.1. Additional References

None.

8. Natural occurrence

Nopol is not reported to occur in foods by the VCF.a


9. REACH dossier

Nopol has been pre-registered for 11/30/10; no dossier available as of 12/23/20. Available for additional material (1R)-nopol (CAS # 35836-73-8); accessed 02/23/21 (ECHA, 2013a).

10. Conclusion

The maximum acceptable concentrationsb in finished products for nopol are detailed below.

<table>
<thead>
<tr>
<th>IFRA Categoryb</th>
<th>Description of Product Type</th>
<th>Maximum Acceptable Concentrationsc in Finished Products (%)d</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Products applied to the lips (lipstick)</td>
<td>0.29</td>
</tr>
<tr>
<td>2</td>
<td>Products applied to the axillae</td>
<td>0.087</td>
</tr>
<tr>
<td>3</td>
<td>Products applied to the face/body using fingertips</td>
<td>0.76</td>
</tr>
<tr>
<td>4</td>
<td>Products related to fine fragrances</td>
<td>1.6</td>
</tr>
<tr>
<td>5A</td>
<td>Body lotion products applied to the face and body using the hands (palms), primarily leave-on</td>
<td>0.41</td>
</tr>
<tr>
<td>5B</td>
<td>Face moisturizer products applied to the face and body using the hands (palms), primarily leave-on</td>
<td>0.41</td>
</tr>
<tr>
<td>5C</td>
<td>Hand cream products applied to the face and body using the hands (palms), primarily leave-on</td>
<td>0.41</td>
</tr>
<tr>
<td>5D</td>
<td>Baby cream, oil, talc</td>
<td>0.14</td>
</tr>
<tr>
<td>6</td>
<td>Products with oral and lip exposure</td>
<td>0.96</td>
</tr>
<tr>
<td>7</td>
<td>Products applied to the hair with some hand contact</td>
<td>0.58</td>
</tr>
<tr>
<td>8</td>
<td>Products with significant anogenital exposure (tampon)</td>
<td>0.14</td>
</tr>
<tr>
<td>9</td>
<td>Products with body and hand exposure, primarily rinse-off (bar soap)</td>
<td>1.9</td>
</tr>
<tr>
<td>10A</td>
<td></td>
<td>2.7</td>
</tr>
</tbody>
</table>
(continued on next column)

Note: *Maximum acceptable concentrations for each product category are based on the lowest acceptable concentrations considered (based on systemic toxicity, skin sensitization, or any other endpoint evaluated in this safety assessment). For nopol, the basis was the reference dose of 0.60 mg/kg/day, a predicted skin absorption value of 80%, and a skin sensitization NESIL of 3800 μg/cm².


cCalculations by Creme RIFM Aggregate Exposure Model v3.1.4.

11. Summary

11.1. Human health endpoint summaries

11.1.1. Genotoxicity

Based on the current existing data, nopol does not present a concern for genotoxicity.

11.1.1. Risk assessment. Nopol was assessed in the BlueScreen assay and found negative for both cytotoxicity and genotoxicity, with and without metabolic activation (RIFM, 2013). BlueScreen is a human cell-based assay for measuring the genotoxicity and cytotoxicity of chemical compounds and mixtures. Additional assays on a more reactive read-across material were considered to fully assess the potential mutagenic or clastogenic effects of the target material.

There are no studies assessing the mutagenic activity of nopol; however, read-across can be made to myrtenol (CAS # 515-00-4; see Section VI). The mutagenic activity of myrtenol has been evaluated in a bacterial reverse mutation assay conducted in compliance with GLP regulations and in accordance with OECD TG471 using the standard plate incorporation method. Salmonella typhimurium strains TA98, TA100, TA1535, TA1537, and Escherichia coli strain WP2uvrA were treated with myrtenol in solvent dimethyl sulfoxide (DMSO) at concentrations up to 5000 μg/plate. Increases in the mean number of revertant colonies were not observed at any tested dose in the presence or absence of metabolic activation (S9) (RIFM, 2015c). Under the conditions of the study, myrtenol was not mutagenic in the Ames test, and this can be extended to nopol.

