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May 2020

NATIONAL INDUSTRIAL CHEMICALS NOTIFICATION AND ASSESSMENT SCHEME (NICNAS)

PUBLIC REPORT

2*H*-Pyran-4-ol, tetrahydro-4-methyl-2-(2-methylpropyl)-, 4-acetate

This Assessment has been compiled in accordance with the provisions of the *Industrial Chemicals (Notification and Assessment) Act 1989* (the Act) and Regulations. This legislation is an Act of the Commonwealth of Australia. The National Industrial Chemicals Notification and Assessment Scheme (NICNAS) is administered by the Department of Health, and conducts the risk assessment for public health and occupational health and safety. The assessment of environmental risk is conducted by the Department of Agriculture, Water and the Environment.

This Public Report is available for viewing and downloading from the NICNAS website or available on request, free of charge, by contacting NICNAS. For requests and enquiries please contact the NICNAS Administration Coordinator at:

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Director
NICNAS

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SUMMARY

The following details will be published in the NICNAS *Chemical Gazette:*

ASSESSMENT REFERENCE	APPLICANT	CHEMICAL OR TRADE NAME	HAZARDOUS CHEMICAL	INTRODUCTION VOLUME	USE
STD/1718	BASF Australia Ltd	2*H*-Pyran-4-ol, tetrahydro-4-methyl-2-(2-methylpropyl)-, 4-acetate	Yes	< 10 tonnes per annum	Fragrance ingredient

CONCLUSIONS AND REGULATORY OBLIGATIONS

Hazard Classification
Based on the available information, the notified chemical is a hazardous chemical according to the *Globally Harmonised System of Classification and Labelling of Chemicals (GHS),* as adopted for industrial chemicals in Australia. The hazard classification applicable to the notified chemical is presented in the following table.

Hazard Classification	*Hazard Statement*
Skin sensitisation (Category 1B)	H317 – May cause an allergic skin reaction

The environmental hazard classification according to the *Globally Harmonised System of Classification and Labelling of Chemicals* (GHS) is presented below. Environmental classification under the GHS is not mandated in Australia and carries no legal status but is presented for information purposes.

Hazard Classification	*Hazard Statement*
Chronic Category 3	H412 – Harmful to aquatic life with long lasting effects

Human Health Risk Assessment
Under the conditions of the occupational settings described, the notified chemical is not considered to pose an unreasonable risk to the health of workers.

When used in the proposed manner, the notified chemical is not considered to pose an unreasonable risk to public health.

Environmental Risk Assessment
On the basis of the PEC/PNEC ratio, the notified chemical is not considered to pose an unreasonable risk to the environment.

Recommendations

REGULATORY CONTROLS

Hazard Classification and Labelling

- The notified chemical should be classified as follows:
  – Skin sensitisation (Category 1B): H317 – May cause an allergic skin reaction

  The above should be used for products/mixtures containing the notified chemical, if applicable, based on the concentration of the notified chemical present.

CONTROL MEASURES

Occupational Health and Safety

- A person conducting a business or undertaking at a workplace should implement the following engineering controls to minimise occupational exposure to the notified chemical during reformulation:
  - Enclosed , automated processes, where possible
  - Local exhaust ventilation and/or appropriate extraction systems, where possible

- A person conducting a business or undertaking at a workplace should implement the following safe work practices to minimise occupational exposure during handling of the notified chemical during reformulation:
  - Avoid contact with skin
  - Avoid inhalation of aerosols or mists

- A person conducting a business or undertaking at a workplace should ensure that the following personal protective equipment is used by workers to minimise occupational exposure to the notified chemical during reformulation :
  - Protective clothing
  - Impervious gloves
  - Respiratory protection if inhalation exposure may occur

  Guidance in selection of personal protective equipment can be obtained from Australian, Australian/New Zealand or other approved standards.

- A copy of the SDS should be easily accessible to employees.

- If products and mixtures containing the notified chemical are classified as hazardous to health in accordance with the *Globally Harmonised System of Classification and Labelling of Chemicals (GHS)* as adopted for industrial chemicals in Australia, workplace practices and control procedures consistent with provisions of State and Territory hazardous substances legislation should be in operation.

Health Surveillance

- As the notified chemical is a skin sensitiser, employers should carry out health surveillance for any worker who has been identified in the workplace risk assessment as having a significant risk of sensitisation.

Storage

- The handling and storage of the notified chemical should be in accordance with the Safe Work Australia Code of Practice for *Managing Risks of Hazardous Chemicals in the Workplace* (SWA, 2012) or relevant State or Territory Code of Practice.

Disposal

- Where reuse or recycling are not appropriate, dispose of the notified chemical in an environmentally sound manner in accordance with relevant Commonwealth, state, territory and local government legislation.

Regulatory Obligations

Secondary Notification
This risk assessment is based on the information available at the time of notification. The Director may call for the reassessment of the chemical under secondary notification provisions based on changes in certain circumstances. Under Section 64 of the *Industrial Chemicals (Notification and Assessment) Act (1989)* the notifier, as well as any other importer or manufacturer of the notified chemical, have post-assessment regulatory obligations to notify NICNAS when any of these circumstances change. These obligations apply even when the notified chemical is listed on the Australian Inventory of Chemical Substances (AICS).

Therefore, the Director of NICNAS must be notified in writing within 28 days by the notifier, other importer or manufacturer:

(1) Under Section 64(1) of the Act; if
- the final use concentration of the notified chemical exceeds 0.2% in cosmetic and household products;
- additional toxicological information becomes available on the notified chemical, in particular, studies on developmental toxicity;

or

(2) Under Section 64(2) of the Act; if
- the function or use of the chemical has changed from a fragrance ingredient, or is likely to change significantly;
- the amount of chemical being introduced has increased, or is likely to increase, significantly;
- the chemical has begun to be manufactured in Australia;
- additional information has become available to the person as to an adverse effect of the chemical on occupational health and safety, public health, or the environment.

The Director will then decide whether a reassessment (i.e. a secondary notification and assessment) is required.

Safety Data Sheet
The SDS of the notified chemical provided by the notifier was reviewed by NICNAS. The accuracy of the information on the SDS remains the responsibility of the applicant.

ASSESSMENT DETAILS

1. APPLICANT AND NOTIFICATION DETAILS

APPLICANT
BASF Australia Ltd (ABN: 62 008 437 867)
Level 12, 28 Freshwater Place
SOUTHBANK VIC 3006

NOTIFICATION CATEGORY
Standard: Chemical other than polymer (more than 1 tonne per year)

EXEMPT INFORMATION (SECTION 75 OF THE ACT)
Data items and details exempt from publication include: analytical data, degree of purity, impurities, import volume and identity of manufacturer.

VARIATION OF DATA REQUIREMENTS (SECTION 24 OF THE ACT)
Schedule data requirements are varied for melting point, boiling point, density, vapour pressure, dissociation constant, flammability, explosive properties, oxidising properties, repeated dose toxicity, and *in vitro* and *in vivo* genotoxicity.

PREVIOUS NOTIFICATION IN AUSTRALIA BY APPLICANT
None

NOTIFICATION IN OTHER COUNTRIES
EU (2017), Switzerland (2019)

2. IDENTITY OF CHEMICAL

MARKETING NAME
Pyranyl acetate

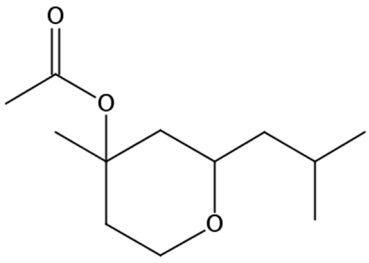
CAS NUMBER
131796-64-0

CHEMICAL NAME
2*H*-Pyran-4-ol, tetrahydro-4-methyl-2-(2-methylpropyl)-, 4-acetate

OTHER NAMES
EC 942-380-9
Tetrahydro-4-methyl-2-(2-methylpropyl)-, 4-acetate

MOLECULAR FORMULA
$C_{12}H_{22}O_3$

STRUCTURAL FORMULA



MOLECULAR WEIGHT
214.30 g/mol

ANALYTICAL DATA
Reference NMR, IR, UV/VIS spectra were provided.

