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Short review

RIFM fragrance ingredient safety assessment, benzyl isobutyrate, CAS Registry Number 103-28-6

A.M. Api [a, *], D. Belsito [b], S. Bhatia [a], M. Bruze [c], P. Calow [d], M.L. Dagli [e], W. Dekant [f], A.D. Fryer [g], L. Kromidas [a], S. La Cava [a], J.F. Lalko [a], A. Lapczynski [a], D.C. Liebler [h], V.T. Politano [a], G. Ritacco [a], D. Salvito [a], T.W. Schultz [i], J. Shen [a], I.G. Sipes [j], B. Wall [a], D.K. Wilcox [a]

[a] Research Institute for Fragrance Materials, Inc., 50 Tice Boulevard, Woodcliff Lake, NJ 07677, USA
[b] Member RIFM Expert Panel, Columbia University Medical Center, Department of Dermatology, 161 Fort Washington Ave., New York, NY 10032, USA
[c] Member RIFM Expert Panel, Malmo University Hospital, Department of Occupational & Environmental Dermatology, Sodra Forstadsgatan 101, Entrance 47, Malmo SE-20502, Sweden
[d] Member RIFM Expert Panel, Humphrey School of Public Affairs, University of Minnesota, 301 19th Avenue South, Minneapolis, MN 55455, USA
[e] Member RIFM Expert Panel, University of Sao Paulo, School of Veterinary Medicine and Animal Science, Department of Pathology, Av. Prof. dr. Orlando Marques de Paiva, 87, Sao Paulo CEP 05508-900, Brazil
[f] Member RIFM Expert Panel, University of Wuerzburg, Department of Toxicology, Versbacher Str. 9, 97078 Würzburg, Germany
[g] Member RIFM Expert Panel, Oregon Health Science University, 3181 SW Sam Jackson Park Rd., Portland, OR 97239, USA
[h] Member RIFM Expert Panel, Vanderbilt University School of Medicine, Department of Biochemistry, Center in Molecular Toxicology, 638 Robinson Research Building, 2200 Pierce Avenue, Nashville, TN 37232-0146, USA
[i] Member RIFM Expert Panel, The University of Tennessee, College of Veterinary Medicine, Department of Comparative Medicine, 2407 River Dr., Knoxville, TN 37996-4500, USA
[j] Member RIFM Expert Panel, Department of Pharmacology, University of Arizona, College of Medicine, 1501 North Campbell Avenue, P.O. Box 245050, Tucson, AZ 85724-5050, USA

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1. Identification

1. **Chemical Name:** Benzyl isobutyrate
2. **CAS Registry Number:** 103-28-6
3. **Synonyms:** Benzyl isobutyrate, Benzyl 2-methylpropanoate, Propanoic acid, 2-methyl-, phenylmethyl ester, ｱﾙｶﾝ酸(C = 1 ~ 6) ﾍﾞﾝｼﾞﾙ, ｱﾙｷﾙ（C = 1 ~ 5）ｶﾙﾎﾞﾝ酸ﾌｪﾆﾙｱﾙｷﾙ（C = 1 ~ 6）
4. **Molecular Formula:** $C_{11}H_{14}O_2$
5. **Molecular Weight:** 178.23
6. **RIFM Number:** 227

2. Physical data

1. **Boiling Point:** 229 °C [FMA database], 241.5 °C [EPI Suite]
2. **Flash Point**: >200 °F;CC [FMA database]
3. **Log K_{OW}**: 2.99 [EPI Suite]
4. **Melting Point**: 10.84 °C [EPI Suite]
5. **Water Solubility:** 157.2 mg/L [EPI Suite]
6. **Specific Gravity:** 1.000–1.005 [FMA], 1.002–1.007 [FMA database]
7. **Vapor Pressure:** 0.0274 mm Hg @ 20 °C [EPI Suite 4.0], 0.04 mm Hg 20 °C [FMA database], 0.0428 mm Hg @ 25 °C [EPI Suite]
8. **UV Spectra:** No absorption between 290 and 400 nm; molar absorption coefficient is below the benchmark (1000 L mol^{-1} cm^{-1}).
9. **Appearance/Organoleptic:** A colorless to pale yellow liquid having a fruity and jasmin-like odor.

* Corresponding author.
E-mail address: AApi@rifm.org (A.M. Api).

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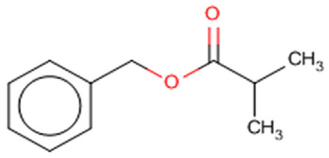
Version: 030816. This version replaces any previous versions.
Name: Benzyl isobutyrate
CAS Registry Number: 103-28-6



