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Review

A toxicologic and dermatologic assessment of cyclic acetates when used as fragrance ingredients ☆

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ABSTRACT

An evaluation and review of a structurally related group of fragrance materials.

Contents

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Introduction

This report summarizes safety data relevant to the risk assessment of the use of some cyclic acetates as fragrance ingredients. These esters are used in decorative cosmetics, fine fragrances, shampoos, toilet soaps and other toiletries as well as in non-cosmetic products such as household cleaners and detergents. This report summarizes animal and human data, including studies with various exposure routes, and evaluates the risk from their use as fragrance ingredients. The scientific evaluation focuses on dermal exposure, which is considered to be the primary exposure route for these fragrance materials. Toxicity, metabolism and kinetic data obtained from studies using other routes of exposure have also been considered to assess the systemic fate and toxicity of the substances.

The selected data from published and unpublished reports were deemed to be relevant based on the nature of the protocols, quality of the data, and appropriate exposure. These data are presented in tabular form.

1. Chemical identity, regulatory status and exposure

The International Joint FAO/WHO Expert Committee on Food Additives (JECFA) has evaluated alicyclic ketones, secondary alcohols and related esters including cyclohexyl acetate, a member of this group summary, as flavouring agents in food. These materials were judged by this Committee not to present a safety concern at the current estimated intake levels (JECFA, 2007).

1.1. Rationale for grouping cyclic acetates together

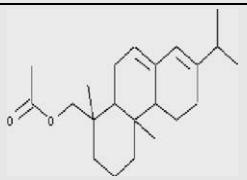
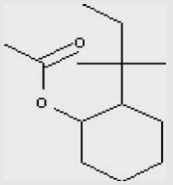
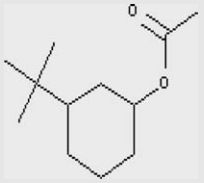
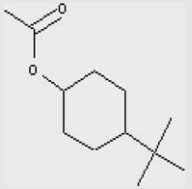
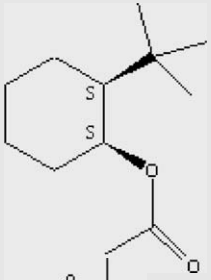
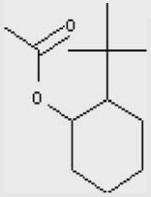
The common characteristic structural element of cyclic acetates is the acetate unit bound to a mono-, bi- or tri-cyclic alcohol. The present group comprises 25 substances which include 15 esters of monocyclic alcohols, three of bicyclic alcohols, and seven of tricyclic alcohols.

The only substituents at the alcohol moiety are alkyl groups. Some esters contain cyclic or exocyclic double bonds or a terminal triple bond. Although data on metabolism are lacking and the toxicity of only a few compounds has been studied, cyclic acetates are assumed to be rapidly hydrolyzed to the alcohol and the carboxylic acid by carboxylesterases. The local and systemic toxicity of the compounds under review is exerted either by the parent compound or by the hydrolysis products. This hydrolysis is on the one hand a detoxification step. It transforms the poorly water-soluble ester into an alcohol, which can subsequently be conjugated and excreted faster than the parent compound, thereby diminishing the potential toxicity of the ester itself. On the other hand, the resulting acid (in this case acetic acid for all but one substance) may cause irritation due to a pH-shift in tissues where hydrolysis initially occurs. Systemic toxicity from acetic acid – an endogenous compound – is very low (SCOEL, 2001) and can be excluded under the conditions of the use of the esters in cosmetic products. As hydrolysis by carboxylesterases might be less efficient for esters with bulky alcohols, it can be expected that the rate of acetate hydrolysis would decline in the order of mono- > bi- > tri-cyclic alcohols. Therefore, systemic toxicity is expected to be inversely related to the hydrolysis rate with acetates of tricyclic alcohols being potentially most toxic.

The toxicity database for the compounds under review is limited. The primary, secondary, and tertiary cyclic alcohols, which are formed from the cyclic acetates, are expected to be of low systemic toxicity. This assumption is based on the five oral repeated dose studies available of which two were performed with cyclic acetates (4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate and 4-*tert*-butylcyclohexyl acetate) and three with metabolites of cyclohexyl acetate and 4-*tert*-butylcyclohexyl acetate (cyclohexanone (2) and 4-*tert*-butyl-cyclohexanol (1), respectively). Results of these repeated dose studies show that the substances are of low systemic toxicity as assumed from the chemical structures. Additionally, the structurally related *cis*- and *trans*-*para*-1(7),8-menthadien-2-yl acetate, 3,3,5-trimethylcyclohexanol, 2-, 3-, and 4-methylcyclohexanone and 2-*sec*-butylcyclohexanone were evaluated by the Joint FAO/WHO Expert Committee on Food Additives and shown to be of no safety concern when used as flavouring agents in food (JECFA, 2007). The study with repeated dermal application of a tricyclic alcohol acetate (4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate) should be regarded according to the considerations outlined above as a worst-case example for systemic toxicity of the esters under review.

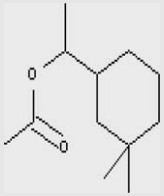
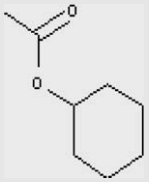
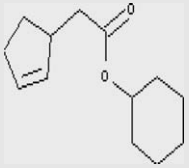
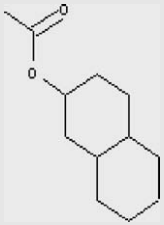
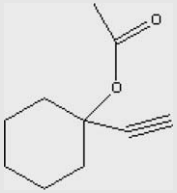
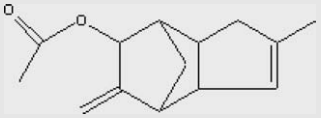
Some of the esters possess cyclic or exocyclic double bonds or a terminal triple bond. The triple bond of 1-ethynylcyclohexyl acetate is not expected to be metabolized to an epoxide; however, the genotoxicity of this compound has not been tested. The esters that contain carbon–carbon double bonds might give rise to potentially mutagenic or carcinogenic epoxides during metabolism. Several of these compounds have been tested and found to be not mutagenic in the Ames test. This indicates that the metabolism to epoxides or the mutagenic potential of the formed epoxides is

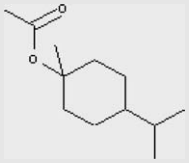
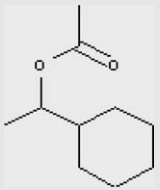
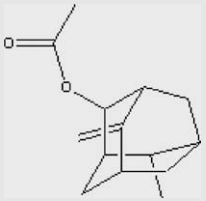
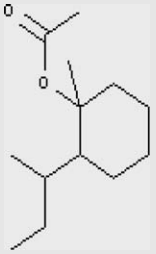
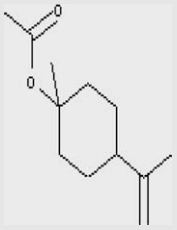
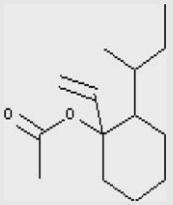
Table 1
Material identification and summary of volume of use and dermal exposure

Material	Synonyms	Structure	Worldwide metric tons	Dermal systemic exposure in cosmetic products (mg/kg/day)	Maximum skin level[a] (%)
Abietyl acetate CAS # 54200-50-9 Log K_{ow} (calculated): 7.24 Molecular weight: 330.12	1*R*-(1.α.,4a.β.,4b.α.,10a.α.)]-1,2,3,4,4a,4b,5,6,10,10a-Decahydro-7-isopropyl-1,4a-dimethylphenanthren-1-methanol acetate; 1-phenanthrenemethanol, 1,2,3,4,4a,4b,5,6,10,10a-decahydro-1,4a-dimethyl-7-(1-methylethyl)-, acetate, [1*R*-(1.alpha)]		0.1–1	0.0089	0.08
Amylcyclohexyl acetate (mixed isomers) CAS # 67874-72-0 Log K_{ow} (calculated): 4.91 Molecular weight: 212.33	2-*t*-Amylcyclohexyl acetate; cyclohexanol, 2-(1,1-dimethylpropyl)-, acetate; 2-*tert*-pentylcyclohexyl acetate		10–100	0.0087	0.06
3-tert-Butylcyclohexyl acetate CAS # 31846-06-7 Log K_{ow} (calculated): 4.42 Molecular weight: 198.31	Cyclohexanol, 3-(1,1-dimethylethyl)-, acetate		0.1–1	0.0008	0.004
4-tert-Butylcyclohexyl acetate CAS # 32210-23-4 Log K_{ow} (measured): 4.48 Log K_{ow} (calculated): 4.42 Molecular weight: 198.31	Acetic acid, *p-tert*-butylcyclohexyl ester; *p-tert*-butylcyclohexyl acetate; 4-*tert*-butylhexahydrophenyl acetate; Oryclon; PTBCHA (*para-tertiary*-butyl-cyclo-hexyl-acetate); Vertenex		>1000	0.175	3.33
cis-2-tert-Butylcyclohexyl acetate CAS # 20298-69-5 Log K_{ow} (calculated): 4.42 Molecular weight: 198.31	Cyclohexanol, 2-(1,1-dimethylethyl)-, acetate, *cis*-		10–100	0.122	1.94
2-tert-Butylcyclohexyl acetate CAS # 88-41-5 Log K_{ow} (calculated): 4.42 Log K_{ow} (measured) (OECD 117): 4.7 at 25 °C trans isomer Log K_{ow} (measured) (OECD 117): 4.8 at 25 °C *cis*-isomer Molecular weight: 198.31	1-Acetoxy-2-*t*-butylcyclohexane; Agrumex; Beldox; 2-*t*-Butylcyclohexanol acetate; *o-tert*-butylcyclohexyl acetate; 2-*t*-butylcyclohexyl acetate; cyclohexanol, 2-(1,1-dimethylethyl)-, acetate; 2-(1,1-dimethylethyl)cyclohexyl acetate; ortho tertiary butyl cyclohexyl acetate; Verdox		>1000	0.138	3.56

(*continued on next page*)

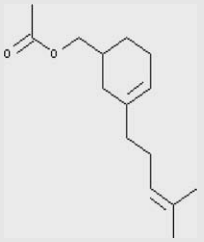
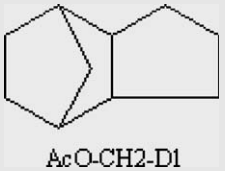
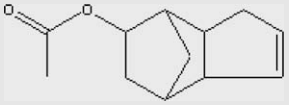
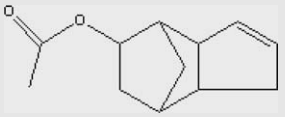
Table 1 (*continued*)

Material	Synonyms	Structure	Worldwide metric tons	Dermal systemic exposure in cosmetic products (mg/kg/day)	Maximum skin level[a] (%)
d-Cyclocitronellene acetate CAS # 25225-10-9 Log K_{ow} (calculated): 4.42 Molecular weight: 198.31	Cyclohexanemethanol, .α.,3,3-trimethyl-, acetate; α,3,3-trimethylcyclohexylmethyl acetate		10–100	0.246	0.39
Cyclohexyl acetate CAS # 622-45-7 Log K_{ow} (calculated): 2.64 Molecular weight: 142.2	Acetic acid, cyclohexyl ester; cyclohexane acetate		<0.1	0.0003	0.001
Cyclohexyl cyclopent-2-ene-1-acetate CAS # 65405-69-8 Log K_{ow} (calculated): 4.7 Molecular weight: 208.01	2-Cyclopentene-1-acetic acid, cyclohexyl ester		0.1–1	0.0003	0.006
Decahydro-beta-naphthyl acetate CAS # 10519-11-6 Log K_{ow} (calculated): 3.66 Molecular weight: 196.29	Decahydro-2-naphthyl acetate; 2-naphthalenol, decahydro-, acetate		1–10	0.0168	0.17
1-Ethynylcyclohexyl acetate CAS # 5240-32-4 Log K_{ow} (calculated): 2.82 Molecular weight: 166.22	1-Acetoxy-1-ethynylcyclohexane; cyclohexanol, 1-ethynyl-, acetate; Herbacet # 1		1–10	0.0025	0.005
(3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-Hexahydro-2-methyl-5-methylene-4,7-methano-1H-inden-6-yl acetate CAS # 81836-13-7 Log K_{ow} (calculated): 3.81 Molecular weight: 218.96	4,7-Methano-1*H*-inden-6-ol, 3a,4,5,6,7,7a-hexahydro-2-methyl-5-methylene-, acetate, (3a.α.,4.α.,6.α.,7.a		<0.1	0.0005*	0.02

4-(Isopropyl)-1-methylcyclohexyl acetate CAS # 26252-11-9 Log K_{ow} (calculated): 4.42 Molecular weight: 198.06	1-Acetoxy-*p*-menthane; cyclohexanol, 1-methyl-4-(1-methylethyl)-, acetate; dihydro-.β.-terpinyl acetate; *p*-menthan-1-ol, acetate; (±)-*p*-menthan-1-ol, acetate		10–100	0.0005*	0.02*
Alpha-methylcyclohexylmethyl acetate CAS # 13487-27-9 Log K_{ow} (calculated): 3.55 Molecular weight: 170.52	Cyclohexanemethanol, .α.-methyl-, acetate; cyclohexylmethylcarbinyl acetate		1–10	0.076	0.55
4-Methyl-8-methylenetricyclo[3.3.1.(3,7)]decan-2-yl acetate CAS # 122760-85-4 Log K_{ow} (calculated): 4.23 Molecular weight: 220.12	Prismylate; tricyclo[3.3.1(3,7)]decan-2-ol, 4-methyl-8-methylene-, acetate		<0.1	0.0005*	0.02
1-Methyl-2-(1-methylpropyl)cyclohexyl acetate CAS # 72183-75-6 Log K_{ow} (calculated) :4.91 Molecular weight: 212.33	Cyclohexanol, 1-methyl-2-(1-methylpropyl)-, acetate		0.1–1	0.041	0.35
1-Methyl-4-(1-methylvinyl)cyclohexyl acetate CAS # 10198-23-9 Log K_{ow} (calculated): 4.42 Molecular weight: 196.9	Cyclohexanol, 1-methyl-4-(1-methylethenyl)-, acetate; β.-terpinyl acetate		1–10	0.0005*	0.02
2-(1-Methylpropyl)-1-vinylcyclohexyl acetate CAS # 37172-02-4 Log K_{ow} (calculated): 5.27 Molecular weight: 224.44	Cyclohexanol, 1-ethenyl-2-(1-methylpropyl)-, acetate; dihydro abmrate		0.1–1	0.079	1.09

(*continued on next page*)

Table 1 (*continued*)

Material	Synonyms	Structure	Worldwide metric tons	Dermal systemic exposure in cosmetic products (mg/kg/day)	Maximum skin level[a] (%)
Myraldyl acetate CAS # 72403-67-9 Log K_{ow} (calculated): 5.76 Log K_{ow} (measured) (OECD 117: 5.6 and 5.7 at 30 °C Molecular weight: 236.36	3-Cyclohexene-1-methanol, 3(or 4)-(4-pentenyl)-, acetate; 4(or 3)-(4-methyl-3-pentenyl)-3-cyclohexenylmethyl acetate and isomers; 3(or 4)-(4-methylpenten-3-yl)cyclohex-3-ene-1-methyl acetate		1–10	0.186	2.12
Octahydro-4,7-methano-1H-indenemethyl acetate CAS # 30772-69-1 Log K_{ow} (calculated): 3.9 Molecular weight: 208.01	4,7-Methano-1*H*-indenemethanol, octahydro-, acetate		1–10	0.043	0.4
Tricyclodecanyl acetate CAS # 64001-15-6 Log K_{ow} (calculated): 3.4 Molecular weight: 194.27	Dihydrocyclacet; 4,7-methano-1*H*-inden-5-ol, octahydro-, acetate; octahydro-4,7-methano-1*H*-indene-5-yl acetate		10–100	0.002	0.16
Tricyclodecenyl acetate CAS # 5413-60-5 Log K_{ow} (calculated): 3.19 Log K_{ow} (measured) (OECD 117): 3.8 at 25 °C Molecular weight: 192.26	Dihydro-norbicyclopentadienyl acetate; greenylacetate; Herbaflorat; 3a,4,5,6,7,7a-hexahydro-4,7-methanoinden-6-ylacetate; jasmacyclene; 4,7-methano-1*H*-inden-6-ol, 3a,4,5,6,7,7a-hexahydro-, acetate; tricyclodecen-4-yl 8-acetate; verdyl acetate		100–1000	0.112	1.18
Tricyclo[5.2.1.02.6]dec-4-en-8-yl acetate CAS # 2500-83-6 Log K_{ow} (calculated): 3.19 Molecular weight: 192.26	3a,4,5,6,7,7a-Hexahydro-4,7-methano-1*H*-inden-5-yl acetate; 4,7-methano-1*H*-inden-5-ol, 3a,4,5,6,7,7a-hexahydro-, acetate		100–1000	0.143	1.38

1,7,7-Trimethylbicyclo[4.4.0]dec-3-yl acetate CAS # 24238-95-7 Log K_{ow} (calculated): 4.99 Molecular weight: 238.37	Decahydro-5,5,8a-trimethyl-2-naphthyl acetate; 2-naphthalenol, decahydro-5,5,8a-trimethyl-, acetate; 2-naphthol, decahydro-5,5,8a-trimethyl-, acetate; polywood; 1,7,7-trimethylbicyclo[4.4.0]decan-3-yl acetate	0.1–1	0.0005*	0.02
1,3,3-Trimethyl-2-norbornanyl acetate CAS # 13851-11-1 Log K_{ow} (calculated): 3.86 Molecular weight: 196.29	Bicyclo[2.2.1]heptan-2-ol, 1,3,3-trimethyl-, acetate; 3,3-dimethyl-8,9-dinorbornan-2-yl acetate; fenchyl acetate	10–100	0.0003	0.011

[a] The maximum skin levels are based on the assumption that the fragrance mixture is used at 20% in a consumer product (IFRA use level survey).
[*] A default value of 0.02% was used to calculate dermal systemic exposure.

low. It remains unclear whether this also applies to those compounds not tested for mutagenicity (1-methyl-4-(1-methylvinyl)cyclohexyl acetate, cyclohexyl cyclopent-2-ene-1-acetate, tricyclo[5.2.1.$0^{2,6}$]dec-4-en-8-yl acetate, (3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate). These esters as well as 1-ethynylcyclohexyl acetate are exempt from this category evaluation until proved to be non-mutagenic.