3. COMPOSITION

DEGREE OF PURITY
> 98%

4. PHYSICAL AND CHEMICAL PROPERTIES

APPEARANCE AT 20 °C AND 101.3 kPa: colourless to yellow liquid

Property	*Value*	*Data Source/Justification*
Melting Point	< -20 °C	SDS
Boiling Point	223 °C at 101.3 kPa	SDS
Density	969 kg/m^3 at 20 °C	SDS
Vapour Pressure	1.95 x 10^{-5} kPa at 20 °C	SDS
Water Solubility	814 mg/L at 20 °C	Measured
Hydrolysis as a Function of pH	Not hydrolysable at pH 4-9	Measured
Partition Coefficient (n-octanol/water)	log Pow = 3.1 – 3.2 at 23 °C	Measured
Adsorption/Desorption	log K_{oc} = 1.96	Measured
Dissociation Constant	Not determined	Contains no dissociable functional groups
Flash Point	104 °C	Measured
Flammability	Combustible liquid	Based on flashpoint
Autoignition Temperature	284 °C	Measured
Explosive Properties	Not determined	Contains no functional groups that would imply explosive properties
Oxidising Properties	Not determined	Contains no functional groups that would imply oxidising properties

DISCUSSION OF PROPERTIES
For details of tests on physical and chemical properties, refer to Appendix A.

Reactivity
The notified chemical is expected to be stable under normal conditions of use.

Physical Hazard Classification
Based on the submitted physico-chemical data depicted in the above table, the notified chemical is not recommended for hazard classification according to the *Globally Harmonised System of Classification and Labelling of Chemicals (GHS),* as adopted for industrial chemicals in Australia.

The notified chemical has a flash point of 104 ºC which is greater than 93 °C. Based on *Australian Standard AS1940* definitions for combustible liquid, the notified chemical may be considered as a Class C2 combustible liquid if the chemical has a flash point below the boiling point.

5. INTRODUCTION AND USE INFORMATION

MODE OF INTRODUCTION OF NOTIFIED CHEMICAL (100%) OVER NEXT 5 YEARS
The notified chemical will not be manufactured in Australia. The notified chemical will be introduced into Australia in neat form for reformulation or in finished consumer products at ≤ 0.2% concentration.

MAXIMUM INTRODUCTION VOLUME OF NOTIFIED CHEMICAL (100%) OVER NEXT 5 YEARS

Year	*1*	*2*	*3*	*4*	*5*
Tonnes	< 2	< 5	< 10	< 10	< 10

PORT OF ENTRY
Melbourne, Sydney, Brisbane, Adelaide and Perth

TRANSPORTATION AND PACKAGING
The notified chemical will be imported neat in 217 L steel drums. Within Australia the drums will be transported by road to the warehouse for storage and later distributed to industrial customers by road for reformulation.

The notified chemical will also be imported as a component of finished consumer products at ≤ 0.2% concentration packed in containers suitable for retail sale. Finished consumer products containing the notified chemical will be transported primarily by road to retail stores in packages suitable for retail sale.

USE
The notified chemical will be used as a fragrance ingredient in cosmetic and household products at final use concentrations of ≤ 0.2% concentration.

OPERATION DESCRIPTION
Reformulation of the notified chemical into finished consumer goods may vary depending on the type of product and may involve both automated and manual transfer steps. Typically, reformulation processes may incorporate blending operations that are highly automated and occur in a fully enclosed/contained environment, followed by automated filling of the reformulated end-use products into containers of various sizes.

End-use products containing the notified chemical at ≤ 0.2% concentration will be used by consumers and professionals such as hairdressers, beauticians or cleaners. Depending on the nature of the product, these could be applied in a number of ways, such as by hand, using an applicator or sprayed.

6. HUMAN HEALTH IMPLICATIONS

6.1. Exposure Assessment

6.1.1. Occupational Exposure

CATEGORY OF WORKERS

Category of Worker	*Exposure Duration (hours/day)*	*Exposure Frequency (days/year)*
Transport and warehouse	none	incidental
Mixer	4	10 - 20
Drum handling	4	10 - 20
Drum cleaning/washing	4	10 - 20
Maintenance	4	10 - 20
Quality control	0.5	10 - 20
Professional end users	8	240

EXPOSURE DETAILS
Transport and storage
Transport, storage and warehouse workers may come into contact with the notified chemical in neat form or as a component of imported preparations, only in the unlikely event of accidental rupture of containers.

Reformulation
During reformulation, dermal, ocular and perhaps inhalation exposure of workers to the notified chemical at ≤ 100% concentration may occur during weighing and transfer stages, blending, quality control analysis, and cleaning and maintenance of equipment. The notifier states that exposure is expected to be minimised through the use of mechanical ventilation and/or enclosed systems, and through the use of personal protective equipment (PPE) such as protective clothing, goggles, impervious gloves and respiratory protection if required.

End-use
Exposure to the notified chemical in end-use products at ≤ 0.2% concentration may occur in professions where the services provided involve the application of cosmetics to clients (e.g. hair dressers and workers in beauty salons), or the use of household products in the cleaning industry. The principal route of exposure will be dermal, while ocular and inhalation exposure are also possible. Such professionals may use some PPE to minimise repeated exposure, and good hygiene practices are expected to be in place. If PPE is used, exposure of such workers is expected to be of a similar or lesser extent than that experienced by consumers using the products containing the notified chemical.

6.1.2. Public Exposure

There will be widespread and repeated exposure of the public to the notified chemical at ≤ 0.2% concentration through the use of a wide range of cosmetic and household products. The main route of exposure will be dermal, while ocular and inhalation exposure are also possible, particularly if products are applied by spray.

Data on typical use patterns of product categories in which the notified chemical may be used are shown in the following tables and these are based on information provided in various literatures (SCCS, 2012; Cadby *et al*., 2002; ACI, 2010; Loretz *et al.,* 2006). For the purposes of the exposure assessment, Australian use patterns for the various product categories are assumed to be similar to those in Europe. A dermal absorption (DA) rate of 100% was assumed for the notified chemical for calculation purposes. For the inhalation exposure assessment, a 2-zone approach was used (Steiling *et al*., 2014; Rothe *et al*., 2011; Earnest, Jr, 2009). An adult inhalation rate of 20 m^3/day (enHealth, 2012) was used and it was conservatively assumed that the fraction of the notified chemical inhaled is 50%. A lifetime average female body weight (BW) of 64 kg (enHealth, 2012) was used for calculation purposes.

Cosmetic products (Dermal exposure):

Product type	Amount (mg/day)	C (%)	RF (unitless)	Daily systemic exposure (mg/kg bw/day)
Body lotion	7820	0.2	1	0.2444
Face cream	1540	0.2	1	0.0481
Hand cream	2160	0.2	1	0.0675
Fine fragrances	750	0.2	1	0.0234
Deodorant spray	1430	0.2	1	0.0469
Shampoo	10460	0.2	0.01	0.0033
Conditioner	3920	0.2	0.01	0.0012
Shower gel	18670	0.2	0.01	0.0058
Hand soap	20000	0.2	0.01	0.0063
Hair styling products	4000	0.2	0.1	0.0125
Total				**0.4594**

C = maximum intended concentration of notified chemical; RF = retention factor.
Daily systemic exposure = (Amount × C × RF × DA)/BW

Household products (Indirect dermal exposure - from wearing clothes):

Product type	Amount (g/use)	C (%)	Product Retained (PR) (%)	Percent Transfer (PT) (%)	Daily systemic exposure (mg/kg bw/day)
Laundry liquid	230	0.2	0.25	10	0.0068
Fabric softener	90	0.2	0.25	10	0.0027
Total					**0.0095**

C = maximum intended concentration of notified chemical
Daily systemic exposure = (Amount × C × PR × PT × DA)/BW

Household products (Direct dermal exposure):