Abbreviation list:
2-Box Model – a RIFM, Inc. proprietary *in silico* tool used to calculate fragrance air exposure concentration
97.5th **percentile** – The concentration of the fragrance ingredient is obtained from examination of several thousand commercial fine fragrance formulations. The upper 97.5th percentile concentration is calculated from these data and is then used to estimate the dermal systemic exposure in ten types of the most frequently used personal care and cosmetic products. The dermal route is the major route in assessing the safety of fragrance ingredients. Further explanation of how the data were obtained and of how exposures were determined has been previously reported by Cadby et al. (2002) and Ford et al. (2000).
AF – Assessment Factor
BCF – Bioconcentration Factor
DEREK – Derek nexus is an *in silico* tool used to identify structural alerts
DST – Dermal Sensitization Threshold
ECHA – European Chemicals Agency
EU – Europe/European Union
GLP – Good Laboratory Practice
IFRA – The International Fragrance Association
LOEL – Lowest Observable Effect Level
MOE – Margin of Exposure
MPPD – Multiple-Path Particle Dosimetry. An *in silico* model for inhaled vapors used to simulate fragrance lung deposition
NA – North America
NESIL – No Expected Sensitization Induction Level
NOAEC – No Observed Adverse Effect Concentration
NOAEL – No Observed Adverse Effect Level
NOEC – No Observed Effect Concentration
OECD – Organisation for Economic Co-operation and Development
OECD TG – Organisation for Economic Co-operation and Development Testing Guidelines
PBT – Persistent, Bioaccumulative, and Toxic
PEC/PNEC – Predicted Environmental Concentration/Predicted No Effect Concentration
QRA – quantitative risk assessment
REACH – Registration, Evaluation, Authorisation, and Restriction of Chemicals
RIFM – Research Institute for Fragrance Materials
RQ – Risk Quotient
TTC – Threshold of Toxicological Concern
UV/Vis Spectra – Ultra Violet/Visible spectra
VCF – Volatile Compounds in Food
VoU – Volume of Use
vPvB – (very) Persistent, (very) Bioaccumulative
WOE – Weight of Evidence

3. Exposure

1. **Volume of Use (worldwide band):** 1–10 metric tons per year (IFRA, 2011)
2. **Average Maximum Concentration in Hydroalcoholics:** 0.16% (IFRA, 2008)
3. **97.5**th **Percentile:** 0.71% (IFRA, 2008)
4. **Dermal Exposure*:** 0.0180 mg/kg/day (IFRA, 2008)
5. **Oral Exposure:** Not applicable
6. **Inhalation Exposures**:** 0.0011 mg/kg/day (IFRA, 2008)
7. **Total Systemic Exposure (Dermal + Inhalation):** (0.018 mg/kg/day x 78.7% absorption) + 0.0011 mg/kg/day = 0.015 mg/kg/day

*Calculated using the reported 97.5th percentile concentration based on the levels of the same fragrance ingredient in ten of the most frequently used personal care and cosmetic products (i.e., anti-perspirant, bath products, body lotion, eau de toilette, face cream, fragrance cream, hair spray, shampoo, shower gel, and toilet soap) (Cadby et al., 2002; Ford et al., 2000).

**Combined (fine fragrances, hair sprays, antiperspirants/deodorants, candles, aerosol air fresheners, and reed diffusers/heated oil plug-ins) result calculated using RIFM's 2-Box/MPPD *in silico* models, based on the IFRA survey results for the 97.5th percentile use in hydroalcoholics for a 60 kg individual.

4. Derivation of systemic absorption

1. **Dermal:** 78.7%

Bronaugh et al., 1990: The skin absorption of read across material [7–^{14}C] benzyl acetate (CAS # 104-11-1; see Section 5) was measured in 4 female rhesus monkeys. The test material in acetone was applied at a concentration of 4 μg/cm^2 to a 1 cm^2 area of abdominal skin for 24 h. Urine was collected for an additional 4 days. The extent of dermal absorption was estimated from the amount of ^{14}C-equivalents excreted in the urine over the 5 day collection period. When the application site was occluded with either plastic wrap or a glass chamber, the absorption of benzyl acetate was 17.3 ± 2.7% and 78.7 ± 7.5%, respectively. When the site was not occluded, the absorption was 34.6 ± 9.4%.

2. **Oral:** Data not available – not considered.
3. Inhalation: Assumed 100%
4. **Total:** Dermal (78.7%) + Inhalation (assume 100%) absorbed = (0.018 mg/kg/day x 78.7%) + 0.0011 mg/kg/day = 0.015 mg/kg/day

RIFM's Expert Panel* concludes that this material is safe under the limits 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 reviews 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 two digit month/day/year), both in the RIFM database (consisting of publicly available and proprietary data) and through publicly available information sources (i.e., 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 end-point value (e.g., PNEC, NOAEL, LOEL, and NESIL).

*RIFM's Expert Panel 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 guidance relevant to human health and environmental protection.

Summary: The use of this material under current use conditions is supported by the existing information.

This material was evaluated for genotoxicity, repeated dose toxicity, developmental toxicity, reproductive toxicity, local respiratory toxicity, phototoxicity, skin sensitization, as well as environmental safety. Data from the suitable read across analog, benzyl acetate (CAS # 140-11-4), show that this material is not genotoxic nor does it have skin sensitization potential. The repeated dose, developmental and reproductive, and local respiratory toxicity endpoints were completed using benzyl acetate (CAS # 140-11-4) as a suitable read across analog, which provided a MOE > 100. The phototoxicity/photoallergenicity endpoint was completed based on suitable UV spectra. The environmental endpoint was completed as described in the RIFM Framework.