There are no studies assessing the clastogenic activity of nopol; however, read-across can be made to myrtenol (CAS # 515-00-4; see Section VI). The clastogenic activity of myrtenol 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 myrtenol in solvent DMSO at concentrations up to 500 μg/mL in the presence and absence of S9 for 3 and 24 h. Myrtenol did not induce binucleated cells with micronuclei when tested up to cytotoxic levels in either non-activated or S9-activated test systems (RIFM, 2015b). Under the conditions of the study, myrtenol was considered to be non-clastogenic in the in vitro micronucleus test, and this can be extended to nopol.

Based on the data available, myrtenol does not present a concern for genotoxic potential, and this can be extended to nopol.

Additional References: None.
A.M. Api et al.

11.1.2. Repeated dose toxicity

The MOE for nopol is adequate for the repeated dose toxicity endpoint at the current level of use.

11.1.2.1. Risk assessment. There are no repeated dose toxicity data on nopol. Read-across material (1R)-nopyl acetate (CAS # 35836-72-7; see Section VI) has sufficient repeated dose toxicity data. In an OECD/GLP 422 study, 3 groups of Sprague Dawley CrI:CD BR rats were administered via dietary admixture of test material, nopol acetate at concentrations of 0, 1000, 3000, or 9000 ppm (equivalent to doses of 0, 56.5, 180.2, or 478.5 mg/kg/day) for up to 63 days (including 3 weeks of maturation phase, pairing, gestation, and early lactation for females). The dose range was determined in a preliminary dose range finding study. Groups of 3 Sprague Dawley CrI:CD BR rats were administered daily via dietary admixture at concentrations of 0, 1500, 7500, or 15000 ppm (equivalent to 0, 106, 490, or 952 mg/kg/day, respectively) nopol acetate for 21 days. The dose range was determined based on reductions in bodyweight gains in the 7500 ppm and above treatment groups and changes in clinical chemistry parameters in 7500 ppm treated males and in 1500 ppm treated females. In the OECD 422 study, each dose group was subdivided into 2 phases: main phase (at 1000 and 3000 ppm: 10 rats/sex/dose; at 0 and 9000 ppm: 5 males and 10 females/sex/dose) and toxicity phase (5 female/sex/dose). A control group was treated with a basal laboratory diet with 2% corn oil. Two recovery groups (5 rats/sex/dose) were treated with 9000 ppm or basal laboratory diet alone for 42 consecutive days and then maintained without treatment for a further 14 days. The clinical condition of offspring, litter size and survival, sex ratio, and offspring body weight were assessed, and macroscopic pathology evaluations were conducted. No treatment-related significant effects were observed on offspring litter size, sex ratio, viability, growth, and development. Thus, the NOAEL for developmental toxicity was considered to be 9000 ppm or 478.5 mg/kg/day, the highest dose tested (ECHA, 2013b). Therefore, the nopol MOE for the developmental toxicity endpoint can be calculated by dividing the (1R)-nopyl acetate NOAEL in mg/kg/day by the total systemic exposure to nopol, 478.5/0.010, or 47850.

There are no fertility data on nopol. Read-across material, (1R)-nopyl acetate (CAS # 35836-72-7; see Section VI) has sufficient fertility data. In an OECD/GLP 422 study, 3 groups of Sprague Dawley CrI:CD BR rats were administered via dietary admixture of test material, nopol acetate at concentrations of 0, 1000, 3000, or 9000 ppm (equivalent to doses of 0, 56.5, 180.2, or 478.5 mg/kg/day) for up to 63 days (including 3 weeks of maturation phase, pairing, gestation, and early lactation for females). Each dose group was subdivided into 2 phases: main phase (at 1000 and 3000 ppm: 10 rats/sex/dose; at 0 and 9000 ppm: 5 males and 10 females/sex/dose) and toxicity phase (5 female/sex/dose). A control group was treated with a basal laboratory diet with 2% corn oil. Two recovery groups (5 rats/sex/dose) were treated with 9000 ppm or basal laboratory diet alone for 42 consecutive days and then maintained without treatment for a further 14 days. No treatment-related effects in mating performance, fertility, and gestation lengths were observed up to the highest dose tested. Thus, the NOAEL for fertility was considered to be