It remains unclear whether acetic acid or the parent substance is responsible for the irritation noted, although there is limited evidence that acetic acid is primarily responsible for the irritation. One of the cyclic acetates (cyclohexyl cyclopent-2-ene-1-acetate) is hydrolyzed to cyclopent-2-enyl acetic acid; data on irritation of this acetate or the hydrolyzed acid are not available. As acidity of organic acids decreases with increasing length of the alkyl chain, it is expected that the hydrolyzed acid is less acidic than acetic acid.

Skin irritation studies in humans are available for 17 esters, and yield non-irritating concentrations. Most of the compounds are, however, irritants, when tested undiluted in animal experiments.

In conclusion, for the purpose of risk assessment local irritation is considered to be the predominant and most sensitive effect of this group of compounds, whereas systemic effects are expected, if at all, at higher doses only.

All data available for the cyclic acetates evaluated are summarized in Tables 2–10. CAS-No., synonyms, structural formulae and exposure data for the esters considered in this review are shown in Table 1.

1.2. Occurrence and use

Three of the cyclic acetates have been detected in fruits, vegetables, herbs and nuts (TNO, 2008).

The annual worldwide production of the individual cyclic acetates varies greatly and ranges from <0.1 to 100 metric tons for most of the compounds. The worldwide production of tricyclodecenyl acetate and tricyclo[5.2.1.$0^{2,6}$]dec-4-en-8-yl acetate is 100–1000 metric tons per year; 4-*tert*-butylcyclohexyl acetate and 2-*tert*-butylcyclohexyl acetate are produced at >1000 metric tons per year (IFRA, 2004) (Table 1).

1.3. Estimated consumer exposure

Potential consumer exposure to fragrance ingredients occurs through the dermal and inhalation routes of exposure and may occur also by other routes when the compounds have other uses. Data on inhalation exposure during use of the substances are not available. It is assumed that the main exposure route will be dermal.

The availability of fragrance ingredients for potential exposure by consumers is estimated in two ways (see Table 1). One estimates potential percutaneous absorption over the entire body due to the use of many different fragranced products. The other estimates potential dermal exposure due to the use of products, such as fine fragrances, that usually contain higher concentrations and are used on smaller localized skin sites. Potential skin exposure to cyclic acetates is estimated based on their concentrations in 10 types of cosmetic products (body lotion, face cream, eau de toilette, fragrance cream, anti-perspirant, shampoo, bath products, shower gel, toilet soap and hair spray). The concentration data in the 10 product types were multiplied by the amount of product applied, the number of applications/ day for each product type, and a "retention factor" (ranging from 0.01–1.0) to account for the length of time a product may remain on the skin and/or the likelihood of it being removed by washing. The value produced represents the maximum skin concentration associated with each product type. As a conservative measure, the total maximum skin concentration

Table 2-1
Acute dermal toxicity studies

Material	Species	No. animals/dose/group	LD_{50}[a] (mg/kg)	References
Amylcyclohexyl acetate (mixed isomers)	Rabbit	10	>5000	RIFM (1974a)
2-*tert*-Butylcyclohexyl acetate	Rabbit	10	>5000	RIFM (1976a)
4-*tert*-Butylcyclohexyl acetate	Rabbit	4	>5000	RIFM (1976b)
	Rabbit	10	>5000	RIFM (1976a)
	Rabbit	6 (3/Sex)	>~4700	RIFM (1979a)
d-Cyclocitronellene acetate	Rabbit	10	>5000	RIFM (1982a)
Cyclohexyl acetate	Rabbit	10	>5000	RIFM (1977a)
	Rabbit	4	>5000	RIFM (1974b)
	Rabbit	4 (Male)	10,100 (95% C.I. 6170–16,500 mg/kg)	Carpenter et al. (1974)
Decahydro-beta-naphthyl acetate	Rabbit	8	>5000	RIFM (1976a)
(3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-Hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate	Rabbit	6	>2000	RIFM (1982b)
Alpha-methylcyclohexylmethyl acetate	Rabbit	1 or 2	>7940	RIFM (1977b)
4-Methyl-8-methylene-tricyclo[3.3.1.1^{3,7}]decan-2-yl acetate	Rat	10 (5/Sex)	>2000	RIFM (1989a)
Octahydro-4,7-methano-1*H*-indenemethyl acetate	Rabbit	4 (2/Sex)	>3000	RIFM (1976c)
Tricyclodecenyl acetate	Rabbit	10	>5000	RIFM (1977a)
	Rabbit	10	>5000	RIFM (1974a)
1,7,7-Trimethylbicyclo[4.4.0]dec-3-yl acetate	Rabbit	6 (3/Sex)	>2000	RIFM (1979b)
1,3,3-Trimethyl-2-norbornanyl acetate	Rabbit	10	>5000	RIFM (1975a)

[a] Units have been converted to make easier comparisons; original units are in the Fragrance Material Reviews.

Table 2-2
Acute oral toxicity studies

Material	Species	No. animals/dose/group	LD_{50}[a] (mg/kg)	References
Amylcyclohexyl acetate (mixed isomers)	Rat	10	>5000	RIFM (1974a)
	Mouse	10 (5/Sex)	Males: 4034 (95% C.I. 1672–48,757 mg/kg) Females: 4738 (3600–6236) Combined: 4388 (1926–8564)	RIFM (1978a)
2-*tert*-Butylcyclohexyl acetate	Rat	10	4600 (95% C.I. 2700–7800 mg/kg)	RIFM (1976a)
	Mouse	6	310 (95% C.I. 300–450 mg/kg)	RIFM (1978b)
	Mouse	2 or 6	~500	RIFM (1981a)
	Mouse	6	350 (95% C.I. 0.0–580 mg/kg)	RIFM (1977c)
4-*tert*-Butylcyclohexyl acetate	Rat	10	>500, <5000	RIFM (1976b)
	Rat	10	~5000	RIFM (1976a)
	Rat	16 (8/Sex)	Males: 4150 (95% C.I. 3520–4900 mg/kg) Females: 3550 (2930–4300) Combined: 3600 (3170–4090)	RIFM (1979c)
	Mouse	6	~4300 (95% C.I. 2100–6000 mg/kg)	RIFM (1978c)
	Mouse	2, 6, 2	~5000	RIFM (1980a)
d-Cyclocitronellene acetate	Rat	10	>5000	RIFM (1982a)
	Rat	4 (2/Sex)	~8000	RIFM (1981b)
Cyclohexyl acetate	Rat	10	>5000	RIFM (1977a)
	Rat	10	>5000	RIFM (1974b)
	Rat	5 (Males)	6730 (95% C.I. 3790–11,900 mg/kg)	Carpenter et al. (1974)
Decahydro-beta-naphthyl acetate	Rat	10	>5000	RIFM (1976a)
(3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-Hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate	Rat	10 (Males)	>5000	RIFM (1982d)
4-(Isopropyl)-1-methylcyclohexyl acetate	Rat	10 (5/Sex)	>5000	RIFM (1984a)
Alpha-methylcyclohexylmethyl acetate	Rat	5	8200 (95% C.I. 7300–9270 mg/kg)	RIFM (1977b)
4-Methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate	Rat	4 (2/Sex)	>2000	RIFM (1989b)
	Rat	10 (5/Sex)	>5000	RIFM (1989b)
2-(1-Methylpropyl)-1-vinylcyclohexyl acetate	Rat	10 (5/Sex)	>5100	RIFM (1977d)
	Mouse	10	>8000	RIFM (1972a)
Myraldyl acetate	Mouse	10	>4000, <8000	RIFM (1972b)
Octahydro-4,7-methano-1*H*-indenemethyl acetate	Rat	4 (2/Sex)	3362 ± 639.9	RIFM (1976c)
Tricyclodecenyl acetate	Rat	10	>5000	RIFM (1977a)
	Rat	10	>5000	RIFM (1974a)
	Rat	10 (Female)	~4300	RIFM (1958)
	Mouse	2, 6, 2	~5400	RIFM (1980b)
	Mouse	2, 6, 2	~10,800	RIFM (1976d)
1,7,7-Trimethylbicyclo[4.4.0]dec-3-yl acetate	Rat	10 (5/Sex)	>5000	RIFM (1979d)
1,3,3-Trimethyl-2-norbornanyl acetate	Rat	10	>5000	RIFM (1975a)

[a] Units have been converted to make easier comparisons; original units are in the Fragrance Material Reviews.

Table 2-3
Acute miscellaneous toxicity studies

Material	Dose route	Species	No. animals/dose group	LD_{50} and/or clinical signs[a]	References
4-*tert*-Butylcyclohexyl acetate	I.P. injection (water emulsion with traganth)	Mouse	Not reported	400 mm^3/kg (~0.4 mg/kg)	RIFM (1970a)
	Inhalation, exposure to a saturated vapor for 8 h	Rat	12	4800 mm^3/kg (~4.8 mg/kg) No deaths and no lesions at necropsy	RIFM (1970a)
Cyclohexyl acetate	Inhalation, exposure to a vapor for 8 h	Rat	6	No deaths	Carpenter et al. (1974)
Alpha-methylcyclohexylmethyl acetate	Inhalation, 6 h to air passed through test material	Rat	6	No deaths at 0.9 mg/l	RIFM (1977b)
2-(1-Methylpropyl)-1-vinylcyclohexyl acetate	I.P. injection (Gum arabic emulsion)	Mouse	10	24 h: 4000 mg/kg (95% C.I. 2000–8000) 10 days: 1000–2000 mg/kg	RIFM (1972a)
Myraldyl acetate	I.P. injection (gum Arabic emulsion)	Mouse	10	>2000 mg/kg, <4000 mg/kg	RIFM (1972b)

[a] Units have been converted to make easier comparisons; original units are in the Fragrance Material Reviews.

Table 3
Repeated dose toxicity studies

Material	Method	Dose[a]	Species (No./dose group)	Results	References
4-Methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate	Oral (gavage), 28 d	0, 5, 55 or 1000 mg/kg bw/day In corn oil	Rat (5/sex/dose)	5 mg/kg bw/day: NOEL ⩾55 mg/kg bw/day: NOAEL: salivation ↑, blood urea nitrogen ↑ (f); 1000 mg/kg bw/day: red staining around eyes (1m), head and shoulders, hunched posture; body-weight gain ↓ (f) weeks 2–4 (stat. sign. at week 4); neutrophil counts ↑ (stat. sign. (m)); changes in some clinical chemistry parameters (protein and globulin ↑, blood urea nitrogen ↑ (m), glucose ↓ (m), inorganic phosphorous ↓ (m)); rel. liver weights ↑, rel. kidney weights ↑ (m), enlarged liver (m and 1f), minimal centrilobular hepatocytes enlargement (1m), moderate degree of eosinophilic inclusion in the cytoplasm of cortical epithelium of kidneys (m); testes atrophy in 1 male	RIFM (1990)

NOEL: no observed effect level, NOAEL: no observed adverse effect level.
m: male, f: female.
bw: Body weight.
↑ Increased, ↓ decreased.
[a] Units have been converted to make easier comparisons; original units are in the Fragrance Material Reviews.

was calculated to be the sum of the maximum skin concentrations for each of the 10 product categories.

The maximum skin exposure levels of the cyclic acetates that form part of the formulae of fine fragrances vary widely and have been reported to range from 0.0008% to 3.6%. For consideration of potential sensitization, the exposure is calculated as the percent concentration applied to the skin. Exposure to cyclic acetates used in fine fragrance products is calculated based on the use of 20% of the fragrance mixture (the maximum used) in the fine fragrance consumer product (IFRA, 2007). The calculated exposures for cyclic acetates used in cosmetic products are listed in Table 1. Exaggerated maximum daily exposures on the skin range from 0.0003 to 1.94 mg/kg body weight (bw)/day for the individual esters acetates in high end users of cosmetic products containing these materials (see Table 1).

Exposure data for five fragrance materials (1,7,7-trimethylbicyclo[4.4.0]dec-3-yl acetate; 1-methyl-4-(1-methylvinyl)cyclohexyl acetate; 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate; 4-(isopropyl)-1-methylcyclohexyl acetate; (3a.α.,4.α.,6.α., 7.α.,7a.α.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate) were not reported. A default value of 0.02% is used to calculate the maximum daily exposure on the skin which is 0.0005 mg/kg bw for high end users of these products.

Maximum skin exposure data (the total of the 10 individual product categories) for each of the cyclic acetates assessed were used to calculate potential systemic exposures. Systemic exposures (i.e., the dose absorbed through the skin and available to the systemic circulation) were estimated based on dermal absorption rates. Where such data were lacking, as a conservative measure, dermal absorption was considered to be 100% (i.e., the maximum skin exposure value was considered as the estimate of systemic exposure). Systemic exposure estimates were compared to indices of systemic toxicity such as the no-observed-adverse-effect level (NOAEL) and the lowest-observed-adverse-effect level (LOAEL) values from subchronic, chronic, and reproductive toxicity studies.

Exposure data were provided by the fragrance industry. 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).

2. Metabolism

Esters are cleaved by carboxylesterases, members of the family of alpha/beta hydrolases, which show ubiquitous tissue expression with high levels in liver, small intestine and lung. The main two isoforms of carboxylesterases in humans are hCE-1 and hCE2. The substrate specificity differs for both enzymes: whereas hCE-1 cleaves substrates with a small alcohol moiety, hCE2 cleaves substrates with a small acyl group and a large alcohol group (Imai, 2006). Both enzymes also occur in skin (Zhu et al., 2007).

Metabolism studies of the substances under review are not available. However, some data for structurally similar monocyclic esters exist.

Hydrolysis of cyclohexyl esters has been observed with *cis*- and *trans*-1-methylene-4-isopropenylcyclohexan-2-yl acetate *in vitro* in the presence of rat liver homogenate. Incubation of the ester resulted in 92% hydrolysis after 15 min and 100% after 60 min. The

Table 4-1
Mutagenicity and genotoxicity: *in vitro* studies

Material	Test system		Concentrations	Results	References
4-*tert*-Butylcyclohexyl acetate	Ames assay with and without S9 activation	*Salmonella typhimurium* TA98, TA100, TA1535, TA1537, TA1538	~0.0024–9.4 μg/plate	Not mutagenic	RIFM (1980c)
Cyclohexyl acetate	Rec-assay	*Bacillus subtilis* strains H 17 (rec^+) and M 45 (rec^-)	19 μg/disc	Not mutagenic	Oda et al. (1978)
	Rec-assay (spore plate assay)	*Bacillus subtilis* strains H 17 (rec^+) and M 45 (rec^-)	20 μg/disc In DMSO	Not mutagenic	Yoo (1986)
4-Methyl-8-methylenetricyclo [3.3.1.1^{3,7}]decan-2-yl acetate	Ames assay with and without S9 activation (standard plate assay)	*Salmonella typhimurium* TA98, TA100, TA1535, TA1537, TA 1535, *E. coli* WP2 *uvrA*	5–500 μg/plate In DMSO (two independent tests)	Not mutagenic	RIFM (1989c)
	Ames assay with and without S9 activation (standard plate assay)	*Salmonella typhimurium* TA98, TA100, TA1535, TA1537, TA 1535, *E. coli* WP2 *uvrA*	Two independent tests: 0.5–150 or 1500 μg/plate In DMSO (cytotoxic)	Not mutagenic	RIFM (1991)
			1.5–150 or 500 μg/plate In DMSO (500 μg/plate tested in *E. coli* (±S9), in TA 1535, TA1537 (−S9) no cytotoxicity	Not mutagenic	
2-(1-Methylpropyl)-1-vinylcyclohexyl acetate	Ames assay with and without S9 activation (plate incorporation test plate and pre-incubation test)	*Salmonella typhimurium* TA98, TA100, TA102, TA1535, TA1537	33–5000 μg/plate	Not mutagenic	RIFM (2003a)
Myraldyl acetate	Ames assay with and without S9 activation (plate incorporation test plate and pre-incubation test)	*Salmonella typhimurium* TA98, TA100, TA102, TA1535, TA1537	Plate incorporation test:1.6–5000 μg/plate In DMSO (precipitation of test article at highest dose)	Not mutagenic	RIFM (2003b)
			Pre-incubation test: 8.2–5000 μg/plate (TA98(+S9), TA100, TA1535 (±S9): 3.3–2000 μg/plate (others)	Not mutagenic	
Tricyclodecenyl acetate	Ames assay with and without S9 activation	*Salmonella typhimurium* TA98, TA100, TA1535, TA1537, TA1538	~0.00001–0.01 μg/plate In DMSO (higher doses not tested due to cytotoxicity)	Not mutagenic	RIFM (1980d)

Table 4-2
Mutagenicity and genotoxicity: *in vivo* studies

Material	Test system	Species	Dose or concentration	Results	References
4-Methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate	Bone marrow micronucleus assay; sampling times: 24, 48 and 72 h after dosing	CD-1 mouse (15 per sex and group; positive control (mitomycin C) 5 per sex)	0, 1568 mg/kg bw In corn oil by single gavage	Not genotoxic; PCE/NCE not affected	RIFM (1989d)

Table 5
Reproductive and developmental toxicity studies

Material	Method	Concentration(s)/dose	Species	Results	References
4-*tert*-Butylcyclohexyl acetate	25 Pregnant rats were dosed via gavage on GD 7 through 20 and sacrificed on GD 21	0 (Vehicle), 40, 160 or 640 mg/kg bw/day in corn oil	Crl:CD® (SD) IGS BR VAF/PLUS® rats	*Maternal* 160 mg/kg bw/day: Excess salivation 640 mg/kg bw/day: 1 Rat sacrificed on GD 20 (adverse clinical observations: i.e. decreased motor activity, excess salivation, apparent dehydration; necropsy: distention of the stomach with gas and yellow fluid, litter of 17 dead fetuses). No test substance-related gross lesions in the remaining rats. Sparse hair coat on the limbs, red perioral substance; reduced body-weight gains during the entire treatment period; absolute and relative feed consumption values significantly reduced for the dosage period. *Offspring* Developmental observations at 640 mg/kg/day consisted of transient retardations in fetal development (significant reductions in fetal body weight and associated significant increases in moderate dilation of the renal pelvis, delayed ossification of the caudal vertebrae, fore- and hind-limb phalanges and hind-limb metatarsals). No other caesarean-sectioning or litter parameters were affected by 4-*t*BCHA as high as 640 mg/kg/day Both the maternal and developmental no-observable-adverse-effect-levels (NOAELs) for 4-*t*BCHA are 160 mg/kg	RIFM (2007)

GD: gestation day.

structurally related ethylene glycol and propylene glycol carbonate esters of (−)-2-isopropyl-5-methylcyclohexanol are completely hydrolyzed after incubation for 1 h with rat liver homogenate. Sterically hindered esters of cyclohexanol are also readily hydrolyzed in rat liver homogenate: 3,5,5-trimethylcyclohexanyl mandelate (cyclandelate) was completely hydrolyzed to 3,5,5-trimethylcyclohexanol and mandelic acid within 5 min of incubation with rat hepatic microsomal preparations (JECFA, 2007).