Human Health Safety Assessment

Genotoxicity: Not genotoxic. (Tennant et al., 1987; Shelby et al., 1993)

Repeated Dose Toxicity: NOAEL = 14.5 mg/kg/day (NTP, 1993)

Developmental and Reproductive Toxicity: NOAEL = 100 mg/kg/day (Ishiguro et al., 1993)

Skin Sensitization: Not sensitizing (RIFM, 1971; RIFM, 1985a; RIFM, 1986a; RIFM, 1985b; RIFM, 1985c; RIFM, 1988a; RIFM, 1961; RIFM, 1975a; RIFM, 1975b; RIFM, 1975c; RIFM, 1975d; RIFM, 1975e; RIFM, 1988b; RIFM, 1988c; RIFM, 1988d; Klecak, 1985; NTP, 1993)

Phototoxicity/Photoallergenicity: Not phototoxic/photoallergenic (UV Spectra, RIFM Database)

Local Respiratory Toxicity: NOAEC = 10 ppm or 61.4 mg/m^3 (0.0614 mg/L) (RIFM, 2013)

Environmental Safety Assessment

Hazard Assessment:

Persistence: Screening Level: 2.9675 (BIOWIN 3) (EpiSuite ver 4.1)

Bioaccumulation: Screening Level: 43.61 L/kg (EpiSuite ver 4.1)

Ecotoxicity: Screening Level: 96 h Algae EC50: 4.284 mg/L (EpiSuite ver 4.1)

Conclusion: Not PBT as per IFRA Environmental Standard

Risk Assessment:

Screening-Level: PEC/PNEC (North America and Europe) > 1 (Salvito et al., 2002)

Critical Ecotoxicity Endpoint: 96 h Algae EC50: 4.284 mg/L (EpiSuite ver 4.1)

RIFM PNEC is: 0.4284 μg/L

- Revised PEC/PNECs (2011 IFRA VoU): North America and Europe <1

5. Computational toxicology evaluation

1. Cramer Classification: Class I, Low

2. Analogues Selected:
   a. **Genotoxicity:** Benzyl acetate (CAS # 140-11-4)
   b. **Repeated Dose Toxicity:** Benzyl acetate (CAS # 140-11-4)
   c. **Developmental and Reproductive Toxicity:** Benzyl acetate (CAS # 140-11-4)
   d. **Skin Sensitization:** Benzyl acetate (CAS # 140-11-4)
   e. **Phototoxicity/Photoallergenicity:** None
   f. **Local Respiratory Toxicity:** Benzyl acetate (CAS # 140-11-4)
   g. **Environmental Toxicity:** None

3. **Read-across Justification:** See Appendix below.

6. Metabolism

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

Expert judgment	Toxtree v 2.6	OECD QSAR toolbox v 3.2
I	I	I

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

Benzyl isobutyrate is reported to occur in the following foods* and in some natural complex substances (NCS):

Beer.
Cherimoya (Annona cherimolia Mill.)
Honey.
Mentha oils.
Passion fruit (Passiflora species).

*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, contains information on published volatile compounds which have been found in natural (processed) food products. Includes FEMA GRAS and EU-Flavis data.

8. IFRA standard

None.

9. Reach dossier

Pre-Registered for 2010; No dossier available as of 03/8/2016.

10. Summary

10.1. Human health endpoint summaries

Genotoxicity:
Based on the current existing data and use levels, benzyl

isobutyrate does not present a concern for genetic toxicity.

Risk Assessment:

Benzyl isobutyrate was tested by the BlueScreen assay and was found negative for cytotoxicity and genotoxicity indicating a lack of genotoxic potential (Birrell et al., 2013). There are no studies assessing the mutagenic potential of benzyl isobutyrate however, read across can be made to benzyl acetate (CAS # 140-11-4; see Section 5) which was assessed for mutagenicity in an Ames study similar to OECD TG 471 using the plate incorporation method. *S. typhimurium* strains TA98, TA100, TA1535, and TA1537 were exposed to concentrations of benzyl acetate up to 10 mg per plate in the presence and absence of liver S9 fractions. No substantial increases in revertant colonies were seen with benzyl acetate with or without metabolic activation (S9). The study concluded that benzyl acetate is not mutagenic (Tennant et al., 1987).

The clastogenic potential of benzyl acetate was assessed in several *in vitro* mouse lymphoma cell assays with and without metabolic activation with varying results. However, several *in vivo* studies assessing the materials effects on inducing chromosomal aberrations, unscheduled DNA synthesis, demonstrate that benzyl acetate lacks genotoxic potential (NTP, 1993; Steinmetz and Mirsalis, 1984). In an *in vivo* mammalian erythrocyte micronucleus assay performed similar to OECD TG 474, groups of 5–7 male B6C3F1 mice were administered benzyl acetate in corn oil via intraperitoneal injection for 3 consecutive days at doses up to 600 mg/kg. No genotoxic activity was observed, and the material was considered not clastogenic (Shelby et al., 1993).

Taken together, benzyl acetate does not present a concern for genotoxic potential and this can be extended to benzyl isobutyrate.

