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Short Review

RIFM fragrance ingredient safety assessment, acetanisole, CAS Registry Number 100-06-1

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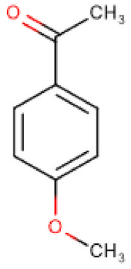
Version: 060822. Initial publication. All fragrance materials are evaluated on a five-year rotating basis. Revised safety assessments are published if new relevant data become available. Open access to all RIFM Fragrance Ingredient Safety Assessments is here: fragrancematerialsafetyresource.elsevier.com.

Name: Acetanisole

CAS Registry Number: 100-06-1



Abbreviation/Definition List:

2-Box Model - A RIFM, Inc. proprietary *in silico* tool used to calculate fragrance air exposure concentration
AF - Assessment Factor

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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; Safford et al., 2015a; Safford et al., 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

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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 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 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. This material has not been fully evaluated for photoallergenic potential.

Acetanisole was evaluated for genotoxicity, repeated dose toxicity, reproductive toxicity, local respiratory toxicity, photoirritation/photoallergenicity, skin sensitization, and environmental safety. Target data and data from read-across analog 4-(*p*-methoxyphenyl)-2-butanone (CAS # 104-20-1) show that acetanisole 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 acetanisole is below the TTC (0.03 mg/kg/day, 0.03 mg/kg/day, and 1.4 mg/day, respectively). Data show that there are no safety concerns for acetanisole for skin sensitization under the current declared levels of use. The photoirritation endpoint was evaluated based on data; acetanisole is not expected to be photoirritating. Acetanisole has not been fully evaluated for photoallergenic potential. The environmental endpoints were evaluated; acetanisole 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, 2017a; RIFM, 2016)

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Repeated Dose Toxicity: No NOAEL available. Exposure is below TTC.
Reproductive Toxicity: No NOAEL available. Exposure is below the TTC.
Skin Sensitization: No concern for skin sensitization. (Ryan et al., 2000)
Photoirritation/Photoallergenicity: Not expected to be photoirritating. Photoallergy has not been evaluated. (RIFM, 2015)
Local Respiratory Toxicity: No NOAEC available. Exposure is below the TTC.

Environmental Safety Assessment

Hazard Assessment:
Persistence:
Critical Measured Value: 100% (EU Method C.4-B) (RIFM, 1992)
Bioaccumulation:
Screening-level: 1.698 L/kg (EPI Suite v4.11; US EPA, 2012a)
Ecotoxicity:
Screening-level: LC50: 308.1 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: LC50: 308.1 mg/L (RIFM Framework; Salvito et al, 2002)

RIFM PNEC is: 0.3081 μg/L
- **Revised PEC/PNECs (2019 IFRA VoU):** North America and Europe: Not applicable; cleared at screening-level

1. Identification

1. **Chemical Name:** Acetanisole
2. **CAS Registry Number:** 100-06-1
3. **Synonyms:** 4-Acetylanisole; p-Acetylanisole; Ethanone, 1-(4-methoxyphenyl)-; p-Methoxyacetophenone; 4′-Methoxyacetophenone; Methyl 4-methoxyphenyl ketone; ﾒﾄｷｼｱｾﾄﾌｪﾉﾝ; 1-(4-Methoxyphenyl)ethanone; Acetanisole Cryst.; Acetanisole
4. **Molecular Formula:** $C_9H_{10}O_2$
5. **Molecular Weight:** 150.17 g/mol
6. **RIFM Number:** 434
7. **Stereochemistry:** Stereoisomer not specified. No stereocenter present and no stereoisomer possible.