<table>
<thead>
<tr>
<th>Potency Classification Based on Animal Data&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Human Data</th>
<th>LLNA Weighted Mean EC&lt;sub&gt;3&lt;/sub&gt; Value&lt;sup&gt;c&lt;/sup&gt;</th>
<th>Mean EC&lt;sub&gt;3&lt;/sub&gt; Value&lt;sup&gt;c&lt;/sup&gt;</th>
<th>[μg/cm&lt;sup&gt;2&lt;/sup&gt;]</th>
</tr>
</thead>
<tbody>
<tr>
<td>NOEL-CNIIH (induction) µg/cm&lt;sup&gt;2&lt;/sup&gt;</td>
<td>NOEL-HMT (induction) µg/cm&lt;sup&gt;2&lt;/sup&gt;</td>
<td>LOEL&lt;sup&gt;b&lt;/sup&gt; (induction) µg/cm&lt;sup&gt;2&lt;/sup&gt;</td>
<td>WoE NESIL&lt;sup&gt;d&lt;/sup&gt; µg/cm&lt;sup&gt;2&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>&gt;6250&lt;sup&gt;a&lt;/sup&gt; [1]</td>
<td>Weak</td>
<td>3897</td>
<td>NA</td>
<td>5000</td>
</tr>
</tbody>
</table>

NOEL = No observed effect level; CNIIH = Confirmation of No Induction in Humans test; HMT = Human Maximization Test; LOEL = lowest observed effect level; NA = Not Available.

<sup>a</sup> Based on animal data using classification defined in ECETOC, Technical Report No. 87, 2003.

<sup>b</sup> Data derived from CNIIH or HMT.

<sup>c</sup> WoE NESIL limited to 2 significant figures.
9000 ppm or 478.5 mg/kg/day (ECHA, 2013b). Therefore, the nopol MOE for the fertility endpoint can be calculated by dividing the (1R)-nonyl acetate NOAEL in mg/kg/day by the total systemic exposure to nopol, 478.5/0.010, or 47850.

Additional References: None.

Literature Search and Risk Assessment Completed On: 02/08/21.

11.1.4. Skin sensitization

Based on the existing data and read-across to 2,4,6-trimethyl-3-cyclohexene-1-methanol (CAS # 68527-77-5), nopol is considered a skin sensitizer with a defined NESIL of 3800 μg/cm².

11.1.4.1. Risk assessment. Limited skin sensitization studies are available on nopol. Based on the existing data and read-across to 2,4,6-trimethyl-3-cyclohexene-1-methanol (CAS # 68527-77-5; see Section VI), nopol is considered a skin sensitizer. The chemical structure of these materials indicates that they would not be expected to react with skin proteins directly (Roberts et al., 2007; Toxtree v3.1.0; OECD Toolbox v4.2). 2,4,6-trimethyl-3-cyclohexene-1-methanol was found to be negative in a vitro Direct Peptide Reactivity Assay (DPRA) and KeratinôSens, but positive in the h-CLAT (RIFM, 2014; RIFM, 2015d; RIFM, 2016). In a murine local lymph node assay (LLNA), nopol (CAS # 35836-73-8) was found to be sensitizing with an EC3 of 6000 μg/cm² (ECHA, 2013a). In another LLNA, the read-across material, 2,4,6-trimethyl-3-cyclohexene-1-methanol, was found to be non-sensitizing up to 25% (6250 μg/cm²) (RIFM, 2005b). In a Buehler study, 60% 2,4,6-trimethyl-3-cyclohexene-1-methanol presented 7/10 reactions indicative of sensitization (RIFM, 1981). In a human maximization test, no sensitization reactions were observed when 8% or 5520 (µg/cm²) in petrolatum was used for induction and challenge (RIFM, 1976). In a Confirmation of No Induction in Humans test (CNH) with 3897 (µg/cm²) of the read-across material, 2,4,6-trimethyl-3-cyclohexene-1-methanol in 1:3 EtOH:DEP, no reactions indicative of sensitization was observed in any of the 103 volunteers (RIFM, 2005c). In additional CNHs with less than 100 subjects, 2,4,6-trimethyl-3-cyclohexene-1-methanol presented 7/10 reactions indicative of sensitization (RIFM, 1983).