Additional References: NTP, 1993; Florin et al., 1980; Mortelmans et al., 1986; Schunk et al., 1986; Rogan et al., 1986; Mirsalis et al., 1989; Steinmetz and Mirsalis, 1984; Mirsalis et al., 1983; Foureman et al., 1994; Matsuoka et al., 1996; Yoshikawa, 1996; Miyagawa et al., 1995; Mitchell and Caspary, 1987; Zimmermann et al., 1989; Honma et al., 1999; Kevekordes et al., 1999, 2001; Rossman et al., 1991; Sekihashi et al., 2002; Yoo, 1985; Demir et al., 2010; Scott et al., 2007; Yasunaga et al., 2004; Witt et al., 2000; Sasaki et al., 2000; Oda et al., 1978; Elmore and Fitzgerald, 1990; Longnecker et al., 1990; Galloway et al., 1987; Caspary et al., 1988; Rudd et al., 1983; Yoo, 1986; McGregor et al., 1988.

Literature Search and Risk Assessment Completed on: 05/06/14.

Repeated Dose Toxicity:

The margin of exposure for benzyl isobutyrate is adequate for the repeated dose toxicity endpoint at the current level of use.

Risk Assessment:

There are no repeated dose toxicity data on benzyl isobutyrate. Read across material benzyl acetate (CAS # 140-11-4; see Section 5) has numerous oral repeated dose toxicity studies conducted in rats and mice. A dietary 2-year chronic toxicity study conducted in rats determined a LOAEL for repeated dose toxicity of 3000 ppm, or 145 mg/kg/day for female rats, based on decreased body weights (NTP, 1993). The NOAEL was derived by dividing the LOAEL by a safety factor of 10, which is equal to 14.5 mg/kg/day. Therefore, the MOE is equal to the benzyl acetate NOAEL in mg/kg/day divided by the total systemic exposure, 14.5/0.015 or 967.

Additional References: McGinty et al., 2012a; Belsito et al., 2012; Longland et al., 1977; McGinty et al., 2012b; RIFM, 2013; RIFM, 1986b; RIFM, 1957; Abdo and Wenk, 1995; Abdo et al., 1998; Longnecker et al., 1986; Longnecker et al., 1990; Young, 1989; Abdo et al., 1985; Caldwell et al., 1987; Snapper et al., 1925; Hotchkiss et al., 1992a; Nasseri-Sina et al., 1992; Chidgey et al., 1986a; Grundschober, 1977; Miyashita and Robinson, 1980; Chidgey and Caldwell, 1986b, Chidgey et al., 1987; McMahon et al., 1989a; Augustinsson and Ekedahl, 1962; Clapp and Young, 1970; McMahon et al., 1989b; Schunk et al., 1986; RIFM, 1989a; Hotchkiss et al., 1992b, 1992c, Hotchkiss, 1998, Hotchkiss et al., 1992b; Caldwell et al., 1987; Hotchkiss, 1992c; Meyer, 1965; Garnett et al., 1994; Jimbo, 1983; Hotchkiss et al., 1988; Hotchkiss, 1989, 1990a, 1992d; Hotchkiss et al., 1990b; RIFM, 1989b; Hotchkiss et al., 1989; Hotchkiss et al., 1992d; McGinty et al., 2012b.

Literature Search and Risk Assessment Completed on: 04/29/14.

Developmental and Reproductive Toxicity:

The margin of exposure for benzyl isobutyrate is adequate for the developmental and reproductive toxicity endpoints at the current level of use.

Risk Assessment:

There are no developmental toxicity data on benzyl isobutyrate. Read across material benzyl acetate (CAS # 140-11-4; see Section 5) has a gavage developmental toxicity study that was conducted in rats. The NOAEL for developmental toxicity was determined to be 100 mg/kg/day, based on minor fetal internal anomalies and fetal weights (Ishiguro et al., 1993). Therefore, the MOE for developmental toxicity is equal to the benzyl acetate NOAEL in mg/kg/day divided by the total systemic exposure, 100/0.015 or 6667.

There are no reproductive toxicity data on benzyl isobutyrate. Read across material benzyl acetate (CAS # 140-11-4; see Section 5) has a gavage developmental toxicity study conducted in rats that determined the NOAEL for maternal toxicity to be 500 mg/kg/day, based on maternal body weight gain (Ishiguro et al., 1993). In 13-week dietary subchronic toxicity studies in rats and mice with benzyl acetate, sperm morphology and vaginal cytology examinations were evaluated (Morrissey et al., 1988). There were no effects on sperm parameters in mice or rats up to the high dosage of 7900 or 3900 mg/kg/day, respectively. There were no effects on estrous cycling in female rats up to the high dosage of 4500 mg/kg/day. Lengthening of the estrous cycle occurred in high-dose female mice (9400 mg/kg/day), which the authors concluded was related to decreases in body weight. These data indicate no specific concern for reproductive toxicity. Therefore, the MOE for reproductive toxicity is equal to the benzyl acetate NOAEL in mg/kg/day divided by the total systemic exposure, 500/0.015 or 33333.