2. Physical data

1. **Boiling Point:** 229.45 °C (EPI Suite)
2. **Flash Point:** >100 °C (Globally Harmonized System), >212 °F; closed cup (Fragrance Materials Association [FMA])
3. **Log K_{OW}:** 1.75 (EPI Suite), 1.79±0.00 at 20 °C (RIFM, 2017d)
4. **Melting Point**: 24.72 °C (EPI Suite)
5. **Water Solubility:** 2474 mg/L (EPI Suite), 1737 ± 76 mg/L at 20 °C (RIFM, 2017c)
6. **Specific Gravity:** Not Available
7. **Vapor Pressure:** 0.00757 mm Hg at 20 °C (EPI Suite v4.0); 0.003 mm Hg at 20 °C (FMA); 0.0133 mm Hg at 25 °C (EPI Suite); 0.42, 0.71, and 7.9 Pa at 20, 25, and 50 °C, respectively (RIFM, 2017b)
8. **UV Spectra:** Significant absorbance between 290 and 700 nm, with a peak at 290 nm and returning to baseline by approximately 330 nm. Molar absorption coefficients (10443, 4817, and 11869 L mol^{-1} • cm^{-1} under neutral, acidic, and basic conditions, respectively) are above the benchmark (1000 L mol^{-1} • cm^{-1})
9. **Appearance/Organoleptic:** Colorless to pale yellow fused solid with odor suggestive of hawthorn and floral note of heliotrope

3. Volume of use (Worldwide band)

1.1–100 metric tons per year (IFRA, 2019)

4. Exposure to fragrance ingredient (Creme RIFM aggregate exposure model v2.0)

1. **95**th **Percentile Concentration in Fine Fragrance:** 0.025% (RIFM, 2019)
2. **Inhalation Exposure*:** 0.00013 mg/kg/day or 0.0091 mg/day (RIFM, 2019)
3. **Total Systemic Exposure**:** 0.0022 mg/kg/day (RIFM, 2019)

*95th percentile calculated exposure derived from concentration survey data in the Creme RIFM Aggregate Exposure Model (Comiskey et al, 2015; Safford et al., 2015; Safford et al, 2017; Comiskey et al, 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 et al, 2015; Safford et al., 2015; Safford et al, 2017; Comiskey et al, 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:** 4-(*p*-Methoxyphenyl)-2-butanone (CAS # 104-20-1)
   b. **Repeated Dose Toxicity:** None
   c. **Reproductive Toxicity:** None
   d. **Skin Sensitization:** None
   e. **Photoirritation/Photoallergenicity:** None
   f. **Local Respiratory Toxicity:** None
   g. **Environmental Toxicity:** None
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

Acetanisole is reported to occur in the following foods by the VCF*:
Anise (*Pimpinella anisum* L.)
Beef.
Black chokeberry (*Aronia melanocarpa* Ell.)
Citrus fruits.
Grape (*Vitis* species)
Honey.
Mentha oils Plum (*Prunus* species)
Sherry.
Star anise.
Tomato (*Lycopersicon esculentum* Mill.)

*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. This is a partial list.

9. Reach Dossier

Available; accessed 12/01/21.

10. Conclusion

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

11. Summary

11.1. Human health endpoint summaries

11.1.1. Genotoxicity

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

11.1.1.1. Risk assessment. The mutagenic activity of acetanisole 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 method. *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537, and *Escherichia coli* strain WP2uvrA were treated with acetanisole in dimethyl sulfoxide (DMSO) 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, 2017a). Under the conditions of the study, acetanisole was not mutagenic in the Ames test.

There are no studies assessing the clastogenic activity of acetanisole; however, read-across can be made to 4-(*p*-methoxyphenyl)-2-butanone (CAS # 104-20-1; see Section VI).

The clastogenic activity of 4-(*p*-methoxyphenyl)-2-butanone 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 4-(*p*-methoxyphenyl)-2-butanone in DMSO; micronuclei analysis was conducted at concentrations up to 1782 μg/mL in the presence and absence of metabolic activation. 4-(*p*-Methoxyphenyl)-2-butanone did induce binucleated cells with micronuclei when tested at 333 μg/mL in the 4-h treatment in the presence of an S9 activation system (RIFM, 2016). However, the binucleated cells with micronuclei at these concentrations were within the vehicle historical control ranges. Therefore, the statistically significant increases at these concentrations were considered biologically non-relevant and not indicative of clastogenic effects. Under the conditions of the study, 4-(*p*-methoxyphenyl)-2-butanone was considered to be non-clastogenic in the *in vitro* micronucleus test, and this can be extended to acetanisole.