In rabbits, 39% of a dose of 350 mg/kg bw of cyclohex-1-en-1-yl acetate (an acetate which is not included in the group of acetates evaluated) was found to be hydrolyzed 18 h after oral administration (Elliott et al., 1959).

Data on hydrolysis of bi- or tri-cyclic esters are not available. As the rate of hydrolysis is determined by steric hindrance of the carboxylesterase substrate, it can be expected that acetates with bulky alcohol moieties are slowly hydrolyzed. However, esters with bulky alcohol moieties like cocaine and even heroin are nevertheless cleaved efficiently by human carboxylesterase hCE-2 (Imai, 2006). Similar to monocyclic esters, hydrolysis to the cyclic alcohol and the aliphatic carboxylic acid *in vivo* can be expected for the bi- or tri-cyclic esters.

After hydrolysis of the ester bond, acetic acid or – for one ester – cyclopent-2-eneacetic acid and primary, secondary or tertiary alcohols are formed. Acetic acid is an endogenous component of intermediary metabolism. Metabolism data for cyclopent-2-eneacetic acid are not available.

Primary alcohols (from abietyl acetate, octahydro-4,7-methano-1*H*-indenemethyl acetate, myraldyl acetate) can either be conjugated (see below) or oxidized via the aldehyde to the corresponding acid. These alicyclic acids are expected to be of a similar acidity as acetic acid; the pK_a value, e.g., for cyclohexanecarboxylic acid is 4.9 (acetic acid: 4.75) (SRC, 2007). Abietic acid, which is expected to be formed from abietyl acetate, was shown to be a weak acid (Söderberg et al., 1996). Further data are not available.

Metabolism data for secondary alicylic alcohols are available. For example, cyclohexanol is rapidly oxidized *in vivo* to the corresponding cyclohexanone derivative by alcohol dehydrogenase. Conversely, the cyclohexanone derivative may be reduced to cyclohexanol by cytosolic carbonyl reductases. Hence, the ketone and alcohol are interconvertible *in vivo*. Many metabolism studies are available on unsubstituted or alkyl-substituted cyclohexanols (cyclohexanol; 2-, 3-, or 4-*tert*-butylcyclohexanol; 2-isopropyl-5-methylcyclohexanol) and cyclohexanones (cyclohexanone; 2-, 3-, or 4-methylcyclohexanone; 2-isopropyl-5-methylcyclohexanone; 3,5,5-trimethyl-2-cyclohexen-1-one). Conjugation with glucuronic acid and excretion in bile and urine provide the predominant pathways for metabolic detoxification and elimination of cyclohexanol (see Fig. 1). Since cyclohexanone is readily converted to cyclohexanol and then to the glucuronic acid conjugate of cyclohexanol *in vivo*, data on cyclohexanone derivatives are directly relevant to the hazard assessment of cyclohexanol derivatives (FFHPVC, 2003; JECFA, 2007; see Fig. 1).

Oxidation of the alkyl ring substituents has been reported as a minor pathway in animals. The number of possible polyoxygenated metabolites increases with an increase in the types of alkyl ring substituents (e.g., methyl and isopropyl substituents) (Asakawa et al., 1986; Madyastha and Srivatsan, 1988; Nelson et al., 1992; Yamaguchi et al., 1994). Although it has been anticipated that lipophilic

Table 6-1
Skin irritation studies in humans

Material	Method	Concentration	Subjects	Results	References
Amylcyclohexyl acetate (mixed isomers)	Induction phase of HRIPT	2.5% In ethanol	42 Volunteers	No irritation	RIFM (1964a)
	48 h, Occlusive (pre-test for a maximization study)	12% In petrolatum	5 Healthy volunteers	No irritation	RIFM (1975b)
2-*tert*-Butylcyclohexyl acetate	48 h, Occlusive (pre-test for a maximization study)	4% In petrolatum	25 Healthy volunteers (females)	No irritation	RIFM (1976e)
4-*tert*-Butylcyclohexyl acetate	Induction phase of HRIPT (semi-occlusive)	6.25% In ethanol	42 Subjects	Scattered transient irritation was observed	RIFM (1964b)
	48 h, Occlusive (pre-test for a maximization study)	4% In petrolatum	31 Healthy volunteers	No irritation	RIFM (1976f)
	48 h, Occlusive (pre-test for a maximization study)	20% In petrolatum	33 Healthy volunteers	No irritation	RIFM (1976f)
d-Cyclocitronellene acetate	48 h, Occlusive (pre-test for a maximization study)	4% In petrolatum	28 Healthy volunteers	No irritation	RIFM (1982e)
Cyclohexyl acetate	Patch test (no further information)	100%	15–30 Subjects	Moderate reaction (classified as slight, moderate or severe, no further information)	Mallette and von Haam (1952)
	48 h, Occlusive (pre-test for a maximization study)	4% In petrolatum	10 Subjects	No irritation	RIFM (1974c)
	48 h, Occlusive (pre-test for a maximization study)	4% In petrolatum	31 Subjects	No irritation	RIFM (1977e)
Decahydro-beta-naphthyl acetate	48 h, Occlusive (pre-test for a maximization study)	12% In petrolatum	27 Volunteers	No irritation	RIFM (1976f)
(3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-Hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate	Induction phase of HRIPT	20% In petrolatum	50 Volunteers	No irritation	RIFM (1982c)
1-Ethynylcyclohexyl acetate	Induction phase of HRIPT	2% In 98% SDA (specially denatured alcohol) 40	39 Subjects	No irritation	RIFM (1971a)
4-(Isopropyl)-1-methylcyclohexyl acetate	Induction phase of HRIPT	6.25% In ethanol	42 Subjects	Scattered transient irritation was observed	RIFM (1964c)
Alpha-methylcyclohexylmethyl acetate	Induction phase of HRIPT	100%	52 Subjects	Mild fatiguing agent	RIFM (1977b)
4-Methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate	Induction phase of HRIPT	0.5% In alcohol SD 39C	48 Subjects	No irritation	RIFM (1989e)
	Induction phase of HRIPT	0.5% In alcohol SD 39C	48 Subjects	No irritation	RIFM (1989f)
	Induction phase of HRIPT	2% In alcohol SD 39C	48 Subjects	No irritation	RIFM (1989g)
	Induction phase of HRIPT	2% In alcohol SD 39C	48 Subjects	No irritation	RIFM (1989h)
	Induction phase of HRIPT	2% In alcohol SD 39C	48 Healthy volunteers	No irritation	RIFM (1989i)
	Induction phase of HRIPT	5% In alcohol SD 39C	49 Subjects	No irritation	RIFM (1996a)
	Induction phase of HRIPT	5% In alcohol SD 39C	49 Subjects	No irritation	RIFM (1996b)
	Induction phase of HRIPT	5% In alcohol SD 39C:DEP (75:25)	52 Subjects	No irritation	RIFM (1996c)
	Induction phase of HRIPT	5% In alcohol SD 39C:DEP (75:25)	52 Subjects	No irritation	RIFM (1996d)
Myraldyl acetate	Induction phase of HRIPT	10% In dimethyl phthalate	52 Healthy volunteers	No irritation	RIFM (1977f)
Octahydro-4,7-methano-1*H*-indenemethyl acetate	Induction phase of HRIPT	20% In petrolatum	50 Subjects	No irritation	RIFM (1976g)
Tricyclodecanyl acetate	Induction phase of HRIPT	6.25% In alcohol	41 Subjects	Irritation observed (1/41)	RIFM (1971b)
Tricyclodecenyl acetate	48 h, Occlusive (pre-test for a maximization study)	20% In petrolatum	35 Healthy volunteers	No irritation	RIFM (1977e)
	48 h, Occlusive (pre-test for a maximization study)	8% In petrolatum	26 Healthy volunteers	No irritation	RIFM (1974d) and Frosch et al. (1995)
	Induction phase of HRIPT	2% In dimethylphthalate	50 Subjects	No irritation	RIFM (1960)
1,7,7-Trimethylbicyclo[4.4.0]dec-3-yl acetate	Induction phase of HRIPT	20% In petrolatum (semi-occlusive)	50 Volunteers	No irritation	RIFM (1979e)
1,3,3-Trimethyl-2-norbornanyl acetate	48 h, Occlusive (pre-test for a maximization study)	5% In petrolatum	5 Healthy volunteers	No irritation	RIFM (1975b)

Table 6-2
Skin irritation studies in animals

Material	Method	Concentration (%)	Species	Results	References
Amylcyclohexyl acetate (mixed isomers)	Irritation evaluated as a part of LD_{50} study	100	Rabbit ($n = 10$)	Slight (4/10) to moderate (6/10) erythema, slight (3/10) to moderate (7/10) oedema	RIFM (1974a)
2-*tert*-Butylcyclohexyl acetate	Irritation evaluated as a part of LD_{50} study	100	Rabbit ($n = 10$)	Slight (6/10) to moderate redness (4/10), slight (3/10) to moderate edema (7/10)	RIFM (1976a)
	4 h, Semi-occlusive, 0.5 ml as a single application	100	Rabbit ($n = 10$)	Slightly to moderately irritant (slight to fairly distinct erythema and marginal to slight edema still present at 72 h, marginal to fairly distinct cracking, with marginal to fairly distinct scaling in some animals)	RIFM (1977g)
4-*tert*-Butylcyclohexyl acetate	4 h, Semi-occlusive, 0.5 ml as a single application	100	Rabbit ($n = 3$)	Irritation was observed (mean score for erythema 2.0 (all 3 animals) and for edema 1.3, 1.7 or 1.0)	RIFM (1984b)
	4 h, Semi-occlusive, 0.5 ml as a single application	100	Rabbit ($n = 4$)	No irritation	RIFM (1985)
	4 h, Semi-occlusive, 0.5 ml as a single application	100	Rabbit ($n = 8$)	Slight to moderate irritation	RIFM (1979f)
	Single open application on the clipped skin, readings at 4, 24 and 48 h (phototoxicity study)	1%, 3% and 10% in ethanol with 2% DMSO	Guinea pig ($n = 10$)	1%: No reactions 3 and 10%: Slight reaction (erythema and/or oedema) at 4 and 24 h (2/10 at 3%, 3/10 at 10%), no effects at 48 h	RIFM (1983)
	Irritation evaluated as a part of LD_{50} study	100	Rabbit ($n = 10$)	Moderate erythema and edema in all animals	RIFM (1976a)
	Irritation evaluated as a part of LD_{50} study	100	Rabbit ($n = 6$)	Scaliness and drying by d8, persisted through termination (14d)	RIFM (1979a)
	Single application to the back or ear (no further information) Back: 1, 5, 15 min and 20 h Ear: 20 h observation after 24 h and d8	100	Rabbits	Back: 1 and 5 min: Slight erythema 15 min: Severe erythema and slight edema 20 h: Severe erythema and edema At d8: slight redness, severe scaling Ear: 20 h: severe erythema and edema and blister At d8: severe necrosis	RIFM (1970a)
d-Cyclocitronellene acetate	Irritation evaluated as a part of LD_{50} study	100	Rabbit ($n = 10$)	Irritation observed	RIFM (1982a)
	Single application on intact and abraded skin, 24 h under occlusive dressing, readings at patch removal and 72 h (pre-test)	10% and 20% In propylene glycol	Rabbit ($n = 1$ per dose)	20%: Very slight erythema at both skin sides at 24 and 72 h, very slight edema at both sides at 24 h, only intact side at 72 h 10%: Very slight erythema and edema at intact side at 24 h, no effects at 72 h	RIFM (1981c)
	Single application on intact and abraded skin, 24 h under occlusive dressing, readings at patch removal and 72 h (pre-test)	10% In propylene glycol	Rabbit ($n = 6$)	Primary Irritation Index (PII) 0.8 (very slight to well-defined erythema in three rabbits at both the intact and abraded sites, very slight edema in two rabbits (both on the intact side) at 24 h, very slight erythema in two rabbits (both sides) and at abraded side of one rabbit, very slight edema still present at 72 h in one animal)	RIFM (1981c)
Cyclohexyl acetate	Irritation evaluated as a part of LD_{50} study	100	Rabbit ($n = 10$)	Irritation observed	RIFM (1977a)
	Irritation evaluated as a part of LD_{50} study	100	Rabbit ($n = 4$)	No irritation observed	RIFM (1974b)
	24 h, Unoccluded, 0.01 ml as a single application on clipped belly, observations within 24 h of the application	100	Rabbit ($n = 5$)	Irritation was observed	Carpenter et al. (1974)
	Irritation test (protocol details not available)	100	Rabbit ($n = 2$–4)	No irritation	Mallette and von Haam (1952)
Decahydro-beta-naphthyl acetate	Irritation evaluated as a part of LD_{50} study	100	Rabbit ($n = 8$)	Irritation observed	RIFM (1976a)
1-Ethynylcyclo-hexyl acetate	Single application on intact and abraded skin, 0.5 ml, 24 h under occlusive dressing, readings at patch removal and at 72 h	2% In SDA 40	Rabbit ($n = 3$)	No irritation (PII 0.0)	RIFM (1970b)

(*continued on next page*)

Table 6-2 (*continued*)

Material	Method	Concentration (%)	Species	Results	References
(3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-Hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate	Irritation evaluated as a part of LD_{50} study	100	Rabbit (n = 6)	Slight erythema and edema at d1 in all animals, absent by d7	RIFM (1982b)
	Single application on intact and abraded skin, 0.5 ml, 24 h under occlusive dressing, readings at patch removal and at 72 h	100	Rabbit (n = 6)	No irritation (PII 1.25)	RIFM (1982f)
Alpha-methylcyclo-hexylmethyl acetate	Primary irritation test (24 h)	100	Rabbit (n = 6)	Mildly irritating: slight erythema and edema and desquamation in all animals (irritation score for erythema and edema (24 and 72 h): 2.0, max. 8.0), reversible at 168 h	RIFM (1977b)
4-Methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate	Irritation evaluated as a part of LD_{50} study	100	Rat (n = 10, 5/sex)	Slight erythema in all five males and well-defined erythema in all five females by d3, cleared by d4 in males and by d6 in all females	RIFM (1989a)
	Single application on the clipped skin, 6 h under occlusive dressing reading 24 h after application (pre-test for Buehler test)	1%, 10%, 25% and 50% In ethanol (80%)	Guinea pig (n = 4, 2/sex)	1%: Only slightly patchy erythema in 1/4 animals, concentration chosen for Buehler test 10%: Slight to severe erythema with/without edema 25% and 50%: Moderate to severe erythema with/without edema	RIFM (1989j), RIFM (1989k)
	4 h, Semi-occlusive, 0.5 ml as a single application	100	Rabbit (n = 3)	Well-defined erythema with slight oedema in all three animals by d2, reactions ameliorated by d7 and resolved by d8, d11 or d13	RIFM (1989m)
2-(1-Methyl-propyl)-1-vinylcyclohexyl acetate	Single application on clipped skin, 0.1 ml, 24 h uncovered (OET pre-test)	100%, 30%, 3% and 10% In ethanol	Guinea pig (n=6)	30%: Lowest irritant concentration (causing mild erythema in at least 25% of an animal group) 10%: Highest non irritant concentration (does not cause macroscopic reactions in any of the animals of the group)	RIFM (1977h)
	0.1 ml, 24 h Uncovered, 21 consecutive days (OET test)	100%, 30%, 10% and 3% In ethanol	Guinea pig (n = 6)	100%: Moderate skin irritation after 7 days (because of the skin reaction the application site was changed every other week) 30%: Slight skin irritation after 7 days (see above) 10% and 3%: No effects	RIFM (1977h)
	Induction phase of Buehler test. Closed patch for 6 h once a week for three weeks, readings d7, d14, d21	10% In DEP	Guinea pig (20 in test group, 10 in control group)	No irritation observed	RIFM (1976h)
	Single application, 48 h (part of phototoxicity test)	10% In rectified alcohol	Guinea pig (n = 4)	No irritation observed	RIFM (1979g)
	24-h irritation test	100%, 30%, 3% and 10% in diethyl phthalate	Rabbit	3%: No irritation Irritation observed at 100, 30 and 10%.	RIFM (1973)
Myraldyl acetate	Single application, open application	1% and 3% In acetone	Guinea pig (no further information)	3%: Irritant reactions 1%: No irritant reactions	RIFM (1972c)
	24 h Uncovered, 21 consecutive days (OET test)	3%, 10%, 30% and 100% In acetone	Guinea pig (no further information)	30% and 100%: Slight (d7), moderate (d14), severe (d14) redness 10%: Very slight (d7), slight (d14), moderate (d21) redness 3%: Very slight redness (d7–d21)	RIFM (1972c)
Octahydro-4,7-methano-1*H*-indenemethyl acetate	Irritation evaluated as a part of LD_{50} study	100	Rabbit (n = 4)	Mild irritation pale red erythema and mild edema after patch removal; mild to moderate desquamation at d7 and d14)	RIFM (1976c)

Tricyclodecanyl acetate	Single application on intact and abraded skin, 0.5 ml, 24 h under occlusive dressing, readings at patch removal and at 72 h	6.25% In alcohol SDA 39C (mixture of 10 different chemicals)	Rabbit (n = 3)	No irritation observed (Primary irritation index: 0)	RIFM (1971c)
Tricyclodecenyl acetate	Irritation evaluated as a part of LD_{50} study	100	Rabbit (n = 10)	Irritation observed	RIFM (1977a)
	Irritation evaluated as a part of LD_{50} study	100	Rabbit (n = 10)	Irritation observed	RIFM (1974a)
	4 h, Semi-occlusive, 0.5 ml as a single application, observations after patch removal, 24, 48 and 72 h	100	Rabbit (n = 7)	Slight irritation (slight erythema and edema, still present at 72 h, very slight cracking and scaling at that timepoint)	RIFM (1979h)
	4 h, Semi-occlusive, 0.5 ml as a single application, observations after patch removal, 24, 48 and 72 h	100	Rabbit (n = 10)	Slight to moderate irritation (marginal to slight erythema and edema with variable cracking and scaling ranging from slight to distinct)	RIFM (1976i)
1,7,7-Trimethylbicyclo[4.4.0]dec-3-yl acetate	Single application on intact and abraded skin, 0.5 g, 24 h under occlusive dressing, readings at patch removal and at 72 h	100	Rabbit (n = 6)	No irritation observed (PII = 0.12)	RIFM (1979i)
1,3,3-Trimethyl-2-norbornanyl acetate	Irritation evaluated as a part of LD_{50} study	100	Rabbit (n = 10)	Irritation observed	RIFM (1975a)

alcohols or ketones with sterically hindered functional groups undergo more extensive oxidation of alkyl ring substituents (Nelson et al., 1992), studies with 2-, 3-, or 4-methylcyclohexanol, 2-isopropyl-5-methylcyclohexanol, 3,5,5-trimethylcyclohexanol, and even 2-, 3-, or 4-*tert*-butyl-substituted cyclohexanol or cyclohexanones reveal that conjugation of the cyclohexanol moiety by glucuronic acid is the predominant excretion pathway regardless of the size or position of the ring substituent. In general, the metabolic fate of alkyl-substituted cyclohexanol and cyclohexanone derivatives are similar to that of the non-substituted homologues (JECFA, 2007; Lington and Bevan, 1994; Topping et al., 1994).