Product type	Frequency (use/day)	C (%)	Contact Area (cm^2)	Product Use C (g/cm^3)	Film Thickness (cm)	Time Scale Factor	Daily systemic exposure (mg/kg bw/day)
Laundry liquid	1.43	0.2	1980	0.01	0.01	0.007	0.0001
Dishwashing liquid	3	0.2	1980	0.0093	0.01	0.03	0.0005
All-purpose cleaner	1	0.2	1980	1	0.01	0.007	0.0043
Total							**0.0049**

C = maximum intended concentration of notified chemical
Daily systemic exposure = (Frequency × C × Contact area × Product Use Concentration × Film Thickness on skin × Time Scale Factor × DA)/BW

Hairspray (Inhalation exposure):

Product type	Amount (g/use)	C (%)	Inhalation rate (m^3/day)	Exposure duration zone 1 (min)	Exposure duration zone 2 (min)	Fraction inhaled (%)	Volume zone 1 (m^3)	Volume zone 2 (m^3)	Daily systemic exposure (mg/kg bw/day)
Hairspray	9.89	0.2	20	15	20	50	1	10	**0.0064**

C = maximum intended concentration of notified chemical

Total daily systemic exposure = Daily systemic exposure in Zone 1 [(amount × C × inhalation rate × exposure duration (zone 1) × fraction inhaled)/(volume (zone 1) × body weight)] + Daily systemic exposure in Zone 2 [(amount × C × inhalation rate × exposure duration (zone 2) × fraction inhaled)/(volume (zone 2) × body weight)]

The worst-case scenario estimation using these assumptions is for a person who is a simultaneous user of all products listed in the above tables that contain the notified chemical at the maximum intended concentrations specified by the notifier in various product types. This would result in a combined internal dose of 0.4802 mg/kg bw/day for the notified chemical. It is acknowledged that inhalation exposure to the notified chemical from use of other cosmetic and household products (in addition to hair spray) may occur. However, the combination of the conservative hair spray inhalation exposure assessment parameters used and the aggregate exposure from use of the dermally applied products (using a conservative 100% dermal absorption rate), are sufficiently protective to cover additional inhalation exposure to the notified chemical from use of other spray cosmetic and household products containing it with low exposure (e.g. air fresheners).

6.2. Human Health Effects Assessment

The results from toxicological investigations conducted on the notified chemical and an analogue chemical are summarised in the following table. For details of the studies, refer to Appendix B.

Endpoint	*Result and Assessment Conclusion*
Acute oral toxicity – rat	LD50 > 2,000 mg/kg bw; low toxicity
Acute dermal toxicity – rat	LD50 > 2,000 mg/kg bw; low toxicity
Skin irritation – *in vitro* EpiDerm™ reconstructed human epidermis test	non-irritant
Eye irritation – *in vitro* bovine corneal opacity and permeability (BCOP) test	non-irritant
Eye irritation – *in vitro* EpiOcular™ test	no prediction could be made
Skin sensitisation – mouse local lymph node assay	Weak sensitiser (EC3=36.5%)
Skin sensitisation – *in chemico* DPRA test	negative
Skin sensitisation – *in vitro* ARE-Nrf2 luciferase test	positive
Skin sensitisation – *in vitro* human cell line activation test (h-CLAT)	positive
Repeat dose oral toxicity – rat, 28 days*	NOAEL = 1,000 mg/kg bw/day in males and 300 mg/kg bw/day in females
Repeat dose dermal toxicity – rat, 90 days*	NOAEL = 1,000 mg/kg bw/day
Mutagenicity – bacterial reverse mutation	non mutagenic
Genotoxicity – *in vitro* chromosome aberration test in human lymphocytes*	non clastogenic
Genotoxicity – *in vitro* gene mutation test in Chinese hamster V79 cells*	non clastogenic
Genotoxicity – *in vivo* mouse micronucleus test*	non clastogenic
Reproductive/developmental toxicity screening test (dermal) – rat*	NOAEL (systemic and reproductive/developmental) = 1,000 mg/kg bw/day
Reproductive toxicity, extended one generation study (diet) – rat *	NOAEL (systemic) = 359 mg/kg bw/day NOAEL (reproductive/developmental) = 1,139 mg/kg bw/day
Prenatal developmental toxicity (dermal) – rat*	NOAEL (maternal and prenatal developmental) = 1,000 mg/kg bw/day
In vitro dermal penetration study*	17.92% penetration at 8,500 µg/cm^2 and 43.78% penetration at 902 µg/cm^2

*studies conducted on analogue chemical (Pyranol, CAS No. 63500-71-0)

The notified chemical is expected to be metabolised in the liver to pyranol (2*H*-pyran-4-ol, tetrahydro-4-methyl-2-(2-methylpropyl)-, CAS No. 63500-71-0) and acetic acid. The notifier has therefore provided read across data for pyranol (analogue chemical) to estimate the repeated dose toxicity and genotoxicity of the notified chemical.

Toxicokinetics

Given the low molecular weight (214.30 g/mol) of the notified chemical, absorption across biological membranes may occur. This is supported by the results of an *in vitro* dermal penetration study that demonstrate absorption of the analogue chemical through rat skin above 15% and 40% at high and low concentrations, respectively.

Acute Toxicity
The notified chemical is of low acute oral and dermal toxicity based on studies conducted in rats.

Irritation and Sensitisation
In an *in vitro* study using the EpiDerm™ reconstructed human epidermis test model, the notified chemical was determined not to require classification for skin irritation under the GHS according to the test guideline.

In an *in vitro* bovine corneal opacity and permeability (BCOP) test, the notified chemical was determined not to require classification for eye irritation under the GHS according to the test guideline. In another *in vitro* eye irritation test using the EpiOcular™ test method, no prediction could be made on the eye irritation potential of the notified chemical.

Sensitisation
The notified chemical was determined to be a weak skin sensitiser in a mouse local lymph node assay (LLNA) with stimulation indices of 4.57, 11.02 and 15.61 at 25%, 50% and 100%, respectively. The EC3 value (i.e. the estimated concentration of a test substance needed to produce a stimulation index of three) was calculated to be 36.5%.

One *in chemico* and two *in vitro* cell based assays were conducted to evaluate the skin sensitisation potential of the notified chemical. The tests are part of Integrated Approach to Testing and Assessment (IATA) which address specific events of the Adverse Outcome Pathway (AOP) leading to development of skin sensitisation (OECD, 2016). The tests are thus considered relevant for assessment of the skin sensitisation potential of the notified chemical, along with other supporting information.

The *in chemico* direct peptide reactivity assay (DPRA) aims to address the first key event (molecular initiation) of the AOP by measuring the interaction of the notified chemical with cysteine and lysine, small synthetic peptides representing the nucleophilic centres in skin proteins. The ARE-Nrf2 luciferase assay aims to address the second key event (keratinocyte activation) of the AOP by measuring the expression of a report luciferase gene under the control of a promoter from the antioxidant response element (ARE), a responding gene known to be upregulated by contact sensitisers. The *in vitro* h-CLAT assay aims to address the third key event (dendritic cell activation) of the AOP by measuring the expression of cell surface markers (i.e. CD54 and CD86) in human monocyte leukaemia cells (THP-1) upon stimulation with the notified chemical.

The notified chemical showed positive responses in two of the three tests (ARE-Nrf2 luciferase assay and h-CLAT test), suggesting potential for skin sensitisation.

Repeated Dose Toxicity
No repeat dose toxicity data were provided for the notified chemical.

In a 28 day repeated dose oral toxicity in rats with the analogue chemical at dose levels of 0, 100, 300 and 1,000 mg/kg bw/day, the No Observed Adverse Effect Level (NOAEL) was established as 1,000 mg/kg bw/day in males and 300 mg/kg bw/day in females, based on ataxia observed in high dose females. Two high dose females showed ataxia immediately after dosing on days 27 and 28. The study reported this effect was transient but test substance related. Statistically significant increase in mean absolute thyroid glands weight (26% increase compare to control recovery males) was observed in high dose recovery males, but not in high dose males.

