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Short review

RIFM fragrance ingredient safety assessment, 2-methyl-4-phenylbutyraldehyde, CAS Registry Number 40654-82-8

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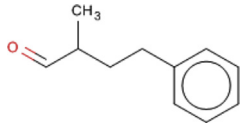
Version: 042518. This version replaces any previous versions.
Name: 2-Methyl-4-phenylbutyraldehyde
CAS Registry Number: 40654-82-8



1. Identification

1. **Chemical Name:** 2-Methyl-4-phenylbutyraldehyde
2. **CAS Registry Number:** 40654-82-8
3. **Synonyms:** Benzenebutanal, α-methyl-; 2-Methyl-4-phenylbutanal; Otropal; 2-Methyl-4-phenylbutyraldehyde
4. **Molecular Formula:** $C_{11}H_{14}O$
5. **Molecular Weight:** 162.23
6. **RIFM Number:** 6763

2. Physical data

1. **Boiling Point:** 246.48 °C (EPI Suite)
2. **Flash Point:** 245.00 °F; TCC (118.33 °C)*
3. **Log K_{ow}:** 2.94 (EPI Suite)
4. **Melting Point**: 12.12 °C (EPI Suite)
5. **Water Solubility:** 205.5 mg/L (EPI Suite)
6. **Specific Gravity:** 0.96800 to 0.97500 @ 25.00 °C*
7. **Vapor Pressure:** 0.0209 mm Hg @ 20 °C (EPI Suite), 0.01 mm Hg 20 °C (FMA), 0.0329 mm Hg @ 25 °C (EPI Suite)

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8. **UV Spectra:** No absorbance between 290 and 700 nm; molar absorption coefficient is below the benchmark (1000 L $mol^{-1} \cdot cm^{-1}$)
9. **Appearance/Organoleptic:** Colorless oily liquid, earthy musty but sweet and also floral; sweet and has a slightly nut-like taste in dilutions below 5 ppm (Arctander, 1969)

*http://www.thegoodscentscompany.com/data/rw1032931.html, retrieved 4/9/2015.

3. Exposure

1. **Volume of Use (worldwide band):** < 0.1 metric tons per year (IFRA, 2015)
2. **95th Percentile Concentration in Hydroalcoholics:** 0.11% (RIFM, 2013a)
3. **Inhalation Exposure*:** 0.00012 mg/kg/day or 0.0078 mg/day (RIFM, 2013a)
4. **Total Systemic Exposure**:** 0.0012 mg/kg/day (RIFM, 2013a)

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

**95th percentile calculated exposure; assumes 100% absorption unless modified by dermal absorption data as reported in Section 4. 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., 2015, 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 I, Low

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

2. Analogs Selected:
   a. **Genotoxicity:** β-methyl-benzenepentanal (CAS # 55066-49-4)
   b. **Repeated Dose Toxicity:** β-methyl-benzenepentanal (CAS # 55066-49-4)
   c. **Developmental and 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

Not considered for this risk assessment.

7. NATURAL OCCURRENCE (discrete chemical) or COMPOSITION (NCS)

2-Methyl-4-phenylbutyraldehyde is not reported to occur in food 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 that contains 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 11/30/2010; no dossier available as of 4/24/2018.

10. Summary

10.1. Human health endpoint summaries

10.1.1. Genotoxicity

Based on the current data, 2-methyl-4-phenylbutyraldehyde does not present a concern for genotoxicity.

10.1.1.1. Risk assessment. 2-Methyl-4-phenylbutyraldehyde was assessed in the BlueScreen assay and found negative for genotoxicity, with and without metabolic activation (RIFM, 2013b). There are no data assessing the mutagenic activity of 2-methyl-4-phenylbutyraldehyde. The mutagenic and clastogenic activity of read-across material β-methyl-benzenepentanal (CAS # 55066-49-4; see Section 5) was assessed in a *Salmonella* (Ames) mutagenicity assay in compliance with GLP regulations and in accordance with OECD TG 471. *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537, and TA1538 were treated with β-methyl-benzenepentanal in acetone at concentrations up to 5000 μg/plate. No increases in the mean number of revertant colonies were observed at any tested dose in the presence or absence of S9 (RIFM, 1996). Under the conditions of the study, β-methyl-benzenepentanal was not mutagenic in the Ames test.