Tertiary alcohols cannot be further oxidized; as for secondary alcohols, conjugation of the alcohol group with glucuronic acid and subsequent elimination is to be expected (Bernauer et al., 1998).

Of the cyclic acetates evaluated, 17 are hydrolyzed to a secondary alcohol (2-*tert*-butylcyclohexyl acetate, 4-*tert*-butylcyclohexyl acetate, amylcyclohexyl acetate, *d*-cyclocitronellene acetate, alpha-methylcyclohexylmethyl acetate, *cis*-2-*tert*-butylcyclohexyl acetate, 3-*tert*-butylcyclohexyl acetate, cyclohexyl cyclopent-2-ene-1-acetate, cyclohexyl acetate, 1,3,3-trimethyl-2-norbornanyl acetate, decahydro-beta-naphthyl acetate, 1,7,7-trimethylbicyclo[4.4.0]dec-3-yl acetate, tricyclodecenyl acetate, tricyclo[5.2.1.$0^{2,6}$]dec-4-en-8-yl acetate, tricyclodecanyl acetate, (3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate; 4-methyl-8-methylenetricyclo[3.3.1.$1^{3,7}$]decan-2-yl acetate), three to a primary alcohol (abietyl acetate; octahydro-4,7-methano-1*H*-indenemethyl acetate, myraldyl acetate) and five to a tertiary alcohol (1-ethynylcyclohexyl acetate; 4-(isopropyl)-1-methylcyclohexyl acetate, 1-methyl-2-(1-methylpropyl)cyclohexyl acetate, 1-methyl-4-(1-methylvinyl)cyclohexyl acetate, 2-(1-methylpropyl)-1-vinylcyclohexyl acetate).

3. Pharmacokinetics

Studies with the esters under review are not available. Available data on the hydrolysis products of the esters with monocyclic alcohols (cyclohexanol and cyclohexanone and alkyl-substituted derivatives) have been summarized by the flavor and fragrance high production volume consortia (FFHPVC, 2003). Data on alicyclic ketones, secondary alcohols and related esters including cyclohexyl acetate, a member of this group summary, which are used as food additives, have been summarized by the Joint FAO/WHO Expert Committee on Food Additives (JECFA, 2007).

3.1. Dermal route of exposure

No percutaneous absorption studies in humans or experimental animals are available for the cyclic acetates. For the purpose of exposure and safety assessment 100% dermal absorption is assumed (see Section 1.3).

3.2. Oral route

No studies in humans or experimental animals via the oral route are available for the cyclic acetates.

Alkyl-substituted cyclohexanols are rapidly absorbed, conjugated with glucuronic acid, and excreted mainly in the urine. Alkyl-substituted cyclohexanones are also rapidly absorbed, reduced to the corresponding cyclohexanol derivatives, conjugated with glucuronic acid and then excreted in the urine (FFHPVC, 2003). The size, position, number, or stereochemistry of alkyl substituents on the cyclohexyl ring exerts no significant effect on the rate of absorption and excretion of alkyl-substituted cyclohexanol or cyclohexanone derivatives (FFHPVC, 2003).

Table 7
Mucous membrane irritation studies in rabbits

Material	Method	Results	References
Amylcyclohexyl acetate (mixed isomers)	0.1 ml, 100%, Three animals	Slight to moderate conjunctival irritation with chemosis and discharge in all 3 rabbits, all effects cleared by d7 (mean score for redness: 1.7, mean score for chemosis: 1.4)	RIFM (1964d)
2-*tert*-Butylcyclohexyl acetate	0.1 ml, 50% In Tween 80, observations at 24 h thereafter daily, three animals	Persistent corneal lesions (moderate corneal lesions affecting 25% of corneal surface in all animals, slight to severe conjunctivitis, slight to moderate discharge (2/3), iritis (2/3), one animal was killed on day 8 (lesions increased in severity), the remaining two animals had persistent corneal lesions at test end (21 d)	RIFM (1976j)
	0.1 ml, 100%, Observations at 24 h, four animals	No irritation; one animal showed signs of discomfort when treated	RIFM (1977i)
4-*tert*-Butylcyclohexyl acetate	50 mm^3 (0.05 ml), 100%, (No further information)	Slight redness by 1 and 24 h; cleared by d8	RIFM (1970a)
	0.1 ml, 0.625% (Vehicle not mentioned), observations at d1, d2, d3, d4 and d7, three animals	Slight conjunctival irritation with chemosis and discharge in all three rabbits, all effects cleared by d4 (mean score for redness 1.9 and for chemosis: 1)	RIFM (1964e)
d-Cyclocitronellene acetate	0.1 ml, 0.1%, 0.5%, 1% and 2% In propylene glycol, observations 24, 48 and 72 h and at d4 and d7, one animal/dose	0.1%: No irritation 0.5% and 2%: Mild inflammation of the conjunctivae, cleared by d2 or d4, respectively	RIFM (1981d)
	0.1 ml, 0.25% and 0.5% In propylene glycol, observations 24, 48 and 72 h and at d4 and d7, three animals/dose	1%: Some corneal opacity, which was cleared by d2, diffuse Crimson-red coloration of the conjunctivae with a slight swelling of the eyelid, cleared by d4	
	0.1 ml, 0.1% In propylene glycol, observations 24, 48 and 72 h and at d4 and d7, three animals	0.5%: All three animals were "positive": corneal opacity in two animals, cleared by d2, temporary iritis in one of these animals, mild inflammation or diffuse, Crimson-red coloration of the conjunctivae with or without slight chemosis in all three animals, normal at d3, d7 or d14 0.25%: two animals "positive": diffuse, Crimson-red coloration of the conjunctivae with slight chemosis, normal at d3 or d7 0.1%: Not "positive", mild inflammation of the conjunctivae in one animal, cleared by d3	
Cyclohexyl acetate	100% In propylene glycol, water or Deobase, five animals (no further information.)	Corneal injuries were observed	Carpenter et al. (1974)
1-Ethynylcyclohexyl acetate	0.1 ml, 2% In SDA 40, observations at 1, 2, 3, 4 and 7 days; three animals	Mild transient conjunctival irritation (mild (1/3) to moderate redness (2/3) with mild (2/3) to moderate chemosis (1/3) and mild discharge (3/3) by d1, all effects cleared by d7	RIFM (1970d)
(3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-Hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate	0.1 ml 100%, Observations at 1, 2 and 3 days; six animals	No irritation	RIFM (1982g)
4-(Isopropyl)-1-methylcyclohexyl acetate	0.1 ml, 6.25% (Vehicle not specified), observations at d1, d2, d3, d4 and d7, 3 animals	Mild vessel injection of the palpebral conjunctivae, reversible at day 3 (mean score for redness: 0.7, edema: 0.0)	RIFM (1963)
Alpha-methylcyclohexylmethyl acetate	0.1 ml, 100%, Observations immediately and at 1, 24, 48, 72, 120 and 168 h, six animals	Slightly irritating: slight to moderate erythema and copious discharge in all animals, all effects cleared by 120 h (Irritation score (24, 48 and 78 h): 5.9, max. score = 110)	RIFM (1977b)
4-Methyl-8-methylenetricyclo[3.3.1.$1^{3,7}$]decan-2-yl acetate	0.1 ml, 100%, Observations at 1 h and d1, d2, d3, d4 and d7, three animals	Slight corneal opacity in 2/3 animals, slight (2/3) to moderate redness (1/3), very slight (1/3) to obvious swelling in 2/3; all effects cleared by d3	RIFM (1989l)
2-(1-Methylpropyl)-1-vinylcyclohexyl acetate	0.1 ml, 100% and 3%, 10% and 30% (Vehicle not mentioned), observations immediately and at 1, 24, 48, 72 h and after 7 and 14 d; Three animals/dose	100%: Slight conjunctival erythema in all rabbits, cleared by d7 30%: Very slight conjunctival erythema in all rabbits, cleared by d7 10%: Very slight conjunctival erythema in all rabbits after 1 h, cleared by d1 (2/3 animals), all eyes had normalized by 48 h 3%: very slight conjunctival erythema in 1/3 rabbits after 1 h, all eyes had normalized by 24 h	RIFM (1977j)
Octahydro-4,7-methano-1*H*-indenemethyl acetate	0.1 ml, 100%, Unwashed, six animals	No irritation (Draize scores of 0 at d1, d2, d3, d4, d7)	RIFM (1979j)
1,7,7-Trimethylbicyclo[4.4.0]dec-3-yl acetate	0.1 ml, 100%, Observations on d1, d2, d3, d5 and d7, six animals	No irritation (Draize scores of 0 at d1, d2, d3, d5 and d7)	RIFM (1979k)
Tricyclodecanyl acetate	0.1 ml 6.25% In alcohol SDA 39C, three animals (mixture of 10 different chemicals)	Moderate irritation (only conjunctivae involved, effects cleared by d3)	RIFM (1970c)
Tricyclodecenyl acetate	0.1 ml 100%, Three animals	Non-persistent corneal lesions (50% of corneal surface in 2/3 animals) associated with moderate corneal swelling, slight conjunctivitis and discharge (1/2), healed within 3 d, one animal unaffected	RIFM (1977k)
	0.1 ml "Crude", 50% in Tween 80, three animals	Persistent corneal lesions (moderate to severe corneal swelling and slight conjunctivitis, iritis in 2/3, pannus in 1/3, moderate corneal opacity affecting 75% of the cornea in all three animals, lesions healed within 10 d (2/3), in the remaining animal still present at test termination (d21)	RIFM (1977l)

Table 8-1A
Skin sensitization studies in humans

Material	Method	Concentration(s)	Subjects	Results	References
Amylcyclohexyl acetate (mixed isomers)	HRIPT	2.5% In ethanol	42 Volunteers	No sensitization reactions	RIFM (1964b)
	Maximization test	12% In petrolatum	25 Healthy volunteers	No sensitization reactions	RIFM, 1975b
2-*tert*-Butylcyclohexyl acetate	Maximization test	4% In petrolatum	25 Healthy volunteers (females)	No sensitization reactions	RIFM (1976e)
4-*tert*-Butylcyclohexyl acetate	HRIPT (semi-occlusive)	6.25% In ethanol	42 Volunteers	No sensitization reactions	RIFM (1964b)
	Maximization test	4% In petrolatum	31 Healthy volunteers	No sensitization reactions	RIFM (1976f)
	Maximization test	20% In petrolatum	33 Healthy volunteers	No sensitization reactions	RIFM (1976f)
d-Cyclocitronellene acetate	Maximization test	4% In petrolatum	28 Healthy volunteers	No sensitization reactions	RIFM (1982e)
Cyclohexyl acetate	Maximization test	4% In petrolatum	31 healthy volunteers	No sensitization reactions	RIFM (1977e)
	Maximization test	4% In petrolatum	25 Healthy volunteers	No sensitization reactions	RIFM (1974c)
Decahydro-beta-naphthyl acetate	Maximization test	12% In petrolatum	27 Healthy volunteers	No sensitization reactions	RIFM (1976f)
1-Ethynylcyclohexyl acetate	HRIPT	2% In alcohol SD 40	39 Volunteers	1 Volunteer exhibited erythematous and papular response at challenge on the original test site, but not on a fresh test site. No rechallenge was made	RIFM (1971a)
(3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-Hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate	HRIPT (semi-occlusive)	20% In petrolatum	50 Volunteers	No sensitization reactions	RIFM (1982c)
4-(Isopropyl)-1-methylcyclohexyl acetate	HRIPT	6.25% In ethanol	42 Volunteers	No sensitization reactions	RIFM (1964c)
Alpha-methylcyclohexylmethyl acetate	HRIPT	100%	52 subjects	No sensitization reactions	RIFM (1977b)
4-Methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate	HRIPT	0.5% in alcohol SD 39C	48 Healthy Volunteers	No sensitization reactions	RIFM (1989e)
	HRIPT	0.5% In alcohol SD 39C	48 Healthy volunteers	No sensitization reactions	RIFM (1989f)
	HRIPT	2% In alcohol SD 39C	48 Healthy volunteers	No sensitization reactions	RIFM (1989g)
	HRIPT	2% In alcohol SD 39C	48 Healthy Volunteers	No sensitization reactions	RIFM (1989i)
	HRIPT	2% In alcohol SD 39C	48 Volunteers	No sensitization reactions	RIFM (1989h)
	HRIPT	5% In alcohol SD 39C	49 Volunteers	No sensitization reactions	RIFM (1996b)
	HRIPT	5% In alcohol SD 39C	49 Volunteers	No sensitization reactions	RIFM (1996a)
	HRIPT	5% In alcohol SD 39C:DEP (75:25)	52 Volunteers	No sensitization reactions	RIFM (1996c)
	HRIPT	5% In alcohol SD 39C:DEP (75:25)	52 Volunteers	No sensitization reactions	RIFM (1996d)
Myraldyl acetate	HRIPT	10% In dimethyl phthalate	52 Healthy volunteers	No sensitization reactions	RIFM (1977f)
Octahydro-4,7-methano-1*H*-indenemethyl acetate	HRIPT	20% In petrolatum	50 Volunteers	No sensitization reactions	RIFM (1976g)
Tricyclodecanyl acetate	HRIPT	6.25% In alcohol	41 Volunteers	No sensitization reactions	RIFM (1971b)
Tricyclodecenyl acetate	Maximization test	20% In petrolatum	26 Healthy volunteers	No sensitization reactions	RIFM (1977e)
	Maximization test	8% In petrolatum	21 Healthy volunteers	No sensitization reactions	RIFM (1974d)
	HRIPT	2% In dimethyl phthalate	50 Volunteers	No sensitization reactions	RIFM (1960)
1,7,7-Trimethylbicyclo[4.4.0]dec-3-yl acetate	HRIPT	20% In petrolatum	50 Volunteers	No sensitization reactions	RIFM (1979e)
1,3,3-Trimethyl-2-norbornanyl acetate	Maximization test	5% In petrolatum	25 Healthy volunteers	No sensitization reactions	RIFM (1975b)

3.3. Respiratory route of exposure

No studies in humans or experimental animals are available for the cyclic acetates.

After exposure of volunteers to concentrations of 101, 207, or 406 mg/m^3 of cyclohexanone for 8 h approximately 60% of the cyclohexanone dose was excreted in the urine within a 72-h period (Mraz et al., 1994).

3.4. Parenteral route of exposure

No studies in humans or experimental animals are available for the cyclic acetates.