In a 90 day repeated dose dermal toxicity study in rats with the analogue chemical at dose levels of 0, 100, 300 and 1,000 mg/kg bw/day, the NOAEL was established as 1,000 mg/kg bw/day in the study. However, mid (12% reduction compare to control females) and high (12% reduction than control females) dose females had statistically significant reduction in mean blood glucose level.

Mutagenicity/Genotoxicity
The notified chemical tested negative in a bacterial reverse mutation assay.

The analogue chemical tested negative in an *in vitro* chromosome aberration test in human lymphocytes, in an *in vitro* gene mutation test in Chinese hamster V79 cells and in an *in vivo* mouse micronucleus test.

Toxicity for Reproduction
No reproductive toxicity data were provided for the notified chemical.

In a reproductive/developmental toxicity screening test, rats were administered the analogue chemical dermally on GD 6-19at dose levels of 0, 100, 300 and 1,000 mg/kg bw/day. The NOAEL for systemic and reproductive/developmental toxicity was established in the study as 1,000 mg/kg bw/day, based on the absence of treatment related adverse effects up to the highest dose tested.

In an extended one generation reproductive toxicity study, rats were administered the analogue chemical in the diet at dose levels of ~ 90 (1,000 ppm), ~ 359 (4,000 ppm) and 1,113 (12,500 ppm) mg/kg bw/day. The NOAEL for systemic toxicity was 359 mg/kg bw/day based on decreased food consumption in F0 parental animals and decreased bodyweight/bodyweight gain in adolescent and adult F1 offspring at 1,113 mg/kg bw/day. The NOAEL for reproductive and developmental toxicity was established as 1,113 mg/kg bw/day in this study. However, there was statistically significant increases in the percentage of abnormal sperms in the cauda epididymidis of high dose males (the value, however, was within the historical control range), mean absolute and relative seminal vessels in mid and high dose males. High dose females had statistically significantly increased mean thyroid weights.

High dose female pups showed statistically significant increase in the mean number of days required to reach vaginal opening and this was 31.5, 31.7, 31.9 and 32.7 for control, low, mid and high dose pups, respectively. However, the study authors stated that the later onset of puberty in high dose pups is most likely a consequence of a general developmental delay and not a specific effect on the timing of puberty.

First preputial separation in male F1 pup was observed on PND 38 and the last was on PND 52. High dose male group showed statistically significant increase in preputial separation and the mean number of days to reach the criterion was 41.6, 42.1, 42.4 and 43.2 for control, low, mid and high dose groups, respectively. The study authors stated the increase in high dose males was within the historical control value.

Statistically significantly reduced blood glucose levels were observed in low, mid and high dose males, and increased blood urea levels in mid and high dose males.

In a prenatal developmental toxicity study, rats were administered the analogue chemical dermally on GD 6-19 at dose levels of 0, 100, 300 and 1,000 mg/kg bw/day. The NOAEL for maternal and prenatal developmental toxicity was established as 1,000 mg/kg bw/day in the study. Reproductive parameters were not affected by the treatment. However there were some foetal effects reported: One high dose male foetus (out of 236 foetuses) showed limb hyperextension (considered as spontaneous in nature); One skeletal malformation affecting the forelimb was observed in high dose group (was within the historical data in a comparable frequency); Skeletal variations of different bone structures (supernumerary thoracic vertebra and wavy ribs)were observed in all groups, with or without effects on corresponding cartilages (comparable to the historical control data) and statistically significant increase in supernumerary thoracic vertebra in mid dose foetus and statistically significant increase in wavy rib in high dose foetus (marginally above the historical control data). These effects could imply that developmental NOAEL could be below the reported value.

Health Hazard Classification

Based on the available information, the notified chemical is a hazardous chemical according to the *Globally Harmonised System of Classification and Labelling of Chemicals (GHS),* as adopted for industrial chemicals in Australia. The hazard classification applicable to the notified chemical is presented in the following table.

Hazard Classification	*Hazard Statement*
Skin sensitisation (Category 1B)	H317 – May cause an allergic skin reaction

6.3. Human Health Risk Characterisation

6.3.1. Occupational Health and Safety

Based on the toxicological information provided, the notified chemical is a weak skin sensitiser. Developmental effects at high doses could not be ruled out based on the information available on the analogue chemical.

Reformulation

Workers may experience dermal, ocular and perhaps inhalation exposure to the notified chemical at ≤ 100% concentration during reformulation. Given the notified chemical is a skin sensitiser caution should be exercised when handling the notified chemical during reformulation processes.

Provided that control measures are in place to minimise worker exposure, including the use of enclosed, automated processes and PPE such as impervious gloves, protective clothing and respiratory protection (if inhalation exposure

may occur), the risk to the health of workers during the handling of the notified chemical is not considered to be unreasonable.

End-use
Cleaners and beauty care professionals will handle the notified chemical at ≤ 0.2% concentration, similar to public use. Such professionals may use PPE to minimise repeated exposure, and good hygiene practices are expected to be in place. Therefore, the risk to workers who use products containing the notified chemical is expected to be of a similar or lesser extent than consumers who use such products on a regular basis. For details of the public health risk assessment see section 6.3.2 below.

6.3.2. Public Health

Members of the public may experience repeated exposure to the notified chemical through the use of cosmetic and household products containing the notified chemical at ≤ 0.2% concentration.

Sensitisation
Based on the results of an LLNA, the notified chemical is a skin sensitiser with an EC3 value of 36.5%. Using deodorant as a worst-case example of leave-on cosmetic products that may contain the notified chemical at ≤ 0.2% concentration, the Consumer Exposure Level (CEL) is estimated to be 15.00 μg/cm^2/day (Cadby *et al*., 2002). Consideration of available information and application of appropriate safety factors, an Acceptable Exposure Level (AEL) of 29.47 μg/cm^2/day is estimated for the notified chemical. In this instance, the factors employed included an interspecies factor (3), intraspecies factor (10), a matrix factor (3.16), use/time factor (3.16) and database factor (1), giving an overall safety factor of 300.

As the AEL > CEL, the risk to the public of the induction of sensitisation that is associated with the use of the notified chemical in deodorants at ≤ 0.2% concentration (a worst-case example of leave-on cosmetic products) is not considered to be unreasonable. Based on lower expected exposure level from other cosmetic and household products, by inference, the risk of induction of sensitisation associated with the use of these products is also not considered to be unreasonable. However, it is acknowledged that consumers may be exposed to multiple products containing the notified chemical, and a quantitative assessment based on aggregate exposure has not been conducted.

Repeated dose toxicity
The repeat dose toxicity potential was estimated by calculation of the margin of exposure (MoE) of the notified chemical using the worst case exposure scenario from use of multiple products by an individual with total exposure of 0.4802 mg/kg bw/day (see Section 6.1.2). Using a NOAEL of 300 mg/kg bw/day for the notified chemical (derived from a 28 day repeated dose toxicity study in rats on an analogue chemical), the margin of exposure (MoE) was estimated to be 624.7. A MoE value ≥ 100 is generally considered to be acceptable for taking into account intra- and inter-species differences.

Overall, based on the information available, the risk to the public associated with use of the notified chemical at ≤ 0.2% in cosmetic and household products, is not considered to be unreasonable.

7. ENVIRONMENTAL IMPLICATIONS

7.1. Environmental Exposure & Fate Assessment

7.1.1. Environmental Exposure

RELEASE OF CHEMICAL AT SITE
The notified chemical is not manufactured in Australia, therefore no release is expected from this activity. The notifier estimates that up to 1% of the import volume may be lost from accidental spills during transport and a further 1% of the import volume may be lost from accidental spills during reformulation. Any accidental spills are to be collected and disposed of in accordance with local government regulations. Wash waters from equipment cleaning containing the notified chemical are expected to be disposed of to sewer as trade waste.

RELEASE OF CHEMICAL FROM USE
The majority of the notified chemical is expected to be washed into sewers as a part of its use in various cosmetic and household products, where it will be treated in sewage treatment plants nationwide before being released into surface waters.