Based on the available data on read-across 2,4,6-trimethyl-3-cyclohexene-1-methanol presented 7/10 reactions indicative of sensitization (RIFM, 1983). In a human maximization test, no sensitization reactions were observed when 8% or 5520 (µg/cm²) in petrolatum was used for induction and challenge (RIFM, 1976). In a Confirmation of No Induction in Humans test (CNH) with 3897 (µg/cm²) of the read-across material, 2,4,6-trimethyl-3-cyclohexene-1-methanol in 1:3 EtOH:DEP, no reactions indicative of sensitization was observed in any of the 103 volunteers (RIFM, 2005c). In additional CNHs with less than 100 subjects, 2,4,6-trimethyl-3-cyclohexene-1-methanol presented 7/10 reactions indicative of sensitization (RIFM, 1983).

Based on the available data on read-across 2,4,6-trimethyl-3-cyclohexene-1-methanol summarized in Table 1 below, nopol is considered to be a weak skin sensitizer with a defined NESIL of 3800 μg/cm².

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, 2020c) and a reference dose of 0.60 mg/kg/day.

Additional References: RIFM, 2005a; ICCVAM, 2011; RIFM, 2009; Api et al., 2015b; Safford et al., 2015b.

Literature Search and Risk Assessment Completed On: 01/20/21.

11.1.5. Phototoxicity/photoallergenicity

Based on the available UV/Vis spectra, nopol 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 nopol in experimental models. UV/Vis absorption spectra indicate no absorption between 290 and 700 nm. The corresponding 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.


11.1.6. Local respiratory toxicity

The MOE could not be calculated due to a lack of appropriate data. The exposure level for nopol is below the Cramer Class III* TTC value for inhalation exposure local effects.

11.1.6.1. Risk assessment. There are no inhalation data available on nopol. Based on the Cramer RIFM Model, the inhalation exposure is 0.050 mg/day. This exposure is 9.4 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: 02/12/21.

11.2. Environmental endpoint summary

11.2.1. Screening-level assessment

A screening-level risk assessment of nopol was performed following the RIFM Environmental Framework (Salvito et al., 2002), which provides 3 tiers of levels of screening for aquatic risk. In Tier 1, only the material’s regional VoU, its log KOW, 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 ecoxicity 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, nopol was identified as a fragrance material with no 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 v4.11 (US EPA, 2012b) identified nopol as possibly persistent but not 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., 2015a). 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 v4.11).

11.2.2. Risk assessment

Based on the current Volume of Use (2015), nopol does not present a risk to the aquatic compartment in the screening-level assessment.

11.2.2.1. Key studies

11.2.2.1.1. Biodegradation. No data available.

11.2.2.1.2. Ecotoxicity. No data available.

11.2.2.1.3. Other available data. Additional material (1R)-nopol (CAS # 35836-73-8) has been registered for REACH, with the following additional data available at this time (ECHA, 2013a):

A ready biodegradability of the test material was evaluated using the closed bottle test according to the OECD 301D guideline. Biodegradation of 5% was observed after 28 days.

An acute fish (Danio rerio) toxicity test was conducted according to the OECD 203 guideline under semi-static conditions. The 96-h LC50 value based on nominal test concentration was reported to be 21.14 mg/L (95% CI: 19.3 – 22.3 mg/L).

A Daphnia magna acute immobilization test was conducted according to the OECD 202 guideline under static conditions. The 48-h EC50 value based on nominal test concentration was reported to be 28.8 mg/L (95% CI: 22.3 – 38.2 mg/L).