Male beagle dogs were given 284 mg/kg bw of cyclohexanone by intravenous injection daily. Cyclohexanol was detected in the plasma within 30 min after injection. The mean distribution and elimination half-lives of cyclohexanone and cyclohexanol are 6.6

Table 8-1B
Diagnostic patch test studies in humans

Material	Method	Concentration(s)	Subjects	Results	References
2-*tert*-Butylcyclohexyl acetate	Patch test	1% and 5% In petrolatum	313 Dermatitis patients	No sensitization	Frosch et al. (1995)
4-*tert*-Butylcyclohexyl acetate	Patch test	1% and 5% In petrolatum	107 Patients	No sensitization reactions	Frosch et al. (1995)
	Closed patch test on the back or forearm or upper arm for 24 to 48 h, results read 30 min after removal	0.05–0.5% (In a base cream or in 99% ethanol)	148 Subjects	Nine reactions were observed Five questionable reactions were also observed	Takenaka et al. (1986)
Cyclohexyl acetate	Patch test	5% In petrolatum	218 Fragrance sensitive patients	One positive reaction (0.5%)	Larsen et al. (2002)

Table 8-2
Skin sensitization studies in animals

Material	Method	Concentration(s)	Species	Results	References
d-Cyclo-citronellene acetate	Maximization test	Induction: 10% in liquid paraffin (intradermal) and 100% (percutaneous); challenge: 10% and 20% in liquid paraffin	Guinea pig	No sensitization reactions (0/14)	RIFM (1981e)
4-Methyl-8-methylene-tricyclo[3.3.1.1^{3,7}]decan-2-yl acetate	Buehler test	Induction: closed patch (dermal dam) topical application of 1% in ethanol for 6 h once a week for three weeks; Challenge: 1% in ethanol; Re-challenge: 0.2% and 1% in ethanol (7 d after primary challenge)	Guinea pig (20 in test group, 10 in control group)	Challenge: five positive reactions (5/20) at 1%, Re-challenge: positive reactions (3/20 at 0.2% and 7/20 at 1% 48 h after re-challenge)	RIFM (1989j, 1989k)
	Local lymph node assay (LLNA)	7.5%, 15%, and 30% (w/v) In EtOH:DEP 1:3	Mouse (five females/group)	No sensitization (stimulation index: 1.19, 1.40 and 1.75, respectively)	RIFM (2004)
2-(1-Methyl-propyl)-1-vinylcyclohexyl acetate	Buehler test	Induction: closed patch for 6 h once a week for three weeks; Challenge: 10% in DEP	Guinea pig (20 in test group, 10 in control group)	No sensitization reactions	RIFM (1976h)
	Freund's complete adjuvant test	Induction: 5% emulsion in FCA; challenge: 3% and 10% (vehicle not mentioned)	Guinea pig (eight in test and control group)	No sensitization reactions	RIFM (1977m)
	Open epicutaneous test (OET)	Induction:100%, 30%, 10% and 3% in ethanol;Challenge: 10% in ethanol	Guinea pig (6/group)	No sensitization reactions	RIFM (1977h)
Myraldyl acetate	Open epicutaneous test (OET)	Induction: 100%, 30%, 10% and 3% in acetone;Challenge: 1% in acetone	Guinea pig	No sensitization reactions	RIFM (1972c)

and 81 min, respectively. The mean steady state volume of distribution for cyclohexanone is 2.6 l/kg, the mean total body clearance for cyclohexanone is 27.4 ml/kg/min (Martis et al., 1980; Koeferl et al., 1981). When 328 mg/kg bw cyclohexanol was administered by intravenous injection, the plasma half-life was 99 min, the apparent distribution volume 1.2 l/kg and the total body clearance 8.8 ml/kg/min. Based on these data cyclohexanone and cyclohexanol are rapidly cleared from the body (Martis et al., 1980). Approximately 60% of cyclohexanone administered was recovered in the urine as glucuronide conjugate of cyclohexanol after 24 h. The direct renal clearance of unmodified cyclohexanone and cyclohexanol is a minor route of elimination accounting for less than 1% of the administered dose. It is proposed that 74–100% of cyclohexanone is converted to cyclohexanol and further metabolized before elimination. The authors assume that some of the cyclohexanone may be exhaled (Martis et al., 1980; Koeferl et al., 1981).

4. Toxicological studies

4.1. Acute toxicity

The acute dermal toxicity has been reported for 13 of the 25 evaluated cyclic acetates. The LD_{50} values are greater than 2000 mg/kg bw or even greater than 5000 mg/kg bw, indicating that these materials are of low toxicity or are practically non toxic via the dermal route (Table 2-1).

Acute oral toxicity has been reported for 16 of the 25 evaluated cyclic acetates. Acute oral toxicity of the cyclic acetates tested is very low with LD_{50} values for most materials in rats and mice reported to be between 2000 and 5000 mg/kg bw or greater. For 4-*tert*-butylcyclohexyl acetate the rat oral LD_{50} value is greater than 500 and below 5000 mg/kg bw; for 2-*tert*-butylcyclohexyl acetate the mouse oral LD_{50} is 310–500 mg/kg bw (Table 2-2).

Reported clinical signs were lethargy and piloerection after dermal or oral administration. Signs of irritation were noted at necropsy in acute oral toxicity tests, for example irritation of different parts of the gastrointestinal tract, chromorrhinorrhea and/or salivation.

Acute toxicity data obtained from studies performed with other than the oral and dermal routes of exposure are summarized in Table 2-3.

4.2. Repeated dose toxicity

Information on toxicity after repeated application is available for only one of the cyclic acetates. The results of this study with the tricyclic 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate are summarized in Table 3 and described below. Furthermore, a developmental toxicity study was conducted with 4-*tert*-butylcyclohexyl acetate, in which pregnant rats were dosed for 14 days (see 4.5 and Table 5). As the unsubstituted monocyclic esters (e.g. cyclohexyl acetate) are rapidly hydrolyzed to the cyclic

Table 9
Phototoxicity and photoallergenicity

Material	Method	Concentration	Species	Results	References
4-*tert*-Butylcyclohexyl acetate	Single open application of 0.025 ml, 30 min after application UV irradiation at 320–400 nm (duration not specified), 10 animals	1%, 3% and 10% In ethanol with 2% DMSO	Guinea pig	No phototoxicity was observed	RIFM (1983)
(3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-Hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate	Photoallergy study as part of HRIPT test with nine applications, irradiation for 15 min (365 nm), irradiation at application 1, 4, 7, 9 and challenge	20% In petrolatum	20 Healthy volunteers	No phototoxicity and no photoallergy were observed	RIFM (1982c)
2-(1-Methylpropyl)-1-vinylcyclohexyl acetate	0.1 ml Every alternate day plus UV B for 15 min and thereafter UV A for 4 h (total nine times in 18 d) on the upper dorsal area; after resting time of 10 d 0.025 ml applied on both flanks, left flank irradiated as for induction; skin reading 24 and 48 h after challenge	10% In rectified alcohol	Guinea pig	No phototoxicity was observed	RIFM (1980e)
	Eight animals, six animals control group (tested and read as exp. group during challenge phase)	3% In rectified alcohol		No photoallergy was observed	
	Four animals per group; patch applied 48 h; 4 h after removal of patches left flank of two groups of experimental animals was radiated: Group A: with UVA (320–400 nm), 30 min Group B: with UVB (280–370 nm), 15 min Group C (control): no radiation Group D (control): not pretreated, radiated from both light sources Observations 4, 16 and 24 h after radiation	10% In rectified alcohol	Guinea pig	No phototoxicity was observed	RIFM (1979g)
1,7,7-Trimethylbicyclo[4.4.0]dec-3-yl acetate	Photoallergy study as part of HRIPT test with nine applications, irradiation for 15 min (365 nm), irradiation at application 1, 4, 7, 9 and challenge	20% In petrolatum	20 Healthy volunteers	No phototoxicity was observed No photoallergy was observed	RIFM (1979e)

Table 10
Summary of UV spectra data on cyclic acetates

Material	UV spectra range of absorption
Amylcyclohexyl acetate	Peaked at 200 nm range
2-*t*-Butylcyclohexanol acetate	No absorption
4-*t*-Butylcyclohexanol acetate	No absorption
Cyclohexyl acetate	200–230 nm range
Decahydro-β-naphthyl acetate	200–230 nm range
(3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-Hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate	200–260 nm range
Octahydro-4,7-1*H*-indenemethyl acetate	200–220 nm range
Tricyclo[5.2.1.02,6]dec-4-en-8-yl acetate	200–210 nm range
1,7,7-Trimethylbicyclo[4.4.0]dec-3-yl acetate	200–270 nm range
1,3,3-Trimethyl-2-norbornanyl acetate	Peaked at 200–210 nm returning to baseline at 300–320 nm

alcohols which (in the case of secondary alcohols) are rapidly further oxidized to the corresponding cyclic ketone, and since the ketone and alcohol are interconvertible, studies with cyclic alcohol (e.g. cyclohexanols) and cyclic ketone derivatives of unsubstituted cyclic esters are also reviewed and evaluated.

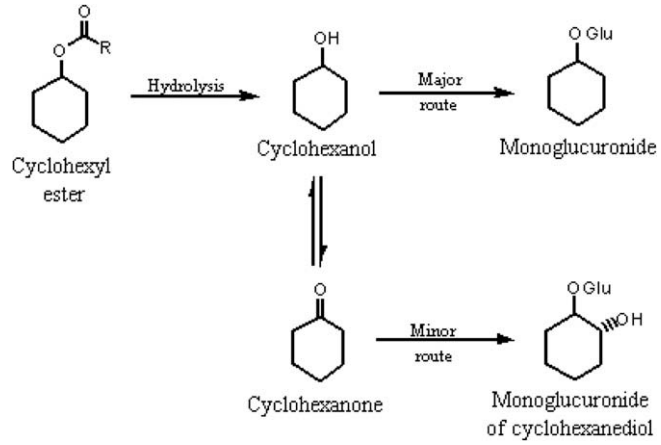


Fig. 1. Metabolic fate of cyclohexyl esters in humans (JECFA, 2007).

4.2.1. Dermal studies

No repeated dose dermal toxicity studies are available for the acetates.

4.2.2. Oral studies

4-Methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate was tested in a 28-day toxicity study in rats. The test substance was administered by gavage to male and female rats at dose levels of 0, 5, 55, or 1000 mg/kg bw/day in corn oil, with five animals per sex and group. Observations included mortality, behavior, body weight, food and water consumption, hematology, clinical chemistry and macroscopic examinations. Microscopic examinations were conducted on adrenals, heart, kidneys, liver, spleen and any other macroscopically abnormal tissue in the control and high-dose group and on kidneys and liver in the low- and mid-dose group. Abnormal clinical signs and higher neutrophil counts (males) and blood urea nitrogen (BUN) were noted in the high-dose groups, as were lower body-weight gains during weeks 2–4 (females), statistically significantly higher serum total protein and globulin and reduced phosphorus. In females given 55 mg/kg bw BUN was elevated. Increased relative kidney weights and moderate degrees of eosinophilic inclusions in the cytoplasm of the cortical epithelium of the kidneys were observed in high-dose group males. Increased relative liver weights were noted in high-dose groups. The authors

concluded a NOAEL of 55 mg/kg bw/day because the changes at this dose (salivation, raised blood urea nitrogen levels in females) were considered to be of minor toxicological importance (RIFM, 1990). Taking into account these slight alterations at that dosage, the NOEL is 5 mg/kg bw/day (see Table 3).

A 28-day toxicity study was conducted with 4-*tert*-butylcyclohexanol, the corresponding alcohol of 4-*tert*-butylcyclohexyl acetate. Groups of rats received 0, 50, 150, or 300 mg/kg bw/day of 4-*tert*-butylcyclohexanol by gavage for 28 days with a 14-day recovery period (HPVIS, 2008). Clinical signs immediately after dosing at the two highest doses included convulsions, squatting position, straub tail and vocalization, all of which disappeared within a few hours. No clinical abnormalities were observed in the controls. After 2, 3 and 4 weeks of treatment, observations in individual rats (predominantly from the high-dose group) included ataxia, fasciculations, padding movements, a guard against touching, aggressiveness, hunchback/squatting position, reduced respiration, hyperactivity, straub tail, and minimal convulsions. In the recovery period, no significant treatment-related clinical signs were observed in any treatment group. Treatment-related histopathological findings were restricted to eosinophilic hyaline droplets in the renal epithelial cell cytoplasm of the proximal tubules of high-dose males (5/10 treated compared to 1/10 control). The authors proposed that the effect may be indicative of the alpha-2 micro-globulin nephropathy syndrome that is a male rat-specific effect. The authors concluded, based on clinical signs, that the NOAEL was 50 mg/kg bw/day and the LOAEL was 150 mg/kg bw/day.

A 13-week toxicity study was performed in which groups of 10 $B6C3F_1$ mice of each sex received drinking-water containing cyclohexanone, a corresponding ketone of cyclohexyl acetate, at a concentration of 0, 400, 2300, 6500, 13,000, 25,000, 34,000 or 47,000 ppm, corresponding to 0, 100, 580, 1600, 3200, 6200, 8500, or 12,000 mg/kg bw/day, respectively (Lijinsky and Kovatch, 1986). No effects were reported at 100, 580 or 1600 mg/kg bw/day. Male mice given 6200 mg/kg bw/day dose showed a 19% decrease in body-weight gain; among animals at 8500 mg/kg bw/day, one male died before 13 weeks, and weight gain was depressed by 15% in females and 24% in males. At the highest dose, 6/10 males and 3/10 females died, some mice showed focal coagulative liver necrosis and two females showed hyperplasia of the thymus. The maximal tolerated dose was estimated by the authors to be 6200 mg/kg bw/day for females and 3200 mg/kg bw/day for males (Lijinsky and Kovatch, 1986). The NOAEL of this study is 1600 mg/kg bw/day.

A 25-week toxicity study was performed in which groups of five Fischer 344 rats of each sex were given drinking-water containing cyclohexanone at a concentration of 0, 190, 400, 800, 1600, 3300, 4700, or 6500 ppm, corresponding to 0, 30, 60, 120, 240, 500, 720, or 1000 mg/kg bw/day, respectively (Lijinsky and Kovatch, 1986). All the rats survived to termination of the study at 25 weeks. No observable effects were reported in male or female rats at doses up to 500 mg/kg bw/day. Two males at 720 mg/kg bw/day developed degenerative changes of the thyroid gland that were not seen in other animals in this study. Animals at the highest dose had a 10% depression in weight gain. No other adverse effects were observed. The maximal tolerated dose was calculated to be 1000 mg/kg bw/day (Lijinsky and Kovatch, 1986). The NOAEL of this study is 500 mg/kg bw/day.

4.2.3. Inhalation studies

No repeated dose inhalation toxicity studies are available for the cyclic acetates.

4.3. Mutagenicity and genotoxicity

Mutagenicity and genotoxicity testing with cyclic acetates or metabolites has been performed mostly *in vitro*, with one material (4-methyl-8-methylenetricyclo[3.3.1.$1^{3,7}$]decan-2-yl acetate) tested *in vivo*. The results of these tests are summarized in Tables 4-1 and 4-2.

4.3.1. In vitro mutagenicity studies

4.3.1.1. Indicator studies. In an assay for sister chromatid exchange, cyclohexanone at a concentration of 7.5 μl/ml gave weakly positive results in Chinese Hamster Ovary (CHO) cells in the absence of metabolic activation and negative results in the presence of metabolic activation (Aaron et al., 1985). A test for induction of unscheduled DNA synthesis was negative in fibroblasts in concentrations up to 9.48 mg/ml (NIOSH, 1980).

4.3.1.2. Mutation studies. The cyclic acetates 4-*tert*-butylcyclohexyl acetate, 4-methyl-8-methylenetricyclo[3.3.1.$1^{3,7}$]decan-2-yl acetate, 2-(1-methylpropyl)-1-vinyl-cyclohexyl acetate, myraldyl acetate and tricyclodecenyl acetate were inactive in bacterial mutagenicity assays (Ames tests) and cyclohexyl acetate was inactive in three rec-assays tested up to cytotoxic concentrations (see Table 4-1).

4-*tert*-Butylcyclohexanol was inactive in the Ames test using *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537, TA1538 with or without metabolic activation (HPVIS, 2008).

Negative results were reported in the standard assay for reverse mutation when various strains of *S. typhimurium* (TA98, TA100, TA1535, TA1537 and TA1538) were incubated with up to 10,000 μg/plate of cyclohexanone with or without metabolic activation (Florin et al., 1980; Haworth et al., 1983).

A positive result in an HPRT-test with cyclohexanone in CHO-cells was reported in absence but not in presence of S9-Mix in concentrations from 2.5 to 12.5 μg/ml (DuPont, 1984). However, in a mouse lymphoma test with up to 5 mg/ml no mutations were induced with and without metabolic activation (McGregor et al., 1988).

Abietic acid was negative in the Salmonella mutagenicity test in strains TA98, TA100, TA1535 and TA1548 with and without metabolic activation. It is unclear whether the substance was tested up to toxic concentrations (Greim, 2002).

4.3.1.3. Chromosome aberration studies. 4-*tert*-Butylcyclohexanol, was studied in Chinese Hamster V79 cells in the presence and absence of S9, and biologically significant increases in chromosomal aberrations were reported (HPVIS, 2008).

Older studies of chromosome aberrations with cyclohexanone (Collin, 1971; Lederer et al., 1971; Dyshlovoi et al., 1981). The studies by Collin and Dyshlovoi et al. are not reliable because of inadequate methods and reporting (Greim, 1998). Cyclohexanone did not induce chromosomal aberrations in Chinese Hamster ovary cells at a concentration of 7.5 μl/ml, with or without metabolic activation (Aaron et al., 1985).

4.3.2. In vivo studies

4-Methyl-8-methylenetricyclo[3.3.1.$1^{3,7}$]decan-2-yl was not genotoxic in the mouse bone marrow micronucleus test (RIFM, 1989d). No other *in vivo* genotoxicity studies with cyclic acetates are available.

A dominant lethal test in mice with cyclohexanone was negative in concentrations up to 400 ml/m^3 and rats did not show chromosome aberrations in the bone marrow (NIOSH, 1980).

4.4. Carcinogenicity

No bioassays are available for the cyclic acetates.

In a 2-year study groups of 41–52 $B6C3F_1$ mice of each sex were given drinking-water containing cyclohexanone at a concentration

of 0, 6500, 13,000 or 25,000 ppm (females only), corresponding to dietary intakes of 0, 1600, 3200 and 6200 mg/kg bw/day, respectively. Mice at the lowest dose and females at 3200 mg/kg bw/day showed no significant changes in weight; male mice at 3200 mg/kg bw/day and females at 6200 mg/kg bw/day had 15–20% lower weight than controls. No differences in survival rates were reported for animals receiving 1600 mg/kg bw/day. The survival of female mice at the two higher doses was poor. Histopathological examination of male mice at 1600 mg/kg bw/day revealed an increased incidence of proliferative lesions of the liver and lung, combined with a statistically significant ($p = 0.041$) increase in the incidence of benign and malignant hepatocellular neoplasms (25/51, 49%) when compared with controls (16/52, 31%). No benign or malignant neoplasms were seen at the two higher concentrations. Female mice at 1600 mg/kg bw/day had an increased incidence of malignant lymphomas (17/50, 34%), which was significant ($p = 0.036$) suggesting a weak carcinogenic effect (Lijinsky and Kovatch, 1986), although it was within the range of historical control rates (12–62%). A carcinogenic effect is unlikely as no dose-response was obtained; however, the high mortality in the higher dose groups might have masked one (Greim, 1998).