RELEASE OF CHEMICAL FROM DISPOSAL

A small proportion of the notified chemical is expected to remain as residues in empty product containers. These containers are expected to be either recycled or disposed of to domestic landfill. Collected wastes of the notified chemical are to be disposed of by licensed waste contractors to eventually be disposed of to landfill or released into the sewer system.

7.1.2. Environmental Fate

Following its use in cosmetic and household cleaning products, the notified chemical is expected to be primarily released into the sewer system and treated at sewage treatment plants before release to surface waters nationwide.

Two biodegradation tests were conducted on the notified chemical. One test conducted (301B method) showed 86% biodegradation after 28 days, however the 10-day window was not met. The other test conducted (302C method) showed 9% degradation after 28 days, however, transformation products were detected in the test solution indicating primary biodegradation. Based on these tests the notified chemical is not considered readily biodegradable but is expected to degrade into simpler organic compounds. For further details on the biodegradation studies refer to Appendix C. The notified chemical is not expected to bioaccumulate due to its log Pow (log Pow = 3.1 – 3.2). Some of the notified chemical may remain in the end use and bulk containers, which are either recycled or disposed of to landfill. In surface waters and landfill, the notified chemical is expected to degrade into water and oxides of carbon.

7.1.3. Predicted Environmental Concentration (PEC)

The use pattern will result in most of the notified chemical being washed into the sewer. The predicted environmental concentration (PEC) has been calculated assuming the realistic worst-case scenario with 100% release of the notified chemical into sewer systems nationwide over 365 days per annum. The extent to which the notified chemical is removed from the effluent in STP processes based on the properties of the notified chemical has not been considered for this scenario, and therefore no removal of the notified chemical during sewage treatment processes, is assumed. The PEC in sewage effluent on a nationwide basis is estimated as follows:

Predicted Environmental Concentration (PEC) for the Aquatic Compartment		
Total Annual Import/Manufactured Volume	10,000	kg/year
Proportion expected to be released to sewer	100%	
Annual quantity of chemical released to sewer	10,000	kg/year
Days per year where release occurs	365	days/year
Daily chemical release:	27.4	kg/day
Water use	200	L/person/day
Population of Australia (Millions)	24.386	Million
Removal within STP	0%	
Daily effluent production:	4,877	ML
Dilution Factor – River	1	
Dilution Factor – Ocean	10	
PEC - River:	5.62	µg/L
PEC - Ocean:	0.56	µg/L

STP effluent re-use for irrigation occurs throughout Australia. The agricultural irrigation application rate is assumed to be 1000 L/m^2/year (10 ML/ha/year). The notified chemical in this volume is assumed to infiltrate and accumulate in the top 10 cm of soil (density 1500 kg/m^3). Using these assumptions, irrigation with a concentration of 5.618 µg/L may potentially result in a soil concentration of approximately 3.745×10^{-2} mg/kg. Assuming accumulation of the notified chemical in soil for 5 and 10 years under repeated irrigation, the concentration of notified chemical in the applied soil in 5 and 10 years may be approximately 1.873×10^{-1} mg/kg and 3.745×10^{-1} mg/kg, respectively.

7.2. Environmental Effects Assessment

The results from ecotoxicological investigations conducted on the notified chemical are summarised in the table below. Details of these studies can be found in Appendix C.

Endpoint	*Result*	*Assessment Conclusion*
Fish Toxicity	LC50 = 74.9 mg/L	Harmful to fish
Daphnia Toxicity	EC50 = 22.3 mg/L	Harmful to aquatic invertebrates
Algal Toxicity	EC50 = 74.6 mg/L NOEC = 17.1 mg/L	Harmful to algal growth

Inhibition of Bacterial Respiration	IC50 > 1,000 mg/L	Not harmful to bacterial respiration

Based on the above ecotoxicological endpoints for the notified chemical, the notified chemical is expected to be harmful to fish, daphnia and algal growth. Therefore, the notified chemical is classified as 'Category 3 H402 – Harmful to aquatic life' according to the Globally Harmonised System of Classification and Labelling of Chemicals (GHS) (United Nations, 2009). The notified chemical is not biodegradable but is not expected to bioaccumulate. Therefore, the notified chemical is formally classified as 'Category 3 H412 – Harmful to aquatic life with long lasting effects' under the GHS for its long-term hazard.

7.2.1. Predicted No-Effect Concentration

A Predicted No-Effect Concentration (PNEC) was calculated based on the most sensitive acute endpoint for daphnia (EC50 = 22.3 mg/L) using an assessment factor of 100 as three acute trophic endpoints are available.

Predicted No-Effect Concentration (PNEC) for the Aquatic Compartment		
EC50 (Invertebrates).	22.30	mg/L
Assessment Factor	100.00	
Mitigation Factor	1.00	
PNEC:	223.00	µg/L

7.3. Environmental Risk Assessment

Risk Assessment	***PEC µg/L***	***PNEC µg/L***	***Q***
Q - River:	5.62	223	0.03
Q - Ocean:	0.56	223	<0.01

The risk quotient (Q=PEC/PNEC) has been calculated based on the worst-case assumption of complete release into the waterways with no removal in STPs. As the Q value is less than 1 the notified chemical unlikely to reach ecotoxicologically significant concentrations. Therefore, on the basis of the PEC/PNEC ratio, the notified chemical is not considered to pose an unreasonable risk to the environment.

APPENDIX A: PHYSICAL AND CHEMICAL PROPERTIES

Water Solubility 814 mg/L at 20 °C

Method OECD TG 105 Water Solubility
EC Council Regulation No 440/2008 A.6 Water Solubility
Remarks Flask Method
Test Facility BASF (2015a)

Hydrolysis as a Function of pH

Method OECD TG 111 Hydrolysis as a Function of pH

pH	*T (°C)*	*$t_{½}$ (hours)*
4	50	Hydrolytically stable
7	50	Hydrolytically stable
9	50	284.5
9	20	5,800.9

Remarks Measured using HPLC-UV
Test Facility BASF (2015b)

Partition Coefficient (n-octanol/water) log K_{ow} (major component) = 3.2 at 23 °C
log K_{ow} (minor component) = 3.1 at 23 °C

Method OECD TG 117 Partition Coefficient (n-octanol/water).
Remarks HPLC Method
Test Facility BASF (2015c)

Adsorption/Desorption – main test log K_{oc} = 1.96

Method OECD TG 106 Adsorption – Desorption Using a Batch Equilibrium Method

Soil Name	*Soil Type*	*Organic Carbon Content (%)*	*pH*	*Koc (mL/g)*	*log Koc*
Red soils	Agricultural soil	2.0	5.06	67	1.83
Yellow brown soils	Agricultural soil	3.23	6.38	178	2.25
Chestnut soils	Timbered soil	2.67	6.97	167	2.22
Alpine meadow soils	Timbered soil	5.48	7.78	80.3	1.9
Black soils	Timbered soil	4.17	4.39	41	1.61

Remarks Analysis by mass balance recovery.
Test Facility BSAL (2019a)

Flash Point 104 °C

Method DIN EN ISO 2719 (2016-11) Determination of Flash Point
Remarks Determined by Pensky-Martens closed cup method.
Test Facility BASF (2015d)

Autoignition Temperature 284 °C

Method DIN EN 14522 Determination of the Auto-Ignition Temperature of Liquids and Gases
Test Facility BASF (2015d)

APPENDIX B: TOXICOLOGICAL INVESTIGATIONS

B.1. Acute Oral Toxicity – Rat

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 423 Acute Oral Toxicity – Acute Toxic Class Method (2001)
Species/Strain	Rat/Wistar/Crl:WI (Han)
Vehicle	Nil
Remarks – Method	No protocol deviations.