There are no data assessing the clastogenic activity of 2-methyl-4-phenylbutyraldehyde. The clastogenic and aneugenic potential of read-across material β-methyl-benzenepentanal was evaluated in cultured human lymphocytes using OECD TG 473/GLP. Human peripheral blood lymphocytes were treated with β-methyl-benzenepentanal in dimethyl sulfoxide (DMSO) at concentrations up to 1760 μg/mL in the presence and absence of metabolic activation. No statistically significant increases in the frequency of cells with structural chromosomal aberrations or polyploid cells were observed with any dose of the test item, either with or without S9 metabolic activation (RIFM, 1999). Under the conditions of the study, β-methyl-benzenepentanal was considered to be non-clastogenic to human cells.

Based on the available read-across data on β-methyl-benzenepentanal, 2-methyl-4-phenylbutyraldehyde does not present a concern for genotoxic potential.

Additional References: RIFM, 1985.

Literature Search and Risk Assessment Completed On: 06/11/2017.

10.1.2. Repeated dose toxicity

The margin of exposure for 2-methyl-4-phenylbutyraldehyde is adequate for the repeated dose toxicity endpoint at the current level of use.

10.1.2.1. Risk assessment. There are insufficient repeated dose toxicity data on 2-methyl-4-phenylbutyraldehyde. Read-across material β-methyl-benzenepentanal (CAS # 55066-49-4; see Section 5) has sufficient repeated dose toxicity data. An OECD/GLP 407 28-day toxicity study was conducted in Sprague Dawley rats. Groups of 5 rats/sex/dose received β-methyl-benzenepentanal via gavage at doses of 0, 15, 150, or 500 mg/kg/day in an Arachis oil BP vehicle. Treatment with β-methyl-benzenepentanal

resulted in a statistically significant increase in aspartate aminotransferase and an increase (not significant) in alanine transferase levels among high-dose females. Organ weight analysis revealed a statistically significant increase in liver weights, both absolute and relative, among high-dose females as compared to the controls. High-dose males showed statistically significant increases in the relative liver weights as compared to the controls. Both absolute and relative kidney weights were statistically significantly increased among high-dose females as compared to the controls, whereas only the relative kidney weights were statistically significantly increased among mid-dose females. Macroscopic examination revealed no treatment-related alterations among treated animals. Microscopic alterations reported included centrilobular hepatocyte enlargement among high-dose males and an increased severity of glycogen type vacuolation of hepatocytes among high-dose females. Thyroid gland follicular cell hypertrophy with colloid depletion was reported among high-dose females. Thus, it was concluded that treatment with β-methyl-benzenepentanal resulted in liver, kidney, and thyroid organ alterations. Alterations in the liver were considered to be test material–related adverse effects since treatment resulted in liver weight alterations with corresponding histopathological and clinical chemistry correlates. Alterations in the thyroid gland were considered to be a secondary response to hepatocyte hypertrophy due to the induction of hepatocyte drug metabolizing enzymes that may increase the turnover of T4, thereby resulting in secondary thyroid hypertrophy due to stimulation of the hypothalamus-pituitary-thyroid axis (Hall et al., 2012). Alterations in the kidneys of mid- and high-dose females were not associated with any histopathological alterations or changes in kidney function, nor was there any dose response in terms of kidney weights reported. Thus, the alterations in kidney weights were not considered to be adverse. Therefore, the NOAEL for repeated dose toxicity was considered to be 150 mg/kg/day, based on statistically significant increased liver weights with associated histopathological alterations among animals of the high-dose group and alterations in clinical chemistry among high-dose females (RIFM, 2000).

A default safety factor of 3 was used when deriving a NOAEL from a 28-day OECD 407 study. The safety factor has been approved by the Expert Panel for Fragrance Safety*.

Thus, the derived NOAEL for the repeated dose toxicity data is 150/3 or 50 mg/kg/day.