In another 2-year study with the same protocol, groups of 52 Fischer 344/N rats of each sex were given drinking-water containing cyclohexanone at a concentration of 0, 3300 or 6500 ppm, corresponding to 0, 330 or 650 mg/kg bw/day (Lijinsky and Kovatch, 1986). Rats in both treated groups had reduced mean body weights compared with the control group. A statistically significant increase in the incidence of adrenal cortex adenomas (7/52, 13%; $p = 0.030$) was reported in males at 330 mg/kg bw/day but no increase was found at 650 mg/kg bw/day (1/51, 2%). The authors noted that the incidence of adrenal cortex adenomas in Fischer 344/N rats in National Toxicology Program laboratories was approximately 1%. They concluded that, in the absence of a dose-response relationship, the increased incidence of benign neoplasms was not indicative of a carcinogenic response (Lijinsky and Kovatch, 1986). A statistically significant increase of follicular thyroid tumors (adenomas and carcinomas) in male rats was observed in the high-dose group. The study authors did not comment on this finding. Cyclohexanone is converted to cyclohexanol, which is conjugated with glucuronide, and a plausible explanation is that the induction of glucuronosyl transferase leads to a faster elimination of thyroid hormones with compensatory stimulation of thyroid growth causing tumors in the thyroid (Greim, 1998). The NOAEL in this 2-year study is 330 mg/kg bw/day.

4.5. Reproductive and developmental toxicity

4.5.1. Fertility

Histopathological examinations of the reproduction organs of rats in the 28-day repeated dose study with 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl (RIFM, 1990) showed no adverse effects (see Table 3).

Female CF1 mice were given cyclohexanone at a dose of 50 mg/day intraperitoneally for 28 days and were mated on day 10 of this treatment. The numbers of pregnancies and viable fetuses were similar to those of controls. The authors concluded that cyclohexanone had no effect on fertility in mice (Hall et al., 1974).

4.5.2. Developmental toxicity

A reliable developmental toxicity study has been conducted with one of the cyclic acetates (see Table 5).

The developmental toxicity of 4-*tert*-butylcyclohexyl acetate was investigated in Sprague-Dawley rats. Twenty-five presumed pregnant rats were dosed via gavage on days 7–20 of presumed gestation (GDs 7–20) with 4-*tert*-butylcyclohexyl acetate in corn oil at 0, 40, 160, or 640 mg/kg bw/day. One rat from the 640 mg/kg bw/day group was sacrificed on GD 20, due to body-weight loss and adverse clinical condition (i.e. decreased motor activity, apparent dehydration and excess salivation). Clinical signs in the remaining dams included excess salivation in the 160 and 640 mg/kg bw/day groups and sparse hair coat on the limbs, urine-stained abdominal fur and red perioral substance in the 640 mg/kg bw/day group (statistically significant increases). Body-weight gains were reduced (32%) in the 640 mg/kg bw/day group during the dosage period with weight loss at GD 7–8, GD 8–9. Absolute and relative feed consumption values were significantly reduced (29% and 26%, respectively) in the 640 mg/kg bw/day group. Pregnancy incidence and litter parameters were not affected by 4-*tert*-butylcyclohexyl acetate. Fetal body weights were significantly reduced (11%) in the 640 mg/kg bw/day group, and transient delays in fetal development were seen, including statistically significant increases in the fetal (but not the litter) incidences of moderate enlargement of the renal pelvices and reversible delays in ossification of the caudal vertebrae, fore- and hind-limb phalanges and hind-limb metatarsals. The maternal NOAEL was 160 mg/kg bw/day. The developmental NOAEL is 160 mg/kg bw/day based on reduced fetal body weights, transient delay in fetal development and reversible delays in skeletal ossification at 640 mg/kg bw/day. The NOEL was 40 mg/kg bw/day. These findings commonly occur with decreased maternal body weight and feed consumption (RIFM, 2007).

Postnatal behavior was examined in CD-1 mice (number not specified) given cyclohexanone at a dose of 800 mg/kg bw/day orally on days 8–12 of gestation. The offspring were tested for motor activity in the maze on days 22, 58 and 200 after parturition. No effect on motor activity was seen (Gray et al., 1986).

A group of 28 ICR/SIM mice was given cyclohexanone orally at a dose of 2200 mg/kg bw/day on days 8–12 of gestation. Six of the 28 mice died, and the remaining mice showed a significant decrease in body-weight gain. The litter size and number and the 2-day survival of the neonates were unaffected by treatment, but there was a significant decrease in live birth weight. (Seidenberg et al., 1986; Seidenburg and Becker, 1987).

4.6. Skin irritation

4.6.1. Human studies

Seventeen of the cyclic acetates have been well studied for their potential to produce skin irritation in humans (see Table 6-1).

Undiluted cyclohexyl acetate led to moderate skin reactions in a patch test, which was not further specified. Further studies with undiluted cyclic acetates in humans are not available.

No irritation was observed in predictive tests (e.g. in pre-tests for a maximization study with occlusive application for 48 h) with the highest tested concentrations, i.e., 20% 4-*tert*-butylcyclohexyl acetate (RIFM, 1976f), 20% tricyclodecenyl acetate (RIFM, 1977e), 12% amylcyclohexyl acetate (RIFM, 1975b), 12% decahydro-beta-naphthyl acetate (RIFM 1976f), 5% 1,3,3-trimethyl-2-norbornanyl acetate (RIFM 1975b), 4% 2-*tert*-butylcyclohexyl acetate (RIFM, 1976e), 4% *d*-cyclocitronellene acetate (RIFM, 1982e), 4% cyclohexyl acetate (RIFM, 1977e).

After application in human repeated insult patch tests (HRIPTs) little or no irritation was observed with the highest concentrations of the following materials: 20% (3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate (RIFM, 1982c), 20% octahydro-4,7-methano-1*H*-indenemethyl acetate (RIFM, 1976g), 20% 1,3,3-trimthyl-2-norbornanyl acetate (RIFM, 1975b), 10% myraldyl acetate (RIFM, 1977f), 6.25% 4-*tert*-butylcyclohexyl acetate (RIFM, 1964b), 6.25% 4-(isopropyl)-1-methylcyclohexyl acetate (RIFM, 1964c), 6.25% tricyclodecanyl acetate (RIFM, 1971b), 5% 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]-decan-2-yl acetate (RIFM 1996a,b,c,d), 2% 1-ethynylcyclohexyl acetate (RIFM, 1971a) and 2% tricyclodecenyl acetate (RIFM,

1960). Further details on studies of dermal irritation in humans are provided in Table 6-1.

4.6.2. Animal studies

Seventeen of the cyclic acetates have been tested in animal models of skin irritation using rabbits, rats, or guinea pigs (see Table 6-2).

If applied undiluted (15 of 17 studies), cyclic acetates showed slight to moderate irritation in almost all studies with rabbits or guinea pigs irrespective of the method used. Tests performed in accordance with current testing guidelines with 2-*tert*-butylcyclohexyl acetate, 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}] decan-2-yl acetate, and tricyclodecenyl acetate showed slight to moderate irritation.

No or only slight effects were observed in rabbits or guinea pigs with diluted compounds: 4-*tert*-butylcyclohexyl acetate 1% no reaction, 3% and 10% slight reaction with open application; *d*-cyclocitronellene acetate 10% or 20% very slight reaction; 1-ethynylcyclohexyl acetate 2% no reaction; 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]-decan-2-yl acetate 1% slight reaction; 2-(1-methyl-propyl)-1-vinylcyclohexyl acetate 10% no reaction; myraldyl acetate 1% no reaction; tricyclodecanyl acetate 6.25% no reaction.

Further details on studies of dermal irritation in animals are provided in Table 6-2.

4.7. Mucous membrane irritation

No human studies on mucous membrane irritation are available.

The potential to induce eye irritation has been studied in animals for 15 of the cyclic acetates (see Table 7).

Undiluted cyclic acetates showed no or only slight conjunctival reactions on the eye of rabbits: 2-*tert*-butylcyclohexyl acetate (RIFM, 1976j), (3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate (RIFM, 1982g), alpha-methylcyclohexylmethyl acetate (RIFM, 1977b), 2-(1-methylpropyl)-1-vinylcyclohexyl acetate (RIFM, 1977j), octahydro-4,7-methano-1*H*-indenemethyl acetate (RIFM, 1979j), 1,7,7-trimethylbicyclo[4.4.0]dec-3-yl acetate (RIFM, 1979k).

Slight to moderate effects were noted with undiluted amylcyclohexyl acetate (RIFM, 1964d), undiluted 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate (RIFM, 1989l), 2% 1-ethynylcyclohexyl acetate (RIFM, 1970d) and 6.25% tricyclodecanyl acetate (RIFM, 1970c). Only very slight reactions were found for 6.25% 4-(isopropyl)-1-methylcyclohexyl acetate (RIFM, 1963). Application of 0.1% *d*-cyclocitronellene acetate was without reaction; 0.25% produced slight effects, 0.5% and 1% showed corneal involvement with or without iris effects, respectively (RIFM, 1981d). Slight to moderate conjunctival irritation was reported with 0.625% 4-*tert*-butylcyclohexyl acetate (RIFM, 1964e) in an unreported vehicle; however, application of the undiluted substance only caused slight redness (RIFM, 1970a).

Serious corneal and iris effects were caused by a 50% solution 2-*tert*-butylcyclohexyl acetate and tricyclodecenyl acetate in Tween 80 (RIFM, 1976j, 1977l). Tested undiluted, 2-*tert*-butylcyclohexyl acetate was not an irritant and tricyclodecenyl acetate showed non-persistent corneal lesions, which healed within 3 days. It seems likely that Tween 80 contributes to the severity of these effects.

4.8. Skin sensitization

4.8.1. Human studies

Seventeen of the cyclic acetates under review have been evaluated for their potential to induce sensitization in humans (see Table 8-1).

No evidence of a sensitizing effect was observed with the following maximum concentrations of cyclic acetates tested in volunteers: amylcyclohexyl acetate (12% in petrolatum (pet.); RIFM, 1975b), 2-*tert*-butylcyclohexyl acetate (4% in pet.; RIFM, 1976e), 4-*tert*-butylcyclohexyl acetate (20% in pet.; RIFM, 1976f), *d*-cyclocitronellene acetate (4% in pet.; RIFM, 1982e), cyclohexyl acetate (4% in pet.; RIFM, 1974c, 1977e), decahydro-beta-naphthyl acetate (12% in pet.; RIFM, 1976f), 1-ethynylcyclohexyl acetate (2% in alcohol; RIFM, 1971a), (3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate (20% in pet.; RIFM, 1982c), 4-(isopropyl)-1-methylcyclohexyl acetate (6.25% in alcohol; RIFM, 1964c), alpha-methylcyclohexylmethyl acetate (100%; RIFM, 1977b), 4-methyl-8-methylenetricyclo-[3.3.1.1^{3,7}]decan-2-yl acetate (5% in alcohol, and in alcohol:DEP (3:1); RIFM, 1996a,b,c,d), myraldyl acetate (10% in dimethylphthalate; RIFM, 1977f), octahydro-4,7-methano-1*H*-indenemethyl acetate (20% in pet.; RIFM, 1976g), tricyclodecanyl acetate (6.25% in alcohol; RIFM, 1971b) tricyclodecenyl acetate (20% in pet.; RIFM, 1977e), 1.7.7-trimethylbicyclo[4.4.0]dec-3-yl acetate (20% in pet.; RIFM, 1979e) and 1,3,3-trimethyl-2-norbornanyl acetate (5% in pet.; RIFM, 1975b).

With the following substances, no sensitizing reactions were observed in diagnostic patch tests with dermatitis patients: 2-*tert*-butylcyclohexyl acetate and 4-*tert*-butylcyclohexyl acetate (Frosch et al., 1995).

Larsen et al. (2002) reported that 5% cyclohexyl acetate produced 1 (0.5%) reaction in 218 fragrance sensitive dermatitis patients.

There are data on abietic acid, the corresponding acid to the alcoholic moiety of abietyl acetate. Pure abietic acid is not a sensitizer in humans, however, it is a prohapten and oxidation products or structurally similar impurities are known sensitizers (Greim, 2002).

4.8.2. Animal studies

Information on the individual animal studies is provided in Table 8-2. In comparison to humans, potential for the cyclic acetates to induce sensitization in animals has been less studied.

Of the tested cyclic acetates, *d*-cyclocitronellene acetate (RIFM, 1981e), 2-(1-methylpropyl)-1-vinylcyclohexyl acetate (RIFM, 1976h; 1977h,m) and myraldyl acetate (RIFM, 1972c) were negative in guinea pig sensitization tests, and cyclohexyl acetate (Mallette and von Haam, 1952) was negative in a not-further-specified patch test with white rabbits.

4-Methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate gave positive reactions in the guinea pig Buehler test with 1% for challenge and 0.2% rechallenge (RIFM, 1989j,k) but no sensitization reaction was observed in the mouse local lymph node assay with concentrations up to 30% (RIFM, 2004).

4.9. Phototoxicity and photoallergenicity

Only limited data were available with regard to the phototoxicity and photoallergenicity of cyclic acetates (see Table 9). From human or animal studies reliable data were available on the phototoxicity and photoallergenicity of 2-(1-methylpropyl)-1-vinylcyclohexyl acetate, (3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate and 1.7.7-trimethylbicyclo[4.4.0]dec-3-yl acetate. Additionally 4-*tert*-butylcyclohexyl acetate was tested for its phototoxic potential.

No phototoxic and photoallergic reactions were seen in groups of 20 human volunteers exposed to 20% (3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate or 1.7.7-trimethylbicyclo[4.4.0]dec-3-yl acetate, followed by irradiation with UVA (RIFM, 1982c, 1979e). In guinea pigs, 2-(1-methylpropyl)-1-vinylcyclohexyl acetate was not phototoxic at 10% in alcohol and not photoallergic at 3% (RIFM, 1979g, 1980e). 4-*tert*-butylcyclohexyl acetate was found not to be

phototoxic in guinea pigs treated with 1%, 3% or 10% solutions in ethanol with 2% DMSO and irradiated with 20 J/cm^2 for an unspecified period of time (RIFM, 1983).

UV spectra have been obtained for 10 cyclic acetates (cyclohexyl acetate; 1,3,3-trimethyl-2-norbornanyl acetate; 1,7,7-trimethylbicyclo[4.4.0]dec-3-yl acetate; amylcyclohexyl acetate; 2-*t*-butylcyclohexanol acetate; 4-*t*-butylcyclohexanol acetate; decahydro-β-naphthyl acetate; octahydro-4,7-1*H*-indenemethyl acetate; (3a.α., 4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate, tricyclo[5.2.1.02,6]dec-4-en-8-yl acetate). Most of them absorbed UV light peaking in the UVC range (<290 nm). Based on the UV spectra (see Table 10) and review of the phototoxicity/photoallergy data, cyclic acetates would not be expected to elicit phototoxicity or photoallergy under the current conditions of use as a fragrance ingredient.

5. Summary

Maximum daily calculated systemic exposures via the dermal route for the materials under review were estimated by RIFM to range from 0.0003 (cyclohexyl acetate) to 0.246 mg/kg bw/day for *d*-cyclocitronellene acetate in users with high consumption of cosmetic products containing these materials (see Table 1). The maximum concentration for these materials in cosmetic products has been estimated or reported to be 0.001% (cyclohexyl acetate) to 3.56% (2-*tert*-butylcyclohexyl acetate). Data on inhalation exposure to the materials are not available.

The common characteristic structural element of cyclic acetates is the acetate unit bound to a mono-, bi- or tri-cyclic alcohol. After hydrolysis of the ester bond, acetic acid or – for one ester – cyclopent-2-eneacetic acid and primary, secondary or tertiary alcohols are formed.

Acetic acid is an endogenous compound occurring in intermediary metabolism. Metabolism data for cyclopent-2-eneacetic acid are not available.

Metabolism data on primary alcohols (derived from abietyl acetate, octahydro-4,7-methano-1*H*-indenemethyl acetate, and myraldyl acetate) are not available. It is expected that these primary alcohols will be oxidized via the aldehyde to the corresponding acids, the acidity of which is similar to acetic acid. Metabolism data on secondary alcohols such as cyclohexanol or alkyl-substituted cyclohexanols do exist. Unsubstituted or alkyl-substituted cyclohexanol is rapidly oxidized to the corresponding cyclohexanone derivative by alcohol dehydrogenase. Conversely, the cyclohexanone derivative may be reduced to cyclohexanol by cytosolic carbonyl reductases. Conjugation of the alcohol with glucuronic acid and excretion in the bile and urine is the predominant pathway for metabolic detoxification and elimination of cyclohexanol. The size, position, number, or stereochemistry of alkyl substituents on the cyclohexyl ring exerts no significant effect on the rate of metabolism of alkyl-substituted cyclohexanol or cyclohexanone derivatives, which are structurally related to metabolites of secondary monocyclic esters. Tertiary alcohols (derived from 1-ethynylcyclohexyl acetate; 4-(isopropyl)-1-methylcyclohexyl acetate, 1-methyl-2-(1-methylpropyl)cyclohexyl acetate, 1-methyl-4-(1-methylvinyl)cyclohexyl acetate, and 2-(1-methylpropyl)-1-vinylcyclohexyl acetate) can not be further oxidized. Conjugation of the alcohol group with glucuronic acid and elimination is to be expected as with other tertiary alcohols e.g. methyl- and ethy-*tert*-butyl alcohols (Bernauer et al., 1998).

Pharmacokinetic studies for the substances under review are not available. Therefore, dermal absorption is assumed to be 100%. Studies on alkyl-substituted cyclohexanol or cyclohexanone derivatives, which are structurally related to metabolites of acetates of secondary monocyclic alcohols, are rapidly absorbed through the gastrointestinal tract and excreted mainly in the urine. The size, position, number, or stereochemistry of alkyl substituents on the cyclohexyl ring exerts no significant effect on the rate of excretion.

Acute dermal toxicity has been reported for 13 of the 25 evaluated cyclic acetates. The LD_{50} values are generally greater than 2000 mg/kg bw indicating that these materials are of low toxicity or are practically nontoxic via the dermal route. Acute oral toxicity for 16 of the 25 evaluated cyclic acetates is very low with LD_{50} values in rats and mice between 2000 and 5000 mg/kg bw. Clinical signs reported were lethargy and piloerection after dermal or oral administration.

The database on repeated dose toxicity for the cyclic acetates is limited to two studies available with cyclic acetates and three studies with metabolites resulting from ester hydrolysis.

The repeated oral application (gavage) of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate/kg bw/day for 28 days led to increased salivation in all of the male and female rats given 55 mg/kg bw/day and further signs of toxicity were observed at 1000 mg/kg bw/day. The systemic NOAEL relevant for this safety assessment is therefore 55 mg/kg bw/day. Oral application (gavage) of 160 mg 4-*tert*-butylcyclohexyl acetate/kg bw/day for 14 days to pregnant rats also produced increased salivation and is the NOAEL. Further clinical signs and body-weight reduction were observed at 640 mg/kg bw/day.