RESULTS

Group	*Number and Sex of Animals*	*Dose (mg/kg bw)*	*Mortality*
1	3F	2,000	0/3
2	3F	2,000	0/3

LD50	> 2,000 mg/kg bw
Signs of Toxicity	All six animals showed an impaired general state and piloerection 3 hours after dosing which persisted in five animals at the 5 hour observation. The symptoms persisted in one animal at the day 1 observation.
Effects in Organs	No abnormalities were observed at necropsy.
Remarks – Results	Normal bodyweight gain was observed in all animals.
CONCLUSION	The notified chemical is of low acute toxicity via the oral route.
TEST FACILITY	Bioassay (2017a)

B.2. Acute Dermal Toxicity – Rat

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 402 Acute Dermal Toxicity (1987)
Species/Strain	Rat/ Wistar / Crl:WI (Han) SPF
Vehicle	Nil
Type of dressing	Semi-occlusive
Remarks – Method	No protocol deviations.

RESULTS

Group	*Number and Sex of Animals*	*Dose (mg/kg bw)*	*Mortality*
1	5M/5F	2,000	0/10

LD50	> 2,000 mg/kg bw
Signs of Toxicity – Local	No signs of local toxicity were observed.
Signs of Toxicity – Systemic	No signs of systemic toxicity were observed.
Effects in Organs	No abnormalities were observed at necropsy.
Remarks – Results	Normal bodyweight gain was observed during the study.
CONCLUSION	The notified chemical is of low acute toxicity via the dermal route.
TEST FACILITY	Bioassay (2017b)

B.3. Skin Irritation – *In Vitro* EpiDerm™ Reconstructed Human Epidermis Test

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 439 *In vitro* Skin Irritation: Reconstructed Human Epidermis Test Method
Vehicle	Nil

Remarks – Method

In the first test, the standard deviation of %-viability was 22.6% which was out of the acceptance range of ≤ 18%. Moreover, the viability of the single tissues did not indicate a clear prediction (values for single tissues were 60.3%, 33.2% and 78.2%). Hence, a second test was performed to clarify the result.

A pre-test using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolim bromide (MTT) showed the test substance does not directly reduce MTT.

Positive and negative controls were run in parallel with the test substance:
- Negative control: phosphate buffered saline
- Positive control: sodium dodecyl sulphate (5% aqueous solution)

No significant protocol deviations.

RESULTS

Test 1

Test Material	*Mean OD_{570} of Triplicate Tissues*	*Relative Mean Viability (%)*	*SD of Relative Mean Viability*
Negative control	2.090	100	4.4
Test substance	1.196	57.2	22.6
Positive control	0.051	2.4	0.1

Test 2

Test Material	*Mean OD_{570} of Triplicate Tissues*	*Relative Mean Viability (%)*	*SD of Relative Mean Viability*
Negative control	1.821	100	8.8
Test substance	1.333	73.2	4.1
Positive control	0.060	3.3	0.3

OD = optical density; SD = standard deviation

Remarks – Results

Based on the mean tissue viability of > 50% in Test 2, the test substance is not considered as irritating to the skin according to the test guideline.

Positive and negative controls performed as expected.

CONCLUSION

The notified chemical is not considered as irritating to the skin requiring classification of it as a skin irritant according to the GHS criteria.

TEST FACILITY

BASF (2017a)

B.4. Eye Irritation – *In Vitro* Bovine Corneal Opacity and Permeability Test

TEST SUBSTANCE

Notified chemical

METHOD

OECD TG 437 Bovine Corneal Opacity and Permeability Test Method for Identifying i) Chemicals Inducing Serious Eye Damage and ii) Chemicals Not Requiring Classification for Eye Irritation or Serious Eye Damage (2013)

Vehicle

Nil

Remarks – Method

Two positive controls were used.

Negative control: deionised water
Positive controls: ethanol (100%) (PC1) and dimethylformamide (100%) (PC2).

RESULTS

Test Material	*Mean Opacities of Triplicate Tissues (SD)*	*Mean Permeabilities of Triplicate Tissues (SD)*	*IVIS (SD)*
Negative control	5.7 (4.1)	0.005 (0.001)	5.8 (4.1)
*Test substance**	1.5 (2.6)	0.003 (0.003)	1.6 (2.6)
*Positive control 1**	25.6 (3.5)	0.610 (0.094)	34.7 (2.1)
*Positive control 2**	105.7 (4.6)	0.472 (0.185)	112.8 (6.7)

SD = Standard deviation; IVIS = *in vitro* irritancy score
* Corrected for background values

Remarks – Results: The IVIS of the test substance was 1.6. An IVIS ≤ 3 is considered as non-irritating to the eye according to the test guideline.

The controls gave satisfactory results confirming the validity of the test system.

CONCLUSION: The notified chemical is not considered an eye irritant requiring classification of it under the GHS criteria.

TEST FACILITY: BASF (2017b)

B.5. Eye Irritation – *In Vitro* EpiOcular™ Test

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 492 Reconstructed human Cornea-like Epithelium (RhCE) test method for identifying chemicals not requiring classification and labelling for eye irritation or serious eye damage (2015)

Vehicle: Nil

Remarks – Method: Negative control: deionised water
Positive control: methyl acetate (100%)

No significant protocol deviations.

RESULTS

Test Material	*Mean OD_{570} of Duplicate Tissues*	*Relative Mean Viability (%)*
Negative Control	1.736	100
Test Substance	0.864	49.8
Positive Control	0.298	17.2

OD = optical density

Remarks – Results: The relative viability of the test substance was 49.8%. Based on the relative viability score of ≤ 60%, a prediction could not be made under the test guideline.

The controls gave satisfactory results confirming the validity of the test system.

CONCLUSION: No prediction on eye irritation potential could be made for the test substance under the conditions of the test.

TEST FACILITY: BASF (2017b)

B.6. Skin Sensitisation – LLNA

TEST SUBSTANCE: Notified chemical

METHOD	OECD TG 429 Skin Sensitisation: Local Lymph Node Assay (2010)
Species/Strain	Mouse/CBA/CaOlaHsd
Vehicle	Methyl ethyl ketone
Preliminary study	Yes
Positive control	α-hexylcinnamaldehyde (85% technical grade) not conducted in parallel with the study.
Remarks – Method	A preliminary study (three treatment groups, each with two animals) was conducted with the test substance at 10%, 50% and 100%. No significant protocol deviations.

RESULTS

Concentration (% w/w)	*Number and Sex of Animals*	*Proliferative Response (DPM/lymph node)*	*Stimulation Index (test/control ratio)*
Test Substance			
0 (vehicle control)	5F	185.1	1
25	5F	846.8	4.57
50	5F	2,040.5	11.02
100	5F	2,486.9	15.61
Positive Control			
1	Not stated	Not stated	2.91
5	Not stated	Not stated	4.29
15	Not stated	Not stated	9.61

EC3	36.5%
Remarks – Results	No mortalities, signs of systemic toxicity or local effects were observed during the study. The test substance concentrations did not cause increases in ear weights demonstrating the absence of skin irritation. The positive control performed as expected confirming the validity of the study.
CONCLUSION	There was evidence of induction of a lymphocyte proliferative response indicative of skin sensitisation to the notified chemical.
TEST FACILITY	BASF (2018a)

B.7. Skin Sensitisation – *In Chemico* DPRA Test

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 442c *In Chemico* Skin Sensitisation: Direct Peptide Reactivity Assay (DPRA) (2015)
Vehicle	Acetonitrile
Remarks – Method	Vehicle control: acetonitrile Positive control: ethylene glycol dimethacrylate No significant deviations protocol deviations.

RESULTS

Sample	*Cysteine Peptide Depletion (%)*	*Lysine Peptide Depletion (% ± SD)*
Vehicle Control	0.0 (1.75)	0.0 (0.66)
Test Substance	2.68 (1.78)	-4.21 (1.18)
Positive Control	61.06 (1.98)	14.70 (1.68)

SD = Standard Deviation

Remarks – Results	The samples of the test substance with the lysine-peptide were emulsions at the time of preparation and after 24 hours. Due to the insolubility of the

test substance in the lysine-peptide samples calculation of mean peptide depletion is not applicable and the cysteine 1:10 prediction model is used for evaluation.