Therefore, the 2-methyl-4-phenylbutyraldehyde MOE for the repeated dose toxicity endpoint can be calculated by dividing the β-methyl-benzenepentanal NOAEL in mg/kg/day by the total systemic exposure to 2-methyl-4-phenylbutyraldehyde, 50/0.0012 or 41667.

In addition, the total systemic exposure to 2-methyl-4-phenylbutyraldehyde (1.2 μg/kg bw/day) is below the TTC (30 μg/kg bw/day; Kroes et al., 2007) for the repeated dose toxicity endpoint of a Cramer Class I material at the current level of use.

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

Additional References: None.

Literature Search and Risk Assessment Completed On: 06/08/2017.

10.1.3. Developmental and reproductive toxicity

There are insufficient developmental and reproductive toxicity data on 2-methyl-4-phenylbutyraldehyde or any read-across materials. The total systemic exposure to 2-methyl-4-phenylbutyraldehyde is below the TTC for the developmental and reproductive toxicity endpoints of a Cramer Class I material at the current level of use.

10.1.3.1. Risk assessment. There are no developmental or reproductive toxicity data on 2-methyl-4-phenylbutyraldehyde or any read-across materials that can be used to support the developmental or reproductive toxicity endpoints. The total systemic exposure to 2-methyl-4-phenylbutyraldehyde (1.2 μg/kg/day) is below the TTC (30 μg/kg/day; Kroes et al., 2007; Laufersweiler et al., 2012) for the developmental and reproductive toxicity endpoints of a Cramer Class I material at the current level of use.

Additional References: None.

Literature Search and Risk Assessment Completed On: 06/08/17.

10.1.4. Skin sensitization

Based on the existing data, 2-methyl-4-phenylbutyraldehyde 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 be expected to react with skin proteins via Schiff base formation (Toxtree 2.6.13, OECD toolbox v3.4). No predictive tests are available for 2-methyl-4-phenylbutyraldehyde. However, in a human repeat insult patch test (HRIPT), no reactions were observed in any of the 107 volunteers when 15% or 8333 μg/cm^2 2-methyl-4-phenylbutyraldehyde in 1:3 ethanol:diethyl phthalate was used for induction and challenge (IFF, 2004). Based on the weight of evidence from structural analysis and available human data, 2-methyl-4-phenylbutyraldehyde does not present a safety concern for skin sensitization under the current declared levels of use.

Additional References: None.

Literature Search and Risk Assessment Completed On: 06/15/17.

10.1.5. Phototoxicity/photoallergenicity

	Phototoxicity	Photoallergenicity
Step 1: UV benchmark ($1000\ L\ mol^{-1} \cdot cm^{-1}$)	**Below**	
Step 2: Study data		
Step 3: Exposure benchmark		
Step 4: Read-across		
Step 5: Generate data		

Based on available UV/Vis absorption spectra, 2-methyl-4-phenylbutyraldehyde would not be expected to present a concern for phototoxicity or photoallergenicity.

10.1.5.1. Risk assessment. There are no phototoxicity studies available for 2-methyl-4-phenylbutyraldehyde in experimental models. The available UV/Vis spectra for 2-methyl-4-phenylbutyraldehyde indicate no significant absorbance 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). Based on the lack of absorbance in the critical range, 2-methyl-4-phenylbutyraldehyde 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, $1000\ L\ mol^{-1} \cdot cm^{-1}$ (Henry et al., 2009).

Additional References: None.

Literature Search and Risk Assessment Completed On: 05/24/17.

10.1.6. Local respiratory toxicity

The margin of exposure could not be calculated due to lack of appropriate data. The exposure level of 2-methyl-4-phenylbutyraldehyde is below the Cramer Class I TTC value for inhalation exposure local effects.

10.1.6.1. Risk assessment. There are no inhalation data available on 2-

methyl-4-phenylbutyraldehyde. Based on the Creme RIFM model, the inhalation exposure is 0.0078 mg/day. This exposure is 179 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: 05/31/16.