The results of both studies with cyclic acetates indicate that salivation is the most sensitive effect; systemic effects are observed only at higher doses. As clinical signs but no salivation were observed in the study with 4-*tert*-butylcyclohexanol at 150 mg/kg bw/day it can be concluded that the release of the acetate group is responsible for the predominant irritating effects of the cyclic acetates.

Application of cyclohexanone in the drinking water of mice for 13 weeks resulted in reduced weight gain at 3200 mg/kg bw/day in mice. The no observed adverse effect level (NOAEL) was 1600 mg/kg bw/day. Addition of cyclohexanone to the drinking water of rats for 25 weeks resulted in degenerative changes of the thyroid gland in male rats at 720 mg/kg bw/day and depressed body-weight gain at 1000 mg/kg bw/day. The NOAEL was 500 mg/kg bw/day.

In summary, based on the low chemical reactivity of the cyclic alcohols and the studies available the cyclic acetates are considered to share a low systemic toxicity with their alcohol and ketone derivatives, but they exert locally irritating properties due to the acetate moiety. From the available data it can be concluded that with oral application under non-irritating conditions, systemic effects are unlikely to occur.

The five cyclic acetates tested (4-*tert*-butylcyclohexyl acetate, 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate, 2-(1-methylpropyl)-1-vinyl-cyclohexyl acetate, myraldyl acetate and tricyclodecenyl acetate) as well as 4-*tert*-butylcyclohexanol, the corresponding alcohol of 4-*tert*-butylcyclohexyl acetate, were inactive in bacterial genotoxicity tests. 4-*tert*-Butylcyclohexanol showed no clastogenic activity in V79 cells. Cyclohexanone did not induce unscheduled DNA synthesis in fibroblasts, chromosomal aberrations in CHO-cells and was inactive in bacterial mutagenicity tests. The cyclic acetate 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl, which was tested in the *in vivo* mouse micronucleus test, was not genotoxic. Cyclohexanone did not induce dominant lethal mutations in mice and chromosome aberrations in rats. It gave weakly positive results in an assay for sister chromatid exchange, which is only an indicator test, and HPRT mutations were induced only in the absence of the metabolic activation system.

Overall, the cyclic acetates and their metabolites tested were not genotoxic *in vitro* and *in vivo*; the saturated cyclic acetates not tested lack structural alerts for mutagenicity.

No bioassays on carcinogenicity are available for the cyclic acetates.

In 2-year carcinogenicity studies with cyclohexanone, incidences of tumors observed also in controls were increased at some doses, but no dose-dependent carcinogenic effects were observed. The authors concluded that the doses of 3200 mg/kg bw/day cyclohexanone for mice of each sex and 330 mg/kg bw/day for male rats were weakly carcinogenic. These conclusions should be evaluated in relation to the finding of no evidence of carcinogenicity with higher doses in female mice (6200 mg/kg bw/day) and in rats (650 mg/kg bw/day). These data were evaluated independently by a working group convened by the IARC (1989), which determined that cyclohexanone was "not classifiable as to its carcinogenicity to humans" (JECFA, 2007). Another evaluation came to the conclusion that cyclohexanone causes thyroid tumors but the mechanism is not clear. Because of the absence of positive findings in mutagenicity tests, a genotoxic mechanism is not plausible (Greim, 1998).

Reproductive and developmental toxicity data are limited. Histopathological examinations of the reproduction organs of rats in the 28-day repeated dose study with 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl showed no adverse effects up to 55 mg/kg bw/day. In a developmental toxicity study with 4-*tert*-butylcyclohexyl acetate a delay of fetal development was observed at maternal toxic doses of 640 mg/kg bw/day. The NOAEL for developmental toxicity of 160 mg/kg bw/day is far in excess of current human exposure level and raises no safety concern.

The potential for skin irritation by most of the cyclic acetates assessed in this report has been well characterized in humans and in experimental animals.

The animal data with rabbits and guinea pigs and one human study indicate that most of the cyclic acetates tested are likely to be slight to moderate skin irritants when topically applied undiluted. For the most part no or only minimal evidence of skin irritation was associated with concentrations in the range of 1–20%. The human studies performed show no evidence of irritation at current levels of use at 2–20% for individual cyclic acetates.

Cyclic acetates showed no or only slight conjunctival reactions in rabbits. Only some cyclic acetates exerted slight to moderate reactions: amylcyclohexyl acetate, 1-ethynylcyclohexyl acetate, tricyclodecanyl acetate, 4-(isopropyl)-1-methylcyclohexyl acetate, *d*-cyclocitronellene acetate, 4-*tert*-butylcyclohexyl acetate. In contrast, almost all cyclic acetates tested undiluted showed slight to moderate skin irritation.

The sensitization potential for 17 of the cyclic acetates has been well characterized in humans with supporting data from animal experiments for three acetates. For one acetate, only animal data exist.

None of the cyclic acetates tested demonstrated relevant sensitization potential: amylcyclohexyl acetate, 2-*tert*-butylcyclohexyl acetate, 4-*tert*-butylcyclohexyl acetate, *d*-cyclocitronellene acetate, cyclohexyl acetate, decahydro-beta-naphthyl acetate, 1-ethynylcyclohexyl acetate, (3a.α.,4.α.,6.α.,7.α.,7a.α.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1 H-inden-6-yl acetate, 4-(isopropyl)-1-methylcyclohexyl acetate, alpha-methylcyclohexylmethyl acetate, 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate, myraldyl acetate, octahydro-4,7-methano-1*H*-indenemethyl acetate, tricyclodecanyl acetate, tricyclodecenyl acetate, 1.7.7-trimethylbicyclo[4.4.0]dec-3-yl acetate and 1,3,3-trimethyl-2-norbornanyl acetate.

No phototoxic and photoallergic reactions were seen with four cyclic acetates in humans or animals.

6. Conclusion

The panel is of the opinion that there are no safety concerns regarding cyclic acetates under the present declared levels of use and exposure. These materials have not been evaluated at levels other than reported in this group summary. Use of these materials at higher maximum dermal levels or higher systemic exposure levels would require re-evaluation by the panel. This opinion was based on the following reasons:

- Minimal, if any, evidence of skin irritation in humans is associated with current levels of use at 2–20% for individual cyclic acetates. Even esters (octahydro-4,7-methano-1*H*-indenemethyl acetate and myraldyl acetate) that may be metabolized to both acetic acid and a cyclic acid (via the primary alcohol), have no effect concentrations for skin irritation in humans of 10–20%. The same is expected for abietyl acetate, because its possible metabolite abietic acid is a weak acid. Therefore, due to the structural similarities, compounds not tested for skin irritation in humans are expected to be of no concern provided concentrations in end products are in the range of 2–20%.
- These materials have no, or a low, sensitizing potential. Abietic acetate might give rise to sensitizing oxidation products. It should be ensured that abietic acetate is of high purity and, that oxidation is prevented in the end product.
- The cyclic acetates have a low order of acute toxicity.
- The cyclic acetates and the cyclic alcohols tested are of low systemic toxicity upon repeated dermal application. NOAELs for compounds or their metabolites are in the range of 50–500 mg/kg bw/day in rats. Changes indicative of enzyme induction in the liver (liver enlargement), α2micro-globulin-nephropathy in male rats and possibly thyroid hormone imbalance due to enzyme induction have been observed. Lower systemic toxicity in the drinking water studies compared to the oral studies might be related to the delivery method since a gavage bolus leads to higher substance concentrations in the blood than application via drinking water. As dermal application is more comparable to drinking water application, calculating margins of safety for dermal application from NOAELs of gavage studies is an additional precautionary approach.
- Compared to the estimated highest daily uptake of 0.246 mg/kg bw/day of *d*-cyclocitronellene acetate (100% dermal absorption) the margin of safety for this compound is at least 200 (estimated 50% oral absorption). For the other acetates with estimated daily doses (Table 1) the margin of safety ranges from 269 (myraldyl acetate) to 166,666 (cyclohexyl acetate).
- The five tested cyclic acetates (4-*tert*-butylcyclohexyl acetate, 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate, 2-(1-methylpropyl)-1-vinyl-cyclohexyl acetate, myraldyl acetate and tricyclodecenyl acetate) as well as 4-*tert*-butylcyclohexanol were inactive in bacterial mutagenicity tests. 4-*tert*-Butylcyclohexanol showed no clastogenic activity in V79 cells. Cyclohexanone was negative in a variety of *in vitro* and *in vivo* mutagenicity tests. 4-Methyl-8-methylenetricyclo[3.3.1.1^{3,7}]-decan-2-yl which was tested in the *in vivo* mouse micronucleus was not genotoxic *in vivo*.
- Data on carcinogenicity of cyclic acetates are not available but in view of the negative mutagenicity tests so far obtained. This is not of primary concern.
- Available genotoxicity data do not show a genotoxic potential of the substances.

Conflict of interest statement

This research was supported by the Research Institute for Fragrance Materials, an independent research institute that is funded by the manufacturers of fragrances and consumer products containing fragrances. The authors are all members of the Expert Panel of the Research Institute for Fragrance Materials, an independent group of experts who evaluate the safety of fragrance materials.

References

Aaron, C.S., Brewen, J.G., Bleicher, W.T., Spahn, M.C., Stetka, D.G., 1985. Comparative mutagenesis in mammalian cells (CHO) in culture: multiple genetic endpoint analysis of cyclohexanone in vitro. Environmental and Molecular Mutagenesis, 60–61.

Asakawa, Y., Ishida, T., Toyota, M., Takemoto, T., 1986. Terpenoid biotransformation in mammals IV. Biotransformation of (+)-longifolene, (−)-caryophyllene, (−)-caryophyllene oxide, (−)-cyclocolorenone, (+)-nootkatone, (−)-elemol, (−)-abietic acid and (+)-dehydroabietic acid in rabbits. Xenobiotica 16, 753–767.

Bernauer, U., Amberg, A., Scheutzow, D., Dekant, W., 1998. Biotransformation of ^{12}C- and 2-^{13}C-labeled methyl *tert*-butyl ether, ethyl *tert*-butyl ether, and *tert*-butyl alcohol in rats: identification of metabolites in urine by ^{13}C nuclear magnetic resonance and gas chromatography/mass spectrometry. Chemical Research in Toxicology 11, 651–658.

Cadby, A., Troy, W.R., Vey, M., 2002. Consumer exposure to fragrance ingredients: providing estimates for safety evaluation. Regulatory Toxicology and Pharmacology 36, 246–252.

Carpenter, C.P., Weil, C.S., Smyth Jr., H.F., 1974. Range-finding toxicity data: list VIII. Toxicology and Applied Pharmacology 28, 313–319.

Collin, J.P., 1971. Cytogenetic effect of cyclamate, cyclohexanone and cyclohexanol. Diabetes 19, 215–221.

Dyshlovoi, V.D., Boicko, N.L., Shemetum, A.M., Kharchenko, T.I., 1981. Cytogenic effect of cyclohexanone. Gigiena i Sanitariya 5, 76–77.

Elliott, T.H., Parke, D.V., Williams, R.T., 1959. Studies in detoxication. The metabolism of cyclo[(14)]hexane and its derivatives. Biochemical Journal 72, 193–200.

FFHPVC (The Flavor and Fragrance High Production Volume Consortia), 2003. The Cyclohexyl Derivatives Consortium Test Plan for Alkyl-substituted Cyclohexanol Derivatives, August 21. Submitted to EPA under the HPV Challenge Program. AR201. <http://www.epa.gov/opptintr/chemrtk/pubs/summaries/cyclodrv/c14686.pdf>.

Florin, I., Rutberg, L., Curvall, M., Enzell, C.R., 1980. Screening of tobacco smoke constituents for mutagenicity using the Ames' test. Toxicology 18, 219–232.

Ford, R.A., Domeyer, B., Easterday, O., Maier, K., Middleton, J., 2000. Regulatory Toxicology and Pharmacology 31, 166–181.

Frosch, P.J., Pilz, B., Andersen, K.E., Burrows, D., Camarasa, J.G., Dooms-Goossens, A., Ducombs, G., Fuchs, T., Hannuksela, M., Lachapelle, J.M., Lahti, A., Maibach, H.I., Menne, T., Rycroft, R.J.G., Shaw, S., Wahlberg, J.E., White, I.R., Wilkinson, J.D., 1995. Patch testing with fragrances: results of a multicenter study of the European environmental and contact dermatitis research group with 48 frequently used constituents of perfumes. Contact Dermatitis 33, 333–342.

Gray Jr., L.E., Kavlok, R.J., Ostby, J., Ferrell, J., Rogers, J., Gray, K., 1986. An evaluation of figure-eight maze activity and general behavioral development following prenatal exposure to forty chemicals: effects of cytosine arabinoside, dinocap, nitrofen, and vitamin A. Neurotoxicology 7, 449–462.

Greim, H. (Ed.), 1998. Cyclohexanone. Occupational Toxicants, vol. 10. Wiley-VCH, Weinheim.

Greim, H. (Ed.), 2002. Abietinsäure. Toxikologisch-arbeitsmedizinische Begründungen von MAK-Werten, vol. 34. Lieferung, Wiley-VCH, Weinheim.

Hall, I.H., Carlson, G.L., Abernethy, G.S., Piantadosi, C., 1974. Cycloalkanones. 4. Antifertility activity. Journal of Medical Chemistry 17, 1253–1257.

Haworth, S., Lawlor, T., Mortelmans, K., Speck, W., Zeiger, E., 1983. Salmonella mutagenicity test results for 250 chemicals. Environmental Mutagenesis 5 (Suppl. 1), 3–142.

HPVIS (High Production Volume Information System), 2008. Unpublished Reports from Degussa AG, 1988, 1997, 1999 on a Repeated Dose Toxicity Study, a Mutation Assay and a Cytogenetic Assay. Cited from the HPVIS Website. <http://iaspub.epa.gov/oppthpv/quicksearch.display?pChem=102093> (The original reports from these studies have not been reviewed as only summaries of the data have been made available for review).

IARC (International Agency for Research on Cancer), 1989. Monographs on the Evaluation of Carcinogenic Risks to Humans. Re-evaluation of Some Organic Chemicals, Hydrazine and Hydrogen Peroxide, vol. 71. World Health Organization, Lyon, France. pp. 1359–1364.

IFRA (International Fragrance Association), 2004. Volume of Use Survey.

IFRA (International Fragrance Association), 2007. Use Level Survey, September 2007.

Imai, T., 2006. Human carboxylesterase isozymes: catalytic properties and rational drug design. Drug Metabolism and Pharmacokinetics 21, 173–185.

JECFA, 2007. Safety evaluation of certain food additives. Alicyclic ketones, secondary alcohols and related esters. Prepared by the Joint FAO/WHO Expert Committee on Food Additives (JECFA). WHO Food Additives Series: 50. IPCS, WHO, Geneva (Draft).

Koeferl, M.T., Miller, T.R., Fisher, J.D., Martis, L., Garvin, P.J., Dorner, J.L., 1981. Influence of concentration and rate of intravenous administration on the toxicity of cyclohexanone in beagle dogs. Toxicology and Applied Pharmacology 59, 215–229.

Larsen, W., Nakayama, H., Fischer, T., Elsner, P., Frosch, P., Burrows, D., Jordan, W., Shaw, S., Wilkinson, J., Marks Jr., J., Sugawara, M., Nethercott, M., Nethercott, J., 2002. Fragrance contact dermatitis – a worldwide multicenter investigation (part III). Contact Dermatitis 46, 141–144.

Lederer, J., Collin, J.-P., Pottier-Arnould, A.-M., Gondry, E., 1971. L'action cytogenetique et teratogene du cyclamate et de ses metabolites. Therapeutique 47, 357–363.

Lijinsky, W., Kovatch, R.M., 1986. Chronic toxicity study of cyclohexanone in rats and mice. Journal of the National Cancer Institute 77, 941–949.

Lington, A.W., Bevan, C., 1994. Alcohols, fourth ed.. In: Clayton, G.D., Clayton, F.E. (Eds.), Patty's Industrial Hygiene Toxicology, vol. IID John Wiley and Sons, New York, pp. 2585–2760.

Madyastha, K.M., Srivatsan, V., 1988. Biotransformation of alpha-terpineol in the rat: its effects on the liver microsomal cytochrome p-450 system. Bulletin of Environmental Contamination and Toxicology 41, 17–25.

Mallette, F.S., Von Haam, E., 1952. Studies on the toxicity and skin effects of compounds used in the rubber and plastics industries II. Plasticizers. A.M.A. Archives of Industrial Hygiene and Occupational Medicine 6, 231–236.

Martis, L., Tolhurst, T., Koeferl, M.T., Miller, T.R., Darby, T.D., 1980. Disposition kinetics of cyclohexanone in beagle dogs. Toxicology and Applied Pharmacology 55, 545–553.

McGregor, D.B., Brown, A., Cattanach, P., Edwards, I., McBride, D., Riach, C., Caspary, W.J., 1988. Responses of the L5178Y tk+/tk− mouse lymphoma cell forward mutation assay: III 72 coded chemicals. Environmental and Molecular Mutagenesis 12, 85–153.

Mraz, J., Galova, E., Nohova, H., Vitkova, D., 1994. Uptake, metabolism and elimination of cyclohexanone in humans. Journal International Archives of Occupational and Environmental Health 66, 203–208.

Nelson, S.D., McClanahan, R.H., Thomassen, D., Gordon, W.P., Knebel, N., 1992. Investigations of mechanisms of reactive metabolite formation from (*R*)-(+)-pulegone. Xenobiotica 22, 1157–1164.

NIOSH (National Institute for Occupational Safety and Health), 1980. Tier II Mutagenic Screening of 13 NIOSH Priority Compounds: Cyclohexanone. Report No. 24, PB83-127571.

Oda, Y., Hamono, Y., Inoue, K., Yamamoto, H., Niihara, T., Kunita, N., 1978. Mutagenicity of food flavors in bacteria. Osaka-furitsu Koshu Eisei Kenkyu Hokoku Shokuhin Eisei Hen 9, 177–181.