RIFM's Expert Panel* and the adjunct Reproduction Advisory Group reviewed the Ishiguro et al. (1993) results and concluded that the minor fetal anomalies observed at the highest dose level were most likely a developmental delay. An increased fetal body weight may indicate an adverse effect. For example, chemicals which induce maternal diabetes mellitus may increase fetal weight. Early embryocidal effects leading to a reduced litter size may secondarily increase fetal weight. The Panel members and the Reproduction Advisory Group concluded that the increased fetal body weight observed in the 100 and 10 mg/kg/day groups was biologically insignificant and that no additional reproductive or developmental toxicity studies are needed. They concluded that the maternal NOAEL was 500 mg/kg/day based on weight gain and the fetal NOAEL is 100 mg/kg/day based on weight and internal organ malformations.

*RIFM's Expert Panel and adjunct Reproduction Advisory Group are composed of scientific and technical experts in their respective fields. These groups provide advice and guidance.

Additional References: McGinty et al., 2012a; Belsito et al., 2012; Longland et al., 1977; McGinty et al., 2012b; RIFM, 2013; RIFM, 1986b; RIFM, 1957; Abdo and Wenk, 1995, Abdo et al., 1998; Longnecker et al., 1986; Longnecker et al., 1990; Young, 1989; Abdo et al., 1985; Caldwell et al., 1987; Snapper et al., 1925; Hotchkiss et al., 1992a; Nasseri-Sina et al., 1992; Chidgey et al., 1986a; Grundschober, 1977; Miyashita and Robinson, 1980; Chidgey and Caldwell, 1986b; Chidgey et al., 1987; McMahon et al.,

1989a; Augustinsson and Ekedahl, 1962; Clapp and Young, 1970; McMahon et al., 1989b; Schunk et al., 1986; RIFM, 1989a; Hotchkiss et al., 1992b, 1992c, Hotchkiss, 1998; Hotchkiss et al., 1992b; Caldwell et al., 1987; Hotchkiss et al., 1992c; Meyer, 1965; Garnett et al., 1994; Jimbo, 1983; Hotchkiss et al., 1988; Hotchkiss, 1989, 1990a, 1992d; Hotchkiss et al., 1990b; RIFM, 1989b; Hotchkiss et al., 1989; Hotchkiss et al., 1992d; McGinty et al., 2012b.

Literature Search and Risk Assessment Completed on: 04/29/14.

Skin Sensitization:

Based on existing material specific data and read across to benzyl acetate (CAS # 140-11-4); benzyl isobutyrate does not present a concern for skin sensitization.

Risk Assessment:

Based on existing material specific data and read across to benzyl acetate (CAS # 140-11-4; see Section 5), benzyl isobutyrate does not present a concern for skin sensitization. The chemical structure of these materials indicates that they may have a low reactivity with skin proteins (Roberts et al., 2007; Toxtree 2.5.0; OECD toolbox v3.1). In guinea pig test methods no reactions indicative of sensitization were observed to benzyl acetate (RIFM, 1985a; RIFM, 1986a; RIFM, 1985b; Klecak, 1985). Additionally, no reactions indicative of skin sensitization were observed in both the human repeated insult patch test and the human maximization test to benzyl acetate (RIFM, 1975a; RIFM, 1975b; RIFM, 1975c; RIFM, 1975d; RIFM, 1975e; RIFM, 1988a; RIFM, 1988b; RIFM, 1988c; RIFM, 1988d; RIFM, 1961, RIFM, 1987). Finally, in the human maximization test no reactions indicative of sensitization were observed to benzyl isobutyrate (RIFM, 1971).

Additional References: None.

Literature Search and Risk Assessment Completed on: 05/05/14.

Phototoxicity/Photoallergenicity:

Based on UV absorption spectra, benzyl isobutyrate does not present a concern for phototoxicity or photoallergenicity.

Risk Assessment:

There are no phototoxicity studies available for benzyl isobutyrate in experimental models. UV absorption spectra indicate no absorption between 290 and 400 nm. Corresponding molar absorption coefficient is well below the benchmark of concern for phototoxicity and photoallergenicity, 1000 L mol^{-1} cm^{-1} (Henry et al., 2009). Based on lack of absorbance, benzyl isobutyrate does not present a concern for phototoxicity or photoallergenicity.

Additional References: None.

Literature Search and Risk Assessment Completed on: 08/21/15.

Local Respiratory Toxicity:

The margin of exposure for benzyl isobutyrate is adequate for the respiratory endpoint at the current level of use.

Risk Assessment:

The inhalation exposure estimated for combined exposure was considered along with toxicological data observed in the scientific literature to calculate the MOE from inhalation exposure when used in perfumery. A NOAEC of 10 ppm (61.4 mg/m^3) is reported for read across material, benzyl acetate (CAS # 140-11-4; see Section 5), for a 2 week acute inhalation study conducted in rats (RIFM, 2013).

At this level, higher levels of lactate dehydrogenase were noted in the bronchoalveolar lavage fluid. Although the authors did not consider these effects as adverse, for the purpose of estimating local respiratory toxicity MOE, the lower exposure dose (61.4 mg/m^3) was considered.