10.2. Environmental endpoint summary

10.2.1. Screening-level assessment

A screening-level risk assessment of 2-methyl-4-phenylbutyraldehyde 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, 2-methyl-4-phenylbutyraldehyde 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 v4.1 (US EPA, 2012a) did not identify 2-methyl-4-phenylbutyraldehyde 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, 2012). For persistence, if the EPI Suite model BIOWIN 3 predicts a value < 2.2 and either BIOWIN 2 or BIOWIN 6 predicts a value < 0.5, then the material is considered potentially persistent. A material would be considered potentially bioaccumulative if the EPI Suite model BCFBAF predicts a fish BCF ≥2000 L/kg. Ecotoxicity is determined in the above screening-level risk assessment. If, based on these model outputs (Step 1), additional assessment is required, a WoE-based review is then performed (Step 2). This review considers available data on the material's physical–chemical properties, environmental fate (e.g., OECD Guideline biodegradation studies or die-away studies), fish bioaccumulation, and higher-tier model outputs (e.g., US EPA's BIOWIN and BCFBAF found in EPI Suite v4.1). Data on persistence and bioaccumulation are reported below and summarized in the Environmental Safety Assessment section prior to Section 1.

10.2.2. Risk assessment

Based on current Volume of Use (2015), 2-methyl-4-phenylbutyraldehyde does not present a risk to the aquatic compartment in the screening-level assessment.

Biodegradation: No data available.

Ecotoxicity: No data available.

Other available data: 2-Methyl-4-phenylbutyraldehyde has been pre-registered for REACH with no additional data at this time.

10.2.3. Risk assessment refinement

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

Endpoints used to calculate PNEC are underlined.

	LC50 (Fish) (mg/L)	EC50 (*Daphnia*) (mg/L)	EC50 (Algae) (mg/L)	AF	PNEC (µg/L)	Chemical Class
RIFM Framework Screening-level **(Tier 1)**	<u>33.28</u>			1,000,000	0.03328	

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

Exposure	Europe (EU)	North America (NA)
Log K_{ow} Used	2.9	2.9
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 0.03328 µg/L. The revised PEC/PNECs for EU and NA: Not applicable; 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: 6/8/17.

11. Literature Search*

- **RIFM Database:** Target, Fragrance Structure Activity Group materials, other references, JECFA, CIR, SIDS
- **ECHA:** http://echa.europa.eu/

- **NTP:** http://tools.niehs.nih.gov
- OECD Toolbox
- **SciFinder:** https://scifinder.cas.org/scifinder/view/scifinder/scifinderExplore.jsf
- **PubMed:** http://www.ncbi.nlm.nih.gov/pubmed
- **TOXNET:** http://toxnet.nlm.nih.gov/
- **IARC:** http://monographs.iarc.fr
- **OECD SIDS:** http://webnet.oecd.org/hpv/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:** http://www.safe.nite.go.jp/english/db.html
- **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.

Conflicts of interest

The authors declare that they have no conflicts of interest.

Appendix A. Supplementary data

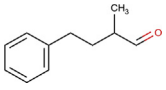
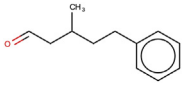
Supplementary data related to this article can be found at https://doi.org/10.1016/j.fct.2018.08.054.

AppendixRead-across Justification

Methods

The read-across analogs were identified following the strategy for structuring and reporting a read-across prediction of toxicity 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 substance 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 v3.4 (OECD, 2012).
- ER binding and repeat dose categorization were generated using OECD QSAR Toolbox v3.4 (OECD, 2012).
- Developmental toxicity was predicted using CAESAR v2.1.7 (Cassano et al., 2010) and skin sensitization was predicted using Toxtree 2.6.13.
- Protein binding was predicted using OECD QSAR Toolbox v3.4 (OECD, 2012).
- The major metabolites for the target and read-across analogs were determined and evaluated using OECD QSAR Toolbox v3.4 (OECD, 2012).