A Daphnia magna reproduction test was conducted according to the OECD 211 guideline under semi-static conditions. The 21-day NOEC value based on time-weighted average value was reported to be 0.4 mg/L.

An algae growth inhibition test was conducted according to the OECD 201 guideline under static conditions. The 72-h EC50 values based on measured concentration for growth rate and yield were reported to be 21.14 mg/L (95% CI: 19.3–23.1 mg/L) and 7.224 mg/L (95% CI: 5.9–8.8 mg/L), respectively.

11.2.2.2. Risk assessment refinement

Since nopol has passed the screening criteria, measured data is included for completeness only and has not been used in PNEC derivation.

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

Endpoints used to calculate PNEC are underlined.

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

<table>
<thead>
<tr>
<th>Exposure</th>
<th>Europe (EU)</th>
<th>North America (NA)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Log Kow Used</td>
<td>3.29</td>
<td>3.29</td>
</tr>
<tr>
<td>Biodegradation Factor Used</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Dilution Factor</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>Regional Volume of Use Tonnage Band*</td>
<td>1-10</td>
<td>&lt;1</td>
</tr>
<tr>
<td>Risk Characterization: PEC/PNEC</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
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</table>

*Combined Regional Volume of Use for both CAS #s.

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

The RIFM PNEC is 0.01692 μg/L. The revised PEC/PNECs for EU and NA are not applicable. The material was cleared at the screening-level; therefore, it does not present a risk to the aquatic environment at the current reported VoU.


11.3. 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/
- SciFinder: https://scifinder.cas.org/scifinder/view/scifinder/scifinderExplore.jsf
- National Library of Medicine’s Toxicology Information Services: https://toxnet.nlm.nih.gov/
- EPA ACToR: https://actor.epa.gov/actor/home.xhtml
- US EPA HPVIS: https://ofmpub.epa.gov/opthpv/public_search.publicdetails?submission_id=24959241&showComments=Yes&sqlstr=null&recordcount=0&User_title=DetailQuery%20Results&EndPointRpt=Y#submission
- Google: https://www.google.com

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 11/17/21.

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. 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

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</table>
Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fct.2022.112925.

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).
- \( J_{\text{max}} \) 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.

To keep continuity and compatibility with in silico alerts, OECD QSAR Toolbox v4.2 was selected as the alert system.

<table>
<thead>
<tr>
<th>Principal Name</th>
<th>Nopol</th>
<th>Myrtenol</th>
<th>2,4,6-Trimethyl-3-cyclohexene-1-methanol</th>
<th>(1R)-Nopyl acetate</th>
</tr>
</thead>
<tbody>
<tr>
<td>CAS No.</td>
<td>128-50-7</td>
<td>515-00-4</td>
<td>68527-77-5</td>
<td>35836-72-7</td>
</tr>
<tr>
<td>Molecular Formulas</td>
<td>C_{11}H_{18}O</td>
<td>C_{10}H_{18}O</td>
<td>C_{10}H_{18}O</td>
<td>C_{13}H_{20}O</td>
</tr>
<tr>
<td>Molecular Weight</td>
<td>166.27</td>
<td>152.24</td>
<td>154.25</td>
<td>208.3</td>
</tr>
<tr>
<td>Melting Point (°C, EPI Suite)</td>
<td>49.60</td>
<td>38.73</td>
<td>13.51</td>
<td>54.2</td>
</tr>
<tr>
<td>Boiling Point (°C, EPI Suite)</td>
<td>250.35</td>
<td>232.42</td>
<td>237.00</td>
<td>259.16</td>
</tr>
<tr>
<td>Log ( K_{\text{ow}} ) (KOWWIN v1.68 in EPI Suite)</td>
<td>3.29</td>
<td>3.22</td>
<td>3.30</td>
<td>4.30</td>
</tr>
<tr>
<td>Water Solubility (mg/L, @ 25°C, WSKOW v1.42 in EPI Suite)</td>
<td>318.1</td>
<td>426.9</td>
<td>360.2</td>
<td>8.429</td>
</tr>
<tr>
<td>( J_{\text{max}} ) (μg/cm²/h, SAM)</td>
<td>94.515</td>
<td>43.314</td>
<td>234.963</td>
<td>0.926</td>
</tr>
<tr>
<td>Henry’s Law (Pa m²/mol, Bond Method, EPI Suite)</td>
<td>9.36E-001</td>
<td>7.05E-001</td>
<td>1.60E+000</td>
<td>6.10E+001</td>
</tr>
</tbody>
</table>