This NOAEC expressed in mg/kg lung weight/day is:

- (61.4 mg/m^3)/(1 m^3/1000 L) = 0.0614 mg/L
- Minute ventilation (MV) of 0.17 L/min for a Sprague–Dawley rat X duration of exposure of 360 minutes per day (min/day) (according to GLP study guidelines) = 61.2 L/d
- (0.0614 mg/L) (61.2 L/d) = 3.76 mg/d
- (3.76 mg/d)/(0.0016 kg lung weight of rat*) = 2350 mg/kg lw/day

Based on the IFRA survey results for hydroalcoholics, the 97.5th percentile was reported to be 0.71%. Assuming the same amount is used in all product types (fine fragrances, hair sprays, antiperspirants/deodorants, candles, aerosol air fresheners, and reed diffusers/heated oil plug-ins), the combined inhalation exposure would be 0.066 mg/day as calculated based on the IFRA survey results for the 97.5th percentile use in hydroalcoholics for a 60 kg individual using RIFM's 2-Box/MPPD *in silico* models. To compare this estimated exposure with the benzyl acetate NOAEC expressed in mg/kg lung weight/day this value is divided by 0.65 kg human lung weight (Carthew et al., 2009) to give 0.102 mg/kg lung weight/day resulting in a MOE of 23039 (i.e., [2350 mg/kg lw/day]/[0.102 mg/kg lung weight/day]).

The MOE is greater than 100. The material exposure by inhalation at 0.71% in a combination of the products noted above is deemed to be safe under the most conservative consumer exposure scenario.

*Phalen, R.F. Inhalation Studies. Foundations and Techniques, 2nd Ed. 2009. Published by, Informa Healthcare USA, Inc., New York, NY. Chapter 9, Animal Models, in section: “Comparative Physiology and Anatomy”, subsection, “Comparative Airway Anatomy.”

Additional References: RIFM, 1977; RIFM, 1997b; Silver, 1992; RIFM, 1997a; Isola et al., 2003a; Rogers et al., 2003; RIFM, 2003a; RIFM, 2003b; Isola et al., 2003b; Isola et al., 2004a, 2004b; Smith et al., 2004; Rogers et al., 2005; Randazzo et al., 2014; Vethanayagam et al., 2013.

Literature Search and Risk Assessment Completed on: 05/09/14

10.2. Environmental endpoint summary

Screening-Level Assessment:

A screening level risk assessment of benzyl isobutyrate was performed following the RIFM Environmental Framework (Salvito et al., 2002) which provides for 3 levels of screening for aquatic risk. In Tier 1, only the material's volume of use in a region, its log K_{ow} and molecular weight are needed to estimate a conservative risk quotient (RQ; Predicted Environmental Concentration/Predicted No Effect Concentration or PEC/PNEC). In Tier 1, a general QSAR for fish toxicity is used with a high uncertainty factor as discussed in Salvito et al., 2002. At Tier 2, the model ECOSAR (providing chemical class specific ecotoxicity estimates) is used and a lower uncertainty factor is applied. Finally, if needed, at Tier 3, measured biodegradation and ecotoxicity data are used to refine the RQ (again, with lower uncertainty factors applied to calculate the PNEC). Following the RIFM Environmental Framework, benzyl isobutyrate was identified as a fragrance material with the potential to present a possible risk to the aquatic environment (i.e., its screening level PEC/PNEC >1).

A screening-level hazard assessment using EPISUITE ver 4.1 did not identify benzyl isobutyrate as either being possibly persistent nor bioaccumulative based on its structure and physical–chemical properties. This screening level hazard assessment is a weight of evidence review of a material's physical–chemical properties, available data on environmental fate (e.g., OECD Guideline biodegradation studies or die-away studies) and fish bioaccumulation, and review of model outputs (e.g., USEPA's BIOWIN

and BCFBAF found in EPISUITE ver 4.1). Specific key data on biodegradation and fate and bioaccumulation are reported below and summarized in the Environmental Safety Assessment section prior to Section 1.

Risk Assessment:

Based on current volume of use (2011), benzyl isobutyrate presents a risk to the aquatic compartment in the screening level assessment.

Key Studies:

Biodegradation: No Data Available.

Ecotoxicity: No Data Available.

Other available data:

Benzyl isobutyrate has been pre-registered for REACH with no additional data at this time.

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)	EC50 (Daphnia)	EC50 (Algae)	AF	PNEC	Chemical Class
RIFM Framework Screening Level **(Tier 1)**	33.08 mg/L			1,000,000	0.0330 μg/L	
ECOSAR Acute Endpoints **(Tier 2)** ***Ver 1.11***	6.297 mg/L	11.77 mg/L	4.28 mg/L	10,000	0.4284 μg/L	Esters
ECOSAR Acute Endpoints **(Tier 2)** ***Ver 1.11***	18.92 mg/L	11.77 mg/L	12.79 mg/L			Neutral Organics SAR (baseline toxicity)

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

Exposure	Europe (EU)	North America (NA)
Log K_{ow} used	2.99	2.99
Biodegradation factor used	1	1
Dilution factor	3	3
Regional volume of use tonnage band	1–10	1–10
Risk characterization: PEC/PNEC	**<1**	**<1**

Based on available data, the RQ for this material is < 1. Additional assessment is not necessary.