	Target material	Read-across material
Principal Name	2-Methyl-4-phenylbutyraldehyde	β-Methyl-benzenepentanal
CAS No.	40654-82-8	55066-49-4
Structure		
Similarity (Tanimoto score)		0.91
Read-across Endpoint		• Genotoxicity • Repeated dose
Molecular Formula	$C_{11}H_{14}O$	$C_{12}H_{16}O$
Molecular Weight	162.23	176.26
Melting Point (°C, EPI SUITE)		22.79
Boiling Point (°C, EPI Suite)	246.48	263.7
Vapor Pressure (Pa @ 25 °C, EPI Suite)	4.38	1.74
Log K_{ow} (KOWWIN v1.68 in EPI Suite)	2.94	3.43
Water Solubility (mg/L, @ 25 °C, WSKOW v1.42 in EPI Suite)	205.5	67.2
J_{max} (mg/cm^2/h, SAM)	57.976	25.487
Henry's Law (Pa·m^3/mol, Bond Method, EPI Suite)	1.30E+000	1.72E+000
Genotoxicity		
DNA Binding (OASIS v 1.4 QSAR Toolbox 3.4)	• No alert found	• No alert found
DNA binding by OECD QSAR Toolbox (3.4)	• Michael Addition • Schiff Base formers	• Michael Addition • Schiff Base formers
Carcinogenicity (Genotox and Non-Genotox) Alerts (ISS)	• Carcinogen (Moderate reliability)	• Carcinogen (Moderate reliability)
DNA Alerts for Ames, MN, CA by OASIS v 1.1	• No alert found	• No alert found

In vitro Mutagenicity (Ames test) Alerts by ISS	• Simple aldehyde	• Simple aldehyde
In vivo Mutagenicity (Micronucleus) Alerts by ISS	• Simple aldehyde	• Simple aldehyde
Oncologic Classification	• Aldehyde type compounds	• Aldehyde type compounds
Repeated dose toxicity		
Repeated Dose (HESS)	• Not categorized	• Not categorized
Metabolism		
OECD QSAR Toolbox (3.4)	See Supplemental Data 1	See Supplemental Data 2
Rat Liver S9 Metabolism Simulator and Structural Alerts for Metabolites		

Summary

There are insufficient toxicity data on the target material 2-methyl-4-phenylbutyraldehyde (CAS # 40654-82-8). Hence, *in silico* evaluation was conducted by determining a read-across analog for this material. Based on structural similarity, reactivity, metabolism data, physical–chemical properties, and expert judgment, analog β-methyl-benzenepentanal (CAS # 55066-49-4) was identified as a read-across material with data for its respective toxicological endpoints.

Conclusions

- β-methyl-benzenepentanal (CAS # 55066-49-4) was used as a read-across analog for the target material 2-methyl-4-phenylbutyraldehyde (CAS # 40654-82-8) for the genotoxicity and repeated dose toxicity endpoints.
  - ○ The target substance and the read-across analog are structurally similar and belong to the structural class of aromatic aldehydes.
  - ○ The target substance and the read-across analog share an aromatic ring with a distant aldehyde moiety.
  - ○ The key difference between the target substance and the read-across analog is that the methyl substitution is at the alpha carbon in the target substance, while the methyl substitution is at the beta carbon in the read-across analog. This structural difference between the target substance and the read-across analog does not affect consideration of the toxicological endpoint.
  - ○ Similarity between the target substance and the read-across analog is indicated by the Tanimoto score in the table above. Differences between the structures that affect the Tanimoto score do not affect consideration of the toxicological endpoint.
  - ○ The physical–chemical properties of the target substance and the read-across analog are sufficiently similar to enable comparison of their toxicological properties.
  - ○ According to the QSAR OECD Toolbox (v3.4), structural alerts for the toxicological endpoints are consistent between the target substance and the read-across analog.
  - ○ The target substance and the read-across analog have a carcinogenicity alert by the ISS model. The target and the read-across analog also have *in vitro* and *in vivo* mutagenicity alerts along with a DNA binding alert by OECD and are classified as simple aldehydes. This shows that the read-across analog is predicted to have comparable reactivity with the target substance. The data described in the genotoxicity section show that the read-across analog does not pose a concern for genotoxicity. Therefore, the alert will be superseded by the available data.
  - ○ The target substance and the read-across analog are expected to be metabolized similarly, as shown by the metabolism simulator.
  - ○ The structural differences between the target material and the read-across analog do not affect consideration of the toxicological endpoints.

References

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

Arctander, S., 1969. Perfume and Flavor Chemicals (Aroma Chemicals), vols. I and II Published by the author: Montclair, NJ (USA).