**Genotoxicity**
- No alert found
- No alert found

**Carcinogenicity (Genotox and Non-genotox) Alerts**
- Non-carcinogen (low reliability)
- Non-carcinogen (low reliability)

**DNA Binding (OASIS V 1.4 QSAR Toolbox 4.2)**
- No alert found
- No alert found

**DNA Binding by OECD QSAR Toolbox (4.2)**
- No alert found
- No alert found

**In Vitro Mutagenicity (Ames Test) Alerts by ISS**
- No alert found
- No alert found

**In Vivo Mutagenicity (Micronucleus) Alerts by ISS**
- No alert found
- No alert found

**Oncologic Classification**
- Not classified
- Not classified

**Repeated Dose Toxicity**
- Not categorized

(continued on next page)
Metabolism identified as read-across materials with sufficient data for toxicological evaluation. analogs myrtenol (CAS # 515-00-4), 2,4,6-trimethyl-3-cyclohexene-1-methanol (CAS # 68527-77-5), and (1R)-nopyl acetate (CAS # 35836-72-7) were across analog for this material. Based on structural similarity, reactivity, metabolism data, physical

### Summary

There are insufficient toxicity data on the target material, nopol (CAS # 128-50-7). Hence in silico evaluation was conducted to determine a read-across analog for this material. Based on structural similarity, reactivity, metabolism data, physical–chemical properties, and expert judgment, analogs myrtenol (CAS # 515-00-4), 2,4,6-trimethyl-3-cyclohexene-1-methanol (CAS # 68527-77-5), and (1R)-nopyl acetate (CAS # 35836-72-7) were identified as read-across materials with sufficient data for toxicological evaluation.

### Metabolism

The metabolism of the target material was not evaluated in the risk assessment. Therefore, metabolism data were not reviewed, except when it may pertain in the read-across for specific endpoint sections above. Metabolism of the read-across material (1R)-nopyl acetate (CAS # 35836-72-7) was predicted using the rat liver S9 Metabolism Simulator (OECD QSAR Toolbox v4.2). Read-across analog (1R)-nopyl acetate (CAS # 35836-72-7) is expected to be metabolized to target material nopol (CAS # 128-50-7) and acetic acid in the first step with 0.95 pre-calculated probability. Hence (1R)-nopyl acetate can be used as a read-across for target material. The target material was out of domain for the in vivo rat and in vitro rat S9 simulator (OASIS TIMES v2.27.19). However, based on expert judgment, the model’s domain exclusion was overridden, and a justification is provided.