The RIFM PNEC is 0.43 μg/L. The revised PEC/PNECs for EU and NA are < 1 and therefore, do not present a risk to the aquatic environment at the current reported volumes of use.

Literature Search and Risk Assessment Completed on: 04/21/14.

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/ntp_tox/index.cfm
- 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://www.chem.unep.ch/irptc/sids/oecdsids/sidspub.html
- **EPA Actor:** http://actor.epa.gov/actor/faces/ACToRHome.jsp;jsessionid=0EF5C212B7906229F477472A9A4D05B7
- **US EPA HPVIS:** http://www.epa.gov/hpv/hpvis/index.html
- **US EPA Robust Summary:** http://cfpub.epa.gov/hpv-s/
- **Japanese NITE:** http://www.safe.nite.go.jp/english/db.html
- **Japan Existing Chemical Data Base:** http://dra4.nihs.go.jp/mhlw_data/jsp/SearchPageENG.jsp
- **Google:** https://www.google.com/webhp?tab=ww&ei=KMSoUpiQK-arsQS324GwBg&ved=0CBQQ1S4

*Information sources outside of RIFM's database are noted as appropriate in the safety assessment.

This is not an exhaustive list.

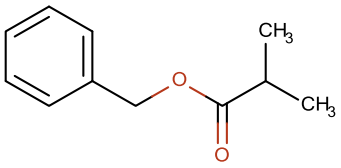
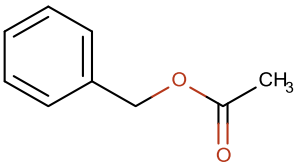
Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.fct.2016.04.027.

Transparency document

Transparency document related to this article can be found online at http://dx.doi.org/10.1016/j.fct.2016.04.027.

Appendix

	Target material	Read across material
Principal name	Benzyl isobutyrate	Benzyl acetate
CAS no.	103-28-6	140-11-4
Structure		
3D structure	http://www.thegoodscentscompany.com/opl/103-28-6.html	http://www.thegoodscentscompany.com/opl/140-11-4.html
Read-across endpoint		• Genotoxicity • Repeated dose • Devel/Repro • Skin sensitization • Respiratory
Molecular formula	$C_{11}H_{14}O_2$	$C_9H_{10}O_2$
Molecular weight	178.23	150.17
Melting point (°C)	10.84	−0.5
Boiling point (°C)	241.5	215.57
Vapor pressure (mmHg @ 25 °C)	0.0428	0.187
Log Kow (KOWWIN v1.68 estimate)	2.99	2.08
Water solubility (mg/L, @ 25 °C, WSKOW v1.42 estimate)	157.2	1605
J_{max} (mg/cm^2/h, RIFM SAM)	45.3	64.0
Henry's law (Pa·m^3/mol, Bond method)	2.53	1.43
Similarity (Tanimoto score)[1]		77%
Skin absorption		
Skin absorption percentage (SAM)	80%	80%
Genotoxicity		
DNA binding by OASIS v 1.1 QSAR Tool Box (3.1)	• No alerts	• Schiff base formers • Schiff base formers ≫ Direct acting Schiff base formers • Schiff base formers ≫ Direct acting Schiff base formers ≫ Specific Acetate Esters • SN1 • SN1 ≫ Carbenium ion formation • SN1 ≫ Carbenium ion formation ≫ Specific Acetate Esters • SN2 • SN2 ≫ Acylating agents • SN2 ≫ Acylating agents ≫ Specific Acetate Esters • SN2 ≫ SN2 at sp3-carbon atom • SN2 ≫ SN2 at sp3-carbon atom ≫ Specific Acetate Esters
DNA binding OECD	• Michael addition • Michael addition ≫ P450 Mediated Activation to Quinones and Quinone-type Chemicals • Michael addition ≫ P450 Mediated Activation to Quinones and Quinone-type Chemicals ≫ Arenes	• Michael addition • Michael addition ≫ P450 Mediated Activation to Quinones and Quinone-type Chemicals • Michael addition ≫ P450 Mediated Activation to Quinones and Quinone-type Chemicals ≫ Arenes
Carcinogenicity (genotox and non-genotox) alerts by ISS	• No alerts	• No alerts
DNA alerts for Ames, MN, CA by OASIS v 1.1	• No alerts	• Schiff base formers • Schiff base formers ≫ Direct acting Schiff base formers • Schiff base formers ≫ Direct acting Schiff base formers ≫ Specific Acetate Esters • SN1 • SN1 ≫ Carbenium ion formation • SN1 ≫ Carbenium ion formation ≫ Specific Acetate Esters • SN2 • SN2 ≫ Acylating agents • SN2 ≫ Acylating agents ≫ Specific Acetate Esters • SN2 ≫ SN2 at sp3-carbon atom • SN2 ≫ SN2 at sp3-carbon atom ≫ Specific Acetate Esters
In-vitro mutagenicity (Ames test) alerts by ISS	• No alerts	• No alerts
In-vivo mutagenicity (Micronucleus) alerts by ISS	• H-acceptor-path3-H-acceptor	• H-acceptor-path3-H-acceptor
Oncologic classification	• Not classified	• Not classified
Repeated dose toxicity		
Repeated dose (HESS)	• Not categorized	• Not categorized
Developmental toxicity		
	• Non toxicant (low reliability)	• Toxicant (medium reliability)