Based on the cysteine 1:10 prediction model and a cysteine peptide depletion of 2.68%, which is ≤ 13.89%, the test substance shows minimal or no reactivity with the peptides (negative prediction for skin sensitisation).

The positive and vehicle controls gave satisfactory results, confirming the validity of the test.

CONCLUSION: The notified chemical was considered to have minimal or no reactivity for peptide depletion under the conditions of the test, showing negative results in the first key event (molecular initiating) of the adverse outcome pathway (AOP) for skin sensitisation as defined in the test guideline.

TEST FACILITY: BASF (2017c)

B.8. Skin Sensitisation – *In Vitro* ARE-Nrf2 Luciferase Test

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 442d *In Vitro* Skin Sensitisation Assays Addressing the AOP Key Event on Keratinocyte Activation (2015)
- The ARE-Nrf2 luciferase LuSens test method (Appendix IB).

Vehicle: Dimethyl sulfoxide (DMSO)

Remarks – Method: Negative control: lactic acid in culture medium (450 µg/mL in 1% DMSO in culture medium)
Vehicle control: 1% DMSO in culture medium
Positive control: ethylene glycol dimethacrylate (18 µg/mL in 1% DMSO in culture medium)

No significant protocol deviations.

RESULTS

Test 1

Sample	*Concentration (µg/mL)*	*Mean Cell viability (%)*	*Mean Luciferase Induction (%)*
Vehicle Control	-	100	1
Negative Control	450	108	0.91
Test substance	76	98	1.44
	92	84	1.36
	110	90	1.48
	132	83	2.05
	158	76	1.52
	190	84	1.71
	228	79	1.55
	273	57	1.77
Positive Control	18	75	5.38

SD = Standard Deviation

Test 2

Sample	*Concentration (µg/mL)*	*Mean Cell viability (%)*	*Mean Luciferase Induction (%)*
Vehicle Control	-	100	1
Negative Control	450	105	0.97
Test substance	76	103	1.29
	91	87	1.43

Sample	*Concentration (µg/mL)*	*Mean Cell viability (%)*	*Mean Luciferase Induction (%)*
	110	83	1.45
	132	93	1.49
	158	85	1.55
	190	84	1.77
	228	86	1.7
	273	76	1.62
Positive Control	18	82	7.04

Remarks – Results

The viability of cells in the two tests were above 50%, therefore IC30 and IC50 values were not calculated.

Test 1
luciferase I_{MAX} = 2.05, EC1.5 = 137 µg/mL

Test 2
luciferase I_{MAX} = 1.77, EC1.5 = 135 µg/mL

The positive and negative controls gave satisfactory results, confirming the validity of the test.

CONCLUSION

The notified chemical was positive in the second key event (keratinocytes response) of the adverse outcome pathway (AOP) for skin sensitisation as defined in the test guideline.

TEST FACILITY

BASF (2017c)

B.9. Skin Sensitisation – *In Vitro* Human Cell Line Activation Test (h-CLAT)

TEST SUBSTANCE

Notified chemical

METHOD

OECD TG 442e *In Vitro* Skin Sensitisation Assays Addressing the Key Event on Activation of Dendritic Cells on the Adverse Outcome Pathway for Skin Sensitisation *In Vitro* Skin Sensitisation (2016)
- Human Cell Line Activation Test (h-CLAT)

Vehicle

DMSO

Remarks – Method

Negative control: lactic acid (1,000 µg/mL in culture medium)
Positive control: 1- chloro-2,4-dinitrobenzene (DNCB) (4.0 µg/mL in 0.2% DMSO in culture medium)
Vehicle control: 0.2% DMSO in culture medium

A test substance is predicted to activate monocytic THP-1 cells when CD86 expression is increased ≥ 150% and/or CD54 expression increased ≥ 200% at any concentration in relation to vehicle control that do not reduce viability below 50% and reproduced in the same cell surface marker in at least two independent experiments.

No significant deviations from the OECD test guideline

RESULTS

Test 1

Sample	*Concentration (µg/mL)*	*Mean RFI* CD86 (%)*	*Mean RFI* CD54 (%)*	*Relative Viability (%)*
Vehicle Control	-	100	100	100
Negative Control	1,000	94	158	99
Test substance	285	80	164	98
	342	63	177	95
	410	54	275	86

Sample	*Concentration (µg/mL)*	*Mean RFI* CD86 (%)*	*Mean RFI* CD54 (%)*	*Relative Viability (%)*
	492	62	310	88
	591	63	282	87
	709	58	295	82
	851	47	381	83
	1,021	30	446	29
Positive Control	4	228	315	74

* RFI = relative fluorescence intensity

Test 2

Sample	*Concentration (µg/mL)*	*Mean RFI* CD86 (%)*	*Mean RFI* CD54 (%)*	*Relative Viability (%)*
Vehicle Control	-	100	100	100
Negative Control	1,000	98	128	97
Test substance	285	84	383	98
	342	75	610	91
	410	76	680	91
	492	68	732	91
	591	76	522	94
	709	66	565	91
	851	68	537	87
	1,021	66	388	94
Positive Control	4	247	723	72

* RFI = relative fluorescence intensity

Remarks – Results

As the CD86 expression was < 150% in both tests, the EC150% (the concentration resulting in a RFI of 150%) for CD86 was not calculated.

The EC200% (the concentration resulting in a RFI of 200%) for CD54 was calculated to be 358 µg/mL (test 1). Calculation of an EC200% for test 2 was not applicable as inductions above 200% fold were obtained in all tested concentrations.

CONCLUSION

The test substance was positive in the third key event (dendritic cell activation) of the adverse outcome pathway (AOP) for skin sensitisation as defined in the test guideline.

TEST FACILITY

BASF (2017c)

B.10. Repeat Dose Oral Toxicity – Rat

TEST SUBSTANCE

Analogue chemical

METHOD

OECD TG 407 Repeated Dose 28-day Oral Toxicity Study in Rodents (2008)

Species/Strain: Rat/Wistar Crl:WI(Han)

Route of Administration: Oral – gavage

Exposure Information: Total exposure days: 28 days
Dose regimen: 7 days per week
Post-exposure observation period: 14 days

Vehicle: Carboxymethylcellulose (1% aqueous solution)

Remarks – Method: No significant protocol deviations.

RESULTS

Group	*Number and Sex of Animals*	*Dose (mg/kg bw/day)*	*Mortality*
Control	5M/5F	0	0/10
Low Dose	5M/5F	100	0/10
Mid Dose	5M/5F	300	0/10

High Dose	5M/5F	1,000	0/10
Control Recovery	5M/5F	0	0/10
High Dose Recovery	5M/5F	1,000	0/10

Mortality and Time to Death

No unscheduled mortalities were observed during the study.

Clinical Observations

Two high dose females showed ataxia immediately after dosing on days 27 and 28. The study authors stated this effect was transient but test substance related.

Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis

The following statistically significant effects were observed:

- Reduction in mean prothrombin time and mean absolute basophil level in high dose males
- Reduction in mean eosinophils level in low dose females
- Reduction in mean alanine aminotransferase level in low dose males and aspartate aminotransferase levels in low and high dose males
- Reduction in mean total bilirubin level in high dose males, butno anaemia (The study authors stated that a decrease of serum bilirubin levels was probably due to an increased conjugation of bilirubin in the liver followed by an accelerated excretion through the bile. This mechanism was regarded as adaptive because of an induction of phase II liver enzymes)
- Increase in mean total protein and mean globulin levels in high dose males. (The study authors stated that the total protein levels were marginally higher than the historical control range and were therefore not considered to be adverse).
- Reduction in mean chloride level in high dose males
- Increase in mean potassium level in high dose females
- Reduction in mean inorganic phosphate level in low and mid dose females
- Reduction in mean urine pH value in high dose males
- Higher incidences of transitional epithelial cells and granulated epithelial and casts in urine in high dose males. (The study authors stated that this finding in male rats of this age combined with the occurrence of α-2u globulins in the renal tubuli is a species-specific effect with no relevance to humans (Hard *et al*., 1993))

The study authors stated most of these changes were considered to be incidental and not treatment related.