Based on the data available, 4-(*p*-methoxyphenyl)-2-butanone does not present a concern for genotoxic potential, and this can be extended to acetanisole.

Additional References: None.

Literature Search and Risk Assessment Completed On: 11/24/21.

11.1.2. Repeated dose toxicity

There are insufficient repeated dose toxicity data on acetanisole or any read-across materials. The total systemic exposure to acetanisole 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 acetanisole or any read-across materials that can be used to support the repeated dose toxicity endpoint. The total systemic exposure (2.2 μg/kg/day) is below the TTC for acetanisole (30 μg/kg/day; Kroes et al., 2007).

Additional References: None.

Literature Search and Risk Assessment Completed On: 11/01/21.

11.1.3. Reproductive toxicity

There are no reproductive toxicity data on acetanisole or any read-across materials. The total systemic exposure to acetanisole 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 reproductive toxicity data on acetanisole or any read-across materials that can be used to support the reproductive toxicity endpoint. The total systemic exposure (2.2 μg/kg/day) is below the TTC for acetanisole (30 μg/kg/day; Kroes et al., 2007; Laufersweiler et al., 2012).

Additional References: None.

Literature Search and Risk Assessment Completed On: 11/01/21.

11.1.4. Skin sensitization

Based on the existing data, acetanisole does not present a concern for skin sensitization.

11.1.4.1. Risk assessment. Based on the existing data, acetanisole is not considered a skin sensitizer. The data are summarized in Table 1. The chemical structure of this material indicates that it would not be expected to react with skin proteins directly (Roberts et al., 2007; Toxtree v3.1.0; OECD Toolbox v4.2). Acetanisole was found to be negative in an *in vitro* direct peptide reactivity assay (DPRA), positive in KeratinoSens, negative in the human cell line activation test (h-CLAT), and positive in the U-SENS test (Natsch et al., 2013; Nukada et al., 2011). In a murine local lymph node assay (LLNA), acetanisole was found to be non-sensitizing when tested up to 50% (12500 μg/cm^2) (Ryan et al., 2000). In a human maximization test, no sensitization reactions were observed at 4140 μg/cm^2 acetanisole (RIFM, 1973).

Based on the weight of evidence (WoE) from structural analysis and *in vitro*, animal, and human studies, acetanisole does not present a concern for skin sensitization.

Additional References: Basketter et al., 2002; Basketter et al., 2003; McKim et al., 2010; Natsch and Gfeller, 2008; Klecak (1985); RIFM, 1970; Klecak (1979); Sharp (1978).

Literature Search and Risk Assessment Completed On: 11/22/21.

11.1.5. Photoirritation/photoallergenicity

Based on available *in vitro* study data, acetanisole does not present a concern for photoirritation. Acetanisole was not evaluated for photoallergy; however, RIFM is sponsoring an *in vitro* photoallergy research program to evaluate its photoallergy potential.

11.1.5.1. Risk assessment. UV/Vis absorbance spectra indicate significant absorbance in the range of 290–700 nm, with a peak absorbance at 290 nm and a return to baseline by approximately 330 nm. Molar absorption coefficients are above the benchmark of concern for photoirritation/photoallergenicity (Henry et al., 2009). In an *in vitro* 3T3-Neutral Red Uptake photoirritation assay (OECD TG 432), acetanisole was not predicted to have photoirritating potential according to the prediction model presented in the test guidelines (RIFM, 2015). Based on the available *in vitro* study data, acetanisole does not present a concern for photoirritation. Acetanisole was not evaluated for photoallergy; however, RIFM is sponsoring an *in vitro* photoallergy research program to evaluate its photoallergy potential.

11.1.5.2. UV spectra analysis. UV/Vis absorption spectra (OECD TG 101) were generated for acetanisole. The spectra demonstrate significant absorbance between 290 and 700 nm, with peak absorbance at 290 nm and returning to baseline by approximately 330 nm. Molar absorption coefficients (10443, 4817, and 11869 L mol^{-1} • cm^{-1} under neutral, acidic, and basic conditions, respectively) are above the benchmark of concern for photoirritating effects, 1000 L mol^{-1} • cm^{-1} (Henry et al., 2009).