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

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

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

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

ECHA, 2012. Guidance on Information Requirements and Chemical Safety Assessment Chapter R.11: PBT Assessment, November 2012 v1.1. http://echa.europa.eu/.

ECHA, 2016. Read-across Assessment Framework (RAAF). Retrieved from. www.echa.europa.eu/documents/10162/13628/raaf_en.pdf.

Hall, A.P., Elcombe, C.R., Foster, J.R., Harada, T., Kaufmann, W., et al., 2012. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes–conclusions from the 3rd International ESTP expert workshop. Toxicol. Pathol. 40 (7), 971–994.

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

IFF Incorporated, 2004. Repeat Insult Patch Test with 2-methyl-4-phenylbutyraldehyde. Carcinogenesis (Unpublished).

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

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

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

OECD, 2012. The OECD QSAR Toolbox, V 3.4. Retrieved from. http://www.qsartoolbox.org/.

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

RIFM (Research Institute for Fragrance Materials, Inc), 1985. Examination of Beta-methyl-benzenepentanal (Mefranal) for Mutagenic Activity in the Ames Test. Unpublished report from Givaudan. RIFM report number 57931. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc), 1996. Beta-methyl-benzenepentanal: Reverse Mutation Assay "Ames Test" Using Salmonella typhimurium. Unpublished report from Firmenich Incorporated. RIFM report number 38473. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc), 1999. Beta-methyl-benzenepentanal (Citralis): Chromosome Aberration Test in Human Lymphocytes in Vitro. Unpublished report from International Flavors and Fragrances. RIFM report number 48891. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc), 2000. Beta-methyl-benzenepentanal (Citralis): Twenty-eight Day Repeated Dose Oral (Gavage) Toxicity Study in the Rat. Unpublished report from International Flavors and Fragrances. RIFM report

number 48896. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc), 2013a. Exposure Survey 01, May 2013.
RIFM (Research Institute for Fragrance Materials, Inc), 2013b. Report on the Testing of 2-methyl-4-phenylbutyraldehyde in the BlueScreen HC Assay (-/+ S9 Metabolic Activation). RIFM report number 66281. RIFM, Woodcliff Lake, NJ, USA.
Rogers, D., Hahn, M., 2010. Extended-connectivity fingerprints. J. Chem. Inf. Model. 50 (5), 742–754.
Safford, B., Api, A.M., Barratt, C., Comiskey, D., Daly, E.J., Ellis, G., McNamara, C., O'Mahony, C., Robison, S., Smith, B., Thomas, R., Tozer, S., 2015. Use of an aggregate exposure model to estimate consumer exposure to fragrance ingredients in personal care and cosmetic products. Regul. Toxicol. Pharmacol. 72, 673–682.
Safford, B., Api, A.M., Barratt, C., Comiskey, D., Ellis, G., McNamara, C., O'Mahony, C., Robison, S., Rose, J., Smith, B., Tozer, S., 2017. Application of the expanded Creme RIFM consumer exposure model to fragrance ingredients in cosmetic, personal care and air care products. Regul. Toxicol. Pharmacol. 86, 148–156.
Salvito, D.T., Senna, R.J., Federle, T.W., 2002. A Framework for prioritizing fragrance materials for aquatic risk assessment. Environ. Toxicol. Chem. 21 (6), 1301–1308.
Schultz, T.W., Amcoff, P., Berggren, E., Gautier, F., Klaric, M., Knight, D.J., Mahony, C., Schwarz, M., White, A., Cronin, M.T.D., 2015. A strategy for structuring and reporting a read-across prediction of toxicity. Regul. Toxicol. Pharmacol. 72 (3), 586–601.
Shen, J., Kromidas, L., Schultz, T., Bhatia, S., 2014. An *in silico* skin absorption model for fragrance materials. Food Chem. Toxicol. 74 (12), 164–176.
US EPA, 2012a. Estimation Programs Interface Suite for Microsoft Windows, v4.0–v4.11. United States Environmental Protection Agency, Washington, DC, USA.
US EPA, 2012b. The ECOSAR (ECOlogical Structure Activity Relationship) Class Program for Microsoft Windows, v1.11. United States Environmental Protection Agency, Washington, DC, USA.