(*continued*)

	Target material	Read across material
Developmental toxicity model by CAESAR v2.1.6		
ER binding by OECD QSAR toolbox (3.1)	• Non binder	• Non binder
Skin sensitization		
Protein binding by OASIS v1.1	• SN2 • SN2 ≫ Nucleophilic substitution at sp3 Carbon atom • SN2 ≫ Nucleophilic substitution at sp3 Carbon atom ≫ Activated alkyl esters	• SN2 • SN2 ≫ Nucleophilic substitution at sp3 Carbon atom • SN2 ≫ Nucleophilic substitution at sp3 Carbon atom ≫ Activated alkyl esters
Protein binding by OECD	• SN2 • SN2 ≫ SN2 reaction at sp3 carbon atom • SN2 ≫ SN2 reaction at sp3 carbon atom ≫ Allyl acetates and related chemicals	• SN2 • SN2 ≫ SN2 reaction at sp3 carbon atom • SN2 ≫ SN2 reaction at sp3 carbon atom ≫ Allyl acetates and related chemicals
Protein binding potency	• Not possible to quantify	• Not possible to quantify
Protein binding alerts for skin sensitization by OASIS v1.1	• SN2 • SN2 ≫ Nucleophilic substitution at sp3 Carbon atom • SN2 ≫ Nucleophilic substitution at sp3 Carbon atom ≫ Activated alkyl esters	• SN2 • SN2 ≫ Nucleophilic substitution at sp3 Carbon atom • SN2 ≫ Nucleophilic substitution at sp3 Carbon atom ≫ Activated alkyl esters
Skin sensitization model (CAESAR) (version 2.1.5)	• Sensitizer (low reliability)	• Sensitizer (low reliability)
Metabolism		
OECD QSAR toolbox (3.1) **Rat liver S9 metabolism simulator**	See Supplemental data 1	See Supplemental data 2

[1] Values calculated using JChem with FCFP4 1024 bits fingerprint (Rogers and Hahn, 2010).

Summary

Benzyl isobutyrate (RIFM# 227, CAS# 103-28-6) lacks toxicity data for the skin absorption, genotoxicity, repeated dose, developmental, reproductive, skin sensitization, respiratory and phototoxicity/photoallergenicity end points. Hence, *in silico* evaluation was conducted by determining suitable read-across analogs for this material. Based on structural similarity, reactivity, metabolism data, physicochemical properties and expert judgment, benzyl acetate (RIFM# 106, CAS# 140-11-4) was identified as proper read across for their respective toxicity endpoints.

Methods

- The identified read-across analogs were confirmed by using expert judgment
- The physicochemical properties of target and analogs were calculated using EPI Suite™ v4.11 developed by US EPA (USEPA, 2012)
- The J_{max} were calculated using RIFM skin absorption model (SAM), the parameters were calculated using consensus model (Shen et al., 2014)
- DNA binding, mutagenicity, genotoxicity alerts and oncologic classification were estimated using OECD QSAR Toolbox (v3.1) (OECD, 2012)
- ER binding and repeat dose categorization were estimated using OECD QSAR Toolbox (v3.1) (OECD, 2012)
- Developmental toxicity and skin sensitization were estimated using CAESAR (v.2.1.6) (Cassano et al., 2010)
- Protein binding were estimated using OECD QSAR Toolbox (v3.1) (OECD, 2012)
- The major metabolites for the target and read-across analogs were determined and evaluated using OECD QSAR Toolbox (v3.1) (OECD, 2012)

Conclusion/rationale

- Benzyl acetate (analog) was used as a read-across for benzyl isobutyrate (target) based on:
  - The target and analog belong to the generic class of aromatic esters, specifically, esters/aryl alkyl alcohol simple acid/benzylic alcohol.
  - The target and analog have similar carboxylic acid part and the same alcohol part.
  - The only difference is that the target is an isobutyrate while the analog is an acetate. The difference between structures does not essentially change the physicochemical properties nor raise any additional structural alerts and therefore, their toxicology profiles are expected to be similar.
  - The analog shows more alerts for DNA binding. This is favorable since it indicates that we are reading from a potentially more reactive to a less reactive species. They show similar alerts for mutagenicity, genotoxicity and oncologic classification.
  - The target and analog show similar alerts for Repeated Dose (HESS) Categorization and ER Binding. ER Binding is molecular initiating event analogous to protein binding. ER binding is not necessarily predictive of endocrine disruption given the complex pre- and post-receptor events that determine activity.
  - The target and analog show similar alerts for protein binding.
  - The target and analog are expected to be metabolized similarly. As per the OECD Toolbox they are predicted to have similar metabolites.

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