Additional References: None.

Literature Search and Risk Assessment Completed On: 11/22/21.

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 acetanisole is below the Cramer Class I TTC value for inhalation exposure local effects.

Table 1
Summary of existing data on acetanisole.

WoE Skin Sensitization Potency Category[a]	Human Data				Animal 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	LLNA Weighted Mean EC3 Value μg/cm^2	GPMT[d]	Buehler[d]
No evidence of sensitization[f]	NA	4140	NA	NA	Negative up to 12500	NA	NA
	***In vitro* Data[e]**				***In silico* protein binding alerts (OECD Toolbox v4.2)**		
	KE 1	**KE 2**	**KE 3**		**Target Material**	**Autoxidation simulator**	**Metabolism simulator**
	Negative	Positive	Negative (h-CLAT) Positive (U-SENS)		No alert found	No alert found	Schiff base formation

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

[a] WoE Skin Sensitization Potency Category is only applicable for identified sensitizers with sufficient data, based on collective consideration of all available data (Na et al., 2021).

[b] Data derived from CNIH or HMT.

[c] WoE NESIL limited to 2 significant figures.

[d] Studies conducted according to OECD TG 406 are included in the table.

[e] Studies conducted according to OECD TG 442, Cottrez et al. (2016), or Forreryd et al. (2016) are included in the table.

[f] Determined based on Criteria for the Research Institute for Fragrance Materials, Inc. (RIFM) safety evaluation process for fragrance ingredients (Api et al., 2015).

11.1.6.1. Risk assessment. There are no inhalation data available on acetanisole. Based on the Creme RIFM Model, the inhalation exposure is 0.0091 mg/day. This exposure is 153.8 times lower than the Cramer Class I TTC value of 1.4 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.

Additional References None.

Literature Search and Risk Assessment Completed On: 11/22/21.

11.2. Environmental endpoint Summary

11.2.1. Screening-level assessment

A screening-level risk assessment of acetanisole 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, acetanisole 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, 2012a) did not identify acetanisole as possibly 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, 2017a). 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). 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 (2019), acetanisole does not present a risk to the aquatic compartment in the screening-level assessment.

11.2.2.2. Key studies

11.2.1.2.1. Biodegradation. **RIFM, 1992:** A 28-day biodegradation test was conducted according to EU Method C.4-B method (Modified OECD Screening Test). Biodegradation of 100% was observed after 28 days.

11.2.1.2.2. Ecotoxicity. **RIFM, 2017e:** An algae growth inhibition test was conducted according to the OECD 201 method. The 72-h EC50 was reported to be 53 mg/L and 29 mg/L based on growth rate and yield, respectively. The 72-h EC10 was reported to be 28 mg/L for growth rate and 13 mg/L for yield.

RIFM, 2017f: A *Daphnia magna* acute immobilization test was conducted according to the OECD 202 method under static conditions. The 48-h EC50 based on nominal concentrations was reported to be 75 mg/L.

RIFM, 2017g: The acute toxicity of the test material to zebrafish (*Danio rerio*) was determined in a 96-h test according to the OECD 203 guidelines. A limit test was performed to demonstrate that the fish is not the most sensitive test organism. Zebrafish were exposed to an aqueous test medium containing the test material at the threshold concentration of nominal 60 mg/L. The 96-h LC50 for fish was reported to be greater than the threshold concentration of nominal 60 mg/L.

11.2.1.2.3. Other available data. Acetanisole has been registered under REACH but has no additional data at this time.

11.2.2. Risk assessment refinement

Since acetanisole has passed screening criteria (Tier 1), measured data are included for completeness only and have not been used in PNEC derivations.

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 Framework: Salvito et al, 2002)

Exposure	Europe	North America
Log K_{ow} Used	1.79	1.79
Biodegradation Factor Used	0	0
Dilution Factor	3	3
Regional Volume of Use Tonnage Band	1–10	1–10
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 0.3081 μg/L. The revised PEC/PNECs for EU and NA are <1; therefore, the material does not present a risk to the aquatic environment at the current reported VoU.