RIFM (Research Institute for Fragrance Materials, Inc.), 1958. Acute oral toxicity study on rats. Unpublished Report from Givaudan Corporation. Report Number 15105, 24 November. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1960. Repeated insult patch test. Unpublished Report from Givaudan Corporation. Report Number 15107, 22 February. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1963. Eye irritation study in rabbits (FDA Draize method). Unpublished Report from IFF Incorporated. Report Number 50648, 23 September. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1964a. Repeated insult patch test. Unpublished Report from IFF Incorporated. Report Number 50545, 28 May. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1964b. Repeated insult patch test. Unpublished Report from IFF Incorporated. Report Number 47081, 28 April. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1964c. Repeated insult patch test. Unpublished Report from IFF Incorporated. Report Number 50647, 28 April. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1964d. Eye irritation test with amylcyclohexyl acetate (mixed isomers) in rabbits. Unpublished report from IFF Incorporated. Report Number 50544, 18 November. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1964e. Eye irritation test in rabbits. Unpublished Report from IFF Incorporated. Report Number 47082. 10 March. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1970a. Acute toxicity studies on *p-tert*-butyl cyclohexylacetate. Unpublished Report from BASF. Report Number 4419, 21 July. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1970b. Skin irritation study in rabbits. Unpublished Report from IFF Incorporated. Report Number 33788, 27 November. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1970c. Eye irritation study in rabbits. Unpublished Report from IFF Incorporated. Report Number 50439, 27 February. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1970d. Eye irritation study in rabbits. Unpublished Report from IFF Incorporated. Report Number 33789, 1 December. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1971a. Repeated insult patch test of group number 71. Unpublished Report from IFF Incorporated. Report Number 33790, 5 August. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1971b. Repeated insult patch test of group number 68. Unpublished Report from IFF Incorporated. Report Number 50441, 23 July. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1971c. Skin irritation study in rabbits. Unpublished Report from IFF Incorporated. Report Number 50438, 6 March. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1972a. Acute oral and intraperitoneal toxicity study of 2-(1-methylpropyl)-1-vinylcyclohexyl acetate in mice. Unpublished Report from Givaudan Corporation. Report Number 41938, 19 October. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1972b. The acute oral and intraperitoneal toxicity of myraldyl acetate in mice. Unpublished Report from Givaudan Corporation. Report Number 17231, 19 October. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1972c. Open epicutaneous testing of myraldyl acetate in guinea pigs. Unpublished Report from Givaudan

Corporation. Report Number 17232, 15 December. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1973. Toxicity study of 2-(1-methylpropyl)-1-vinylcyclohexyl acetate. Unpublished Report from Givaudan Corporation. Report Number 41939, 17 April. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1974a. Acute toxicity studies on rats, rabbits and guinea pigs. RIFM Report Number 1778, January–December. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1974b. Acute toxicity study in rats and rabbits. RIFM Report Number 2025, March–July. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1974c. Determination of potential hazards of fragrances materials by dermal contact. RIFM Report Number 34368, October 10. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1974d. Report on human maximization studies. RIFM Report Number 1801, February–December. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1975a. Acute toxicity studies on rats, rabbits and guinea pigs. RIFM Report Number 2020, May 22. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1975b. Report on human maximization studies. RIFM Report Number 1799, January–December. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1976a. Acute toxicity studies on rats, rabbits and guinea pigs. RIFM Report Number 2019, January–September. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1976b. Acute toxicity studies on rats and rabbits. RIFM Report Number 2016, May 21. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1976c. Acute toxicity studies with octahydro-4,5-methano-1*H*-indenemethyl acetate. Unpublished Report from Firmenich Incorporated. Report Number 37331, 2 August. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1976d. Acute oral toxicity test (mice). RIFM Report Number 49533, December 29. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1976e. Report on human maximization studies. RIFM Report Number 1797, November 11. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1976f. Report on human maximization studies. RIFM Report Number 1796, January–December RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1976g. Human repeated insult patch test with two test materials. Unpublished Report from Firmenich Incorporated. Report Number 37333, 16 August. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1976h. Capacity for allergenic sensitization determined by the test of Buehler performed on guinea pigs with 2-(1-methylpropyl)-1-vinylcyclohexyl acetate. Unpublished Report from Givaudan Corporation. Report Number 41610, 2 December. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1976i. Rabbit covered patch test. Unpublished Report from Quest International. Report Number 49534, 13 December. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1976j. Rabbit eye irritation test. Unpublished Study from Quest International. Report Number 46633, 4 July. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977a. Acute toxicity study in rats, rabbits and guinea pigs. RIFM Report Number 1695, January–October. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977b. Acute toxicity studies with alpha-methylcyclohexylmethyl acetate. Unpublished Report from IFF Incorporated. Report Number 52621, 11 March. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977c. Acute oral toxicity test (mice). Unpublished Report from Quest International. Report Number 46631, 10 June. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977d. Acute oral toxicity of 2-(1-methylpropyl)-1-vinylcyclohexyl acetate in rats. Unpublished Report from Givaudan Corporation. Report Number 41614, 1 March. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977e. Report on human maximization studies. RIFM Report Number 1691, May 16. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977f. Sensitization and irritation studies of myraldyl acetate in human subjects. Unpublished Report from Givaudan Corporation. Report Number 17234, 15 December. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977g. Rabbit covered patch skin irritation test. Unpublished Report from Quest International. Report Number 46632, 1 April. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977h. Skin irritation and capacity for allergic sensitization determined by the open epicutaneous test (OET) on guinea pigs. Unpublished Report from Givaudan Corporation. Report Number 41613, 18 March. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977i. Rabbit eye irritation test with 2-*tert*-butylcyclohexyl acetate. Unpublished Report from Quest International. Report Number 46634, 12 May. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977j. Irritation test on the rabbit eye. Unpublished Report from Givaudan Corporation. Report Number 41612, 18 March. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977k. Rabbit eye test. Unpublished Report from Quest International. Report Number 49535, 31 May. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977l. Rabbit eye test. Unpublished Report from Quest International. Report Number 49536, 19 July. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1977m. Capacity for allergic sensitization determined by the intradermal test with Freund's Complete Adjuvant (FCAT) on guinea pigs. Unpublished Report from Givaudan Corporation. Report Number 41611, 18 March. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1978a. Acute oral toxicity (LD_{50}) of amylcyclohexyl acetate (mixed isomers) in albino mice. Unpublished Report from IFF International. Report Number 50546, 24 May. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1978b. Acute oral toxicity test. Unpublished Report from Quest International. Report Number 46629, 19 June. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1978c. Acute oral toxicity test (mice). Unpublished Report from Quest International. Report Number 46709, 19 June. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979a. Acute dermal toxicity test in rabbits dose determination. Unpublished Report from IFF Incorporated. Report Number 47084, 6 March. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979b. Acute dermal toxicity of 1,7,7-trimethylbicyclo[4.4.0]dec-3-yl acetate in rabbits. Unpublished Report from Firmenich Incorporated. Report Number 17950, 5 September. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979c. Acute oral toxicity test in rats treated with 4-*tert*-butylcyclohexyl acetate. Unpublished Report from IFF Incorporated. Report Number 47083, 30 April. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979d. Acute oral toxicity of 1,7,7-trimethylbicyclo[4.4.0]dec-3-yl acetate in rats. Unpublished Report from Firmenich Incorporated. Report Number 17949, 5 September. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979e. Repeated insult patch test/photosensitization study of 1,7,7-trimethylbicyclo[4.4.0]dec-3-yl acetate in human subjects. Unpublished Report from Firmenich Incorporated. Report Number 17952, 10 September. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979f. Rabbit covered patch skin irritation test. Unpublished Report from Quest International. Report Number 46710, 20 November. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979g. Determination of phototoxicity in guinea pigs. Unpublished Report from Givaudan Corporation. Report Number 41609, 28 November. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979h. Rabbit covered patch test. Unpublished Report from Quest International. Report Number 46287, 8 December. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979i. Primary skin irritation study of 1,7,7-trimethylbicyclo[4.4.0]dec-3-yl acetate in rabbits. Unpublished Report from Firmenich Incorporated. Report Number 17951, 5 September. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979j. Primary ocular irritation (rabbits). Unpublished Report from Firmenich Incorporated. Report Number 37332, 13 August. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1979k. Primary eye irritation study of 1,7,7-trimethylbicyclo[4.4.0]dec-3-yl acetate in rabbits. Unpublished Report from Firmenich Incorporated. Report Number 17964, 5 September. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1980a. Acute oral range finding toxicity test (mice). Unpublished Report from Quest International. Report Number 46712, 4 February. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1980b. Acute oral range finding toxicity test (mice). Unpublished Report from Quest International. Report Number 46285, 4 February. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1980c. Ames metabolic activation test to assess the potential mutagenic effect of 4-*tert*-butylcyclohexyl acetate. Unpublished Report from Quest International. Report Number 46713, 5 February. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1980d. Ames metabolic activation test to assess the mutagenic potential effect of tricyclodecenyl acetate. Unpublished Report from Quest International. Report Number 46286, 20 February. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1980e. A guinea pig assay of photosensitizing potential. Unpublished Report from Givaudan Corporation. Report Number 41608, 4 February. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1981a. Acute oral range finding toxicity test (mice). Unpublished Report from Quest International. Report Number 46872, 8 July. RIFM, Woodcliff Lake, NJ, USA.
RIFM (Research Institute for Fragrance Materials, Inc.), 1981b. Acute oral toxicity to rats of *d*-cyclocitronellene acetate. Unpublished Report from BBA. Report Number 1773, 25 June. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1981c. Irritant effects of *d*-cyclocitronellene acetate on rabbit skin. Unpublished Report from BBA. Report Number 1770, 29 May. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1981d. Irritant effects of *d*-cyclocitronellene acetate on rabbit eye mucosa. Unpublished Report from BBA. Report Number 1772, 14 July and 27 August. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1981e. Screening test for delayed contact hypersensitivity with *d*-cyclocitronellene acetate in the albino guinea pig. Unpublished Report from BBA. Report Number 1771, 30 April. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1982a. Acute toxicity studies. RIFM Report Number 1689, February–September. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1982b. Acute dermal toxicity of (3a.alpha.,4.alpha.,6.alpha.,7.alpha.,7a.alpha.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate in albino rabbits. Unpublished Report from Firmenich Incorporated. Report Number 38871, 21 July. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1982c. Repeated insult patch test/photosensitization study of (3a.a.,4.a.,6.a.,7.a.,7a.a.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate in human subjects. Unpublished Report from Firmenich Incorporated. Report Number 38874, 21 May. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1982d. Single dose oral toxicity study of (3a.alpha.,4.alpha.,6.alpha.,7.alpha.,7a.alpha.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate in rats. Unpublished Report from Firmenich Incorporated. Report Number 38870, 21 July. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1982e. Report on human maximization studies. RIFM Report Number 1643, June 28. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1982f. Primary skin irritation study of (3a.alpha.,4.alpha.,6.alpha.,7.alpha.,7a.alpha.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate in rabbits. Unpublished Report from Firmenich Incorporated. Report number 38872, 30 April. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.). 1982g. Eye irritation study of (3a.alpha.,4.alpha.,6.alpha.,7.alpha.,7a.alpha.)-3a,4,5,6,7,7a-hexahydro-3-methyl-5-methylene-4,7-methano-1*H*-inden-6-yl acetate in rabbits. Unpublished Report from Firmenich Incorporated. Report Number 38873, 21 July. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1983. Determination of phototoxicity in guinea pigs. Unpublished Report from Givaudan Corporation. Report Number 33510, 24 May. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1984a. Acute oral toxicity study in rats. Unpublished Report from IFF Incorporated. Report Number 50649, 4 January. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1984b. Acute dermal irritation study. RIFM Report Number 1795, June 1. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1985. Acute dermal irritation study. RIFM Report Number 3099, June 1. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989a. Acute dermal toxicity to rats of 2-adamantanol, 4-methyl-8-methylene:acetate. Unpublished Report from IFF Incorporated. Report Number 47576, 29 November. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989b. Acute oral toxicity to rats of 2-adamantanol, 4-methyl-8-methylene:acetate. Unpublished Report from IFF Incorporated. Report Number 47541, 26 September. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989c. 2-Adamantanol, 4-methyl-8-methylene:acetate: bacterial mutation assay. Unpublished Report from IFF Incorporated. Report Number 48172, 29 September. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989d. Mouse micronucleus test on 4-methyl-8-methylenetricyclo[3.3.1.(3,7)]decan-2-yl acetate. Unpublished Report from IFF Incorporated. Report Number 48259, 19 December. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989e. Clinical safety evaluation of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate: repeated insult patch test. Unpublished Report from IFF Incorporated. Report Number 48481, 7 July. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989f. Clinical safety evaluation of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate: repeated insult patch test. Unpublished Report from IFF Incorporated, 6 July. Report Number 47623, September 9. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989g. Clinical safety evaluation of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate: repeated insult patch test. Unpublished Report from IFF Incorporated. Report Number 47622, 25 September. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989h. Clinical safety evaluation of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate: repeated insult patch test. Unpublished Report from IFF Incorporated. Report Number 48483, 25 September. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989i. Clinical safety evaluation of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate: repeated insult patch test. Unpublished Report from IFF Incorporated. Report Number 48482, 25 September. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989j. Delayed contact hypersensitivity in guinea pigs of 4-methyl-8-methylenetricyclo[3.3.1.(3,7)]-decan-2-yl acetate. Unpublished Report from IFF Incorporated. Report Number 47885, 26 May. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989k. Delayed contact hypersensitivity in guinea pigs with 4-methyl-8-methylenetricyclo[3.3.1.(3,7)]-decan-2-yl acetate. Unpublished Report from IFF Incorporated. Report Number 47624, 26 May. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989l. Irritant effects on the rabbit eye of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate. Unpublished Report from IFF Incorporated. Report Number 47685, 26 September. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1989m. Irritant effects on the rabbit skin of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate. Unpublished Report from IFF Incorporated. Report Number 47801, 18 September. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1990. Twenty-eight day oral toxicity study in rats with 4-methyl-8-methylenetricyclo[3.3.1.(3,7)]decan-2-yl acetate. Unpublished Report from IFF Incorporated. Report Number 48035, 22 June. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1991. 4-Methyl-8-methylenetricyclo[3.3.1.(3,7)]decan-2-yl acetate. Bacterial mutation assay. Unpublished Report from IFF Incorporated. Report Number 48172, 23 January. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1996a. Clinical safety evaluation of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate: repeated insult patch test. Unpublished Report from IFF Incorporated. Report Number 48484, 6 December. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1996b. Clinical safety evaluation of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate: repeated insult patch test. Unpublished Report from IFF Incorporated. Report Number 48485, 6 December. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1996c. Clinical safety evaluation of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate: repeated insult patch test. Unpublished Report from IFF Incorporated. Report Number 48486, 6 December. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 1996d. Clinical safety evaluation of 4-methyl-8-methylenetricyclo[3.3.1.1^{3,7}]decan-2-yl acetate: repeated insult patch test. Unpublished report from IFF Incorporated. Report Number 48487, 6 December. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 2003a. *Salmonella typhimurium* reverse mutation assay with 2-(1-methylpropyl)-1-vinylcyclohexyl acetate. Unpublished Report from Givaudan Corporation. Report Number 43079, 29 July. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 2003b. Myraldyl acetate: reverse mutation in five histidine-requiring strains of *Salmonella typhimurium*. Unpublished Report from Givaudan Corporation. Report Number 42164, 21 January. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 2004. Local lymph node assay. Unpublished Report from IFF Incorporated. Report Number 47808, 22 November. RIFM, Woodcliff Lake, NJ, USA.

RIFM (Research Institute for Fragrance Materials, Inc.), 2007. Oral (gavage) developmental toxicity study of 4-*tert*-butylcyclohexyl acetate (4-*t*BCHA) in rats. RIFM Report Number 52639, March 12. RIFM, Woodcliff Lake, NJ, USA.

SCOEL (Scientific Committee on Occupational Exposure Limits), 2001. Recommendation of the Scientific Committee on Occupational Exposure Limits for Acetic Acid. SCOEL/SUM 98 Final, June 2001.

Seidenburg, J.M., Becker, R.A., 1987. A summary of the results of 55 chemicals screened for developmental toxicity in mice. Teratogenesis, Carcinogenesis and Mutagenesis 7, 17–28.

Seidenberg, J.M., Anderson, D.G., Becker, R.A., 1986. Validation of an in vivo developmental toxicity screen in the mouse. Teratogenesis, Carcinogenesis and Mutagenesis 6, 361–374.

Söderberg, T.A., Johansson, A., Gref, R., 1996. Toxic effects of some conifer resin acids and tea tree oil on human epithelial and fibroblast cells. Toxicology 107, 99109.

SRC, 2007. Physprop Database, Syracuse Research Corporation.

Takenaka, T., Hasegawa, E., Takenaka, U., Saito, F., Odaka, T., 1986. Fundamental studies of safe compound perfumes for cosmetics. Part 1: The primary irritation of compound materials to the skin. Unknown Journal, 313–329.

TNO, 2008. In: Nijssen, L.M., van Ingen-Visscher, C.A., Donders, J., (Eds.), VCF Volatile Components in Food 10.1.1. <http://www.vcf-online.nl/VcfHome.cfm>.

Topping, D.C., Morgott, D.A., David, R.M., O'Donoghue, J.L., 1994. Ketones. In: Clayton, G.D., Clayton, F.E. (Eds.), Patty's Industrial Hygiene and Toxicology, fourth ed., vol. IIC. John Wiley and Sons, pp. 1739–1878.

Yoo, Y.S., 1986. Mutagenic and antimutagenic activities of flavoring agents used in foodstuffs. Osaka City Medical Journal 34, 267–288.

Yamaguchi, T., Caldwell, J., Farmer, P.B., 1994. Metabolic fate of [(3)*H*]-1-menthol in the rat. Drug Metabolism and Disposition 22, 616–624.

Zhu, Q.-G., Hu, J.-H., Liu, J.-Y., Lu, S.-W., Liu, Y.-X., Wang, J., 2007. Stereoselective characteristics and mechanism of epidermal carboxylesterase metabolism observed in HaCat keratinocytes. Biological and Pharmaceutical Bulletin 30, 532–536.