### Conclusion

- Myrtenol (CAS # 515-00-4) was used as a read-across analog for the target material nopol (CAS # 128-50-7) for the genotoxicity endpoint.
  - The target material and the read-across analog belong to the structural class of primary cyclic unsaturated aliphatic alcohol.
  - The key difference between the target material and the read-across analog is that the target material is a substituted ethanol, whereas the read-across analog is a substituted methanol. This structure difference between the target material and the read-across analog does not affect consideration of the toxicity endpoint.
  - The similarity between the target material and the read-across analog is indicated by the Tanimoto score in the above table. Differences between the structures that affect the Tanimoto score do not affect consideration of the toxicity endpoint.
  - The physical–chemical properties of the target material and the read-across analog are sufficiently similar to enable comparison of their toxicological properties.
  - According to the QSAR OECD Toolbox (v4.2), structural alerts for toxicity endpoints are consistent between the target material and the read-across analog.
  - The target material and the read-across analog are expected to be metabolized similarly, as shown by the metabolism simulator.
- 2,4,6-Trimethyl-3-cyclohexene-1-methanol (CAS # 68527-77-5) was used as a read-across analog for the target material nopol (CAS # 128-50-7) for skin sensitization.
  - The target material and the read-across analog belong to the structural class of primary cyclic unsaturated aliphatic alcohol.
  - The key difference between the target material and the read-across analog is that the target material has a bicyclic ring structure that is not found in the read-across analog 2,4,6-trimethyl-3-cyclohexene-1-methanol. This structure difference between the target material and the read-across analog does not affect consideration of the toxicity endpoint.
  - The similarity between the target material and the read-across analog is indicated by the Tanimoto score in the above table. The Tanimoto score is 0.3 for 2,4,6-trimethyl-3-cyclohexene-1-methanol (CAS # 68527-77-5). This is due to the lack of bridge ring structure in the read-across analog as compared to the target material. Differences between the structures that affect the Tanimoto score do not affect consideration of the toxicity endpoint.
  - The physical–chemical properties of the target material and the read-across analog are sufficiently similar to enable comparison of their toxicological properties.
  - According to the QSAR OECD Toolbox (v4.2), structural alerts for toxicity endpoints are consistent between the target material and the read-across analog.
• The target material and the read-across analog material are predicted to be sensitizers by the CAESAR model for skin sensitization. There are no other protein binding alerts for both of the substances. The data described in the skin sensitization section shows that the read-across analog does not pose a concern for the skin sensitization endpoint. Therefore, this alert will be superseded by the availability of the data.
• The target material and the read-across analog are expected to be metabolized similarly, as shown by the metabolism simulator.
• (1R)-nopyl acetate (CAS # 35836-72-7) was used as a read-across analog for the reproductive toxicity and repeated dose toxicity endpoints.
• The read-across analog, (1R)-nopyl acetate (CAS # 35836-72-7), is an ester formed from the target material nopol (CAS # 128-50-7).
• Structural differences between the target material and the read-across analog are mitigated by the fact that read-across analog (1R)-nopyl acetate (CAS # 35836-72-7) could be metabolically hydrolyzed to the target material nopol (CAS # 128-50-7). Therefore, the toxicity profile of the target is expected to be that of metabolites.
• The differences between the physical–chemical properties of the target material and the read-across analog can be mitigated by the fact that the target material is a direct metabolite of the read-across analog.
• According to the QSAR OECD Toolbox (v4.2), structural alerts for toxic endpoints are consistent between the target material and the read-across analog.
• The target material and the read-across analogs are predicted to be toxicants by the developmental toxicity model by CAESAR. There are no other reproductive toxicity alerts. The data described shows that the read-across analog does not pose a concern for the reproductive endpoint. Therefore, this alert will be superseded by the availability of the data. The structural alerts for toxic endpoints are consistent between the metabolites of the read-across analogs 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. A 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
Q19. Open chain? No
Q23. Aromatic? No
Q24. Monocarboxyclic with simple substituents? No
Q25. Cyclopropane, cyclobutane with substituents in Q24 or mono or bicyclic sulfide or mercaptan? Yes, Class III (Class High)

References


RIFM (Research Institute for Fragrance Materials, Inc), 2005b. 2,4,6-Trimethyl-3-cyclohexene-1-methanol (isocyclogeraniol) Diluted with Vehicle 1:3 EtOH:DEP: Local Lymph Node Assay. RIFM report number 48795. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc), 2005c. Repeated Insult Patch Test with 2,4,6-Trimethyl-3-Cyclohexene-1-Methanol. RIFM report number 49110. RIFM, Woodcliff Lake, NJ, USA.


RIFM (Research Institute for Fragrance Materials, Inc), 2015a. Novel Database for Exposure to Fragrance Ingredients in Cosmetics and Personal Care Products. RIFM report number 68681. RIFM, Woodcliff Lake, NJ, USA.


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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.