Literature Search and Risk Assessment Completed On: 11/09/21.

12. 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
  - **PubChem:** https://pubchem.ncbi.nlm.nih.gov/
  - **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 ChemView:** https://chemview.epa.gov/chemview/
  - **Japanese NITE:** https://www.nite.go.jp/en/chem/chrip/chrip_search/systemTop

	LC50 (Fish) (mg/L)	EC50 (*Daphnia*) (mg/L)	EC50 (Algae) (mg/L)	AF	PNEC (μg/L)	Chemical Class
RIFM Framework Screening-level **(Tier 1)**	308.1			1000000	0.3081	

- **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 06/08/22.

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.2022.113465.

Appendix

Read-across Justification

Methods

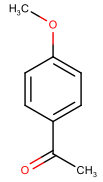
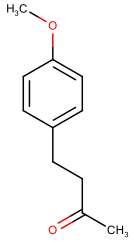
The read-across analog was identified using RIFM fragrance materials chemical inventory clustering and read-across search criteria (Date et al., 2020). These criteria follow the strategy for structuring and reporting a read-across prediction of toxicity as described in Schultz et al. (2015) and are consistent 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, 2017b).

- 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	Read-across Material
Principal Name	Acetanisole	4-(*p*-Methoxyphenyl)-2-butanone
CAS No.	100-06-1	104-20-1
Structure		

(*continued on next page*)

(*continued*)

	Target Material	Read-across Material
		
Similarity (Tanimoto Score)		0.62
SMILES	COc1ccc(cc1)C(C) = O	COc1ccc(CCC(C) = O)cc1
Endpoint		Genotoxicity
Molecular Formula	$C_9H_{10}O_2$	$C_{11}H_{14}O_2$
Molecular Weight (g/mol)	150.177	178.231
Melting Point (°C, EPI Suite)	38.50	8.00
Boiling Point (°C, EPI Suite)	258.00	264.70
Vapor Pressure (Pa at 25 °C, EPI Suite)	8.59E-01	1.64E+00
Water Solubility (mg/L, at 25 °C, WSKOW v1.42 in EPI Suite)	2.47E+03	1.01E+03
Log KOW	1.74	2.04
J_{max} (μg/cm^2/h, SAM)	35.91	11.63
Henry's Law (Pa·m^3/mol, Bond Method, EPI Suite)	5.88E-02	3.18E-02
Genotoxicity		
DNA Binding (OASIS v1.4, QSAR Toolbox v4.2)	No alert found	No alert found
DNA Binding (OECD QSAR Toolbox v4.2)	No alert found	No alert found
Carcinogenicity (ISS)	No alert found	No alert found
DNA Binding (Ames, MN, CA, OASIS v1.1)	No alert found	No alert found
***In Vitro* Mutagenicity (Ames, ISS)**	No alert found	No alert found
***In Vivo* Mutagenicity (Micronucleus, ISS)**	No alert found	No alert found
Oncologic Classification	Not classified	Not classified
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 acetanisole (CAS # 100-06-1). 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, 4-(*p*-methoxyphenyl)-2-butanone (CAS # 104-20-1) was identified as a read-across analog with sufficient data for toxicological evaluation.

Conclusions

- 4-(*p*-Methoxyphenyl)-2-butanone (CAS # 104-20-1) was used as a read-across analog for the target material, acetanisole (CAS # 100-06-1), for the genotoxicity endpoint.
  - o The target material and the read-across analog are structurally similar and belong to a class of aromatic ketones.
  - o The key difference between the target material and the read-across analog is that the target material has a ketone conjugated to the aromatic ring, while the read-across analog has an insulated ketone attached to the aromatic ring. The read-across analog contains the structural features of the target material that are relevant to this endpoint and is expected to have equal or greater potential for toxicity as compared to the target.
  - 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 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 OECD QSAR Toolbox v4.2, structural alerts for toxicological endpoints are consistent between the target material and the read-across analog.
  - o There are no *in silico* alerts for the read-across analog and the target material. *In silico* alerts are consistent with data.
  - 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.

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