Effects in Organs

The following statistically significant findings were observed:

- Increase (dose related) in mean absolute liver weight in high dose males (22% increase compared to control males) and increase (dose related) mean relative liver weight in mid (10% increase compared to control males), high (23.3% increase compared to control males), high dose recovery males (15% increase compared to control recovery males) and high dose females (13% increase compared to control females). As no correlating histopathological findings were observed, the study authors stated these effects were not considered to be adverse.
- Increase in mean absolute adrenal glands in high dose females. The study authors stated that as the relative weight was not significantly changed and no adverse histopathological findings was observed, this change was not considered to be adverse.
- Increase in mean absolute thyroid glands weight in high dose recovery males (26% increase compare to control recovery males).

Uterus: decidual reaction in a high dose female, horn dilation in a high dose recovery female and epithelial hypertrophy in vagina in a high dose female were observed.

The study authors stated that as these findings occurred, either individually, or were biologically equally distributed over control and treatment groups, they were considered to be incidental or spontaneous in nature and without any relation to treatment.

CONCLUSION
The NOAEL was established as 1,000 mg/kg bw/day in males and 300 mg/kg bw/day in females in this study, by the study authors, based on ataxia observed in high dose females.

TEST FACILITY BASF (2013a)

B.11. Repeat Dose Dermal Toxicity – Rat

TEST SUBSTANCE Analogue chemical

METHOD OECD TG 411 Subchronic Dermal Toxicity: 90-day Study (1981)
Species/Strain Rats/Crl:WI(Han)
Route of Administration Dermal – semi-occluded
Exposure Information Total exposure days: 90 days
Dose regimen: 5 days per week
Duration of exposure: 6 hours/day
Vehicle Corn oil
Remarks – Method No significant protocol deviations.

RESULTS

Group	*Number and Sex of Animals*	*Dose (mg/kg bw/day)*	*Mortality*
Control	10M/10F	0	0/20
Low Dose	10M/10F	100	0/20
Mid Dose	10M/10F	300	0/20
High Dose	10M/10F	1,000	0/20

Mortality and Time to Death
No unscheduled mortalities were observed during the study.

Clinical Observations
Skin erosion in the dorsal region and in left flank was observed in one high dose male and one high dose female respectively. The study authors claimed these symptoms were regarded as non adverse due to the temporary occurrence.

Two high dose males, three high dose females and one mid dose male showed slight or moderate erythema during study days 4-92. Diffuse to focal erosions were observed in five high dose and three mid dose males, in four high dose females and in one control female. Scales were observed in two high dose males, five high dose females, one mid dose male and female, and one low dose female. The study authors claimed, as these symptoms occurred temporarily, these were considered to be non adverse.

Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis
Mid and high dose females showed significant reduction (12% reduction compare to control females)in blood glucose level.

Effects in Organs
No adverse findings were observed at necropsy.

Remarks – Results
Although erythema, erosions and scales were most likely related to treatment they were assessed to be non-adverse by the study authors as they occurred only temporarily for a few days in individual animals.

CONCLUSION
The NOAEL was established by the study authors as 1,000 mg/kg bw/day in this study.

TEST FACILITY BASF (2015e)

B.12. Genotoxicity – Bacteria

TEST SUBSTANCE Notified chemical

METHOD	OECD TG 471 Bacterial Reverse Mutation Test (1997) Plate incorporation procedure (test 1) and pre incubation procedure (test 2).
Species/Strain	*Salmonella typhimurium*: TA1535, TA1537, TA98 and TA100, *Escherichia coli*: WP2uvrA
Metabolic Activation System	S9 mix from phenobarbital/β-naphthoflavone induced rat liver
Concentration Range in Main Test	Test 1 a) With metabolic activation: 33, 100, 333, 1,000, 2,600 and 5,200 µg/plate b) Without metabolic activation: 33, 100, 333, 1,000, 2,600 and 5,200 µg/plate Test 2 a) With metabolic activation: 10 (TA strains only), 33, 100, 333, 1,000, 2,600, and 5,200 µg/plate (WP2uvrA strain only) b) Without metabolic activation: 10 (TA strains only), 33, 100, 333, 1,000, 2,600 and 5,200 µg/plate (WP2uvrA strain only)
Vehicle	DMSO
Remarks – Method	No information on dose range finding study. Vehicle and positive control studies were conducted in parallel with the main study. Positive control: with S9 mix: 2-aminoanthracene (TA1535, TA100, TA1537, TA98 and WP2 uvrA) without S9 mix: *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine (TA1535, TA100), 4-nitro-o-phenylenediamine (TA98), 9-aminoacridine (TA1537) and 4-nitroquinoline-*N*-oxide (WP2 uvrA) No protocol deviations.

RESULTS

Metabolic Activation	*Test Substance Concentration (µg/plate) Resulting in:*			
	Cytotoxicity in Preliminary Test	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Absent				
Test 1	Not stated	≥ 1,000	> 5,200	Negative
Test 2	Not stated	≥ 333	> 5,200	Negative
Present				
Test 1	Not stated	≥ 333	> 5,200	Negative
Test 2	Not stated	≥ 333	> 5,200	Negative

Remarks – Results	No biologically relevant increases in revertant colony numbers of any of the tester strains were observed during the test in either the presence or absence of metabolic activation. The positive controls induced a distinct increase of revertant colonies during the study indicating the validity of the test system.
CONCLUSION	The notified chemical was not mutagenic to bacteria under the conditions of the test.
TEST FACILITY	BASF (2017d)

B.13. Genotoxicity – *In Vitro* Chromosome Aberration Test in Human Lymphocytes

TEST SUBSTANCE	Analogue chemical
METHOD	OECD TG 473 *In vitro* Mammalian Chromosome Aberration Test

Species/Strain	Human
Cell Type/Cell Line	Lymphocytes
Metabolic Activation System	S9 mix from Aroclor 1254 induced rat liver
Vehicle	Eagle's minimal essential medium (EMEM)
Remarks – Method	Negative control: EMEM Positive control: without metabolic activation: ethyl methanesulfonate with metabolic activation: cyclophosphamide The short term exposure time was only 2 hours rather than 3-6 hours as stated in the guideline. A continuous exposure without metabolic activation was also not conducted.

Metabolic Activation	*Test Substance Concentration (µg/mL)*	*Exposure Period*	*Harvest Time*
Absent			
Test 1	312.5*, 625*, 1,250*, 2,500* and 5,000	2 h	24 h
Present			
Test 1	312.5*, 625*, 1,250*, 2,500* and 5,000	2 h	24 h

*Cultures selected for metaphase analysis

RESULTS

Metabolic Activation	*Test Substance Concentration (µg/mL) Resulting in:*			
	Cytotoxicity in Preliminary Test	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Absent				
Test 1	Not stated	≥ 5,000	Not sated	Negative
Present				
Test 1	Not stated	≥ 5,000	Not stated	Negative

Remarks – Results	Haemolysis and a marked reduction in the mitotic index were observed at 5,000 ug/ml. No statistically significant or biologically relevant increase in the number of cells with aberrations was observed at any concentration, with and without metabolic activation. The positive controls behaved as expected, confirming the validity of the test system.
CONCLUSION	The test substance was not clastogenic to human lymphocytes treated *in vitro* under the conditions of the test.
TEST FACILITY	Safepharm (1988)

B.14. Genotoxicity – *In Vitro* Gene Mutation Test in Chinese Hamster V79 Cells

TEST SUBSTANCE	Analogue chemical
METHOD	OECD TG 476 *In vitro* Mammalian Cell Gene Mutation Test (1997)
Species/Strain	Chinese hamster
Cell Type/Cell Line	V79
Metabolic Activation System	S9 mix from phenobarbital/β-naphthoflavone induced rat liver
Vehicle	Deionised water
Remarks – Method	Negative control: deionised water Positive control: Without S9: ethylmethane sulfonate With S9: 7,12-dimethylbenz(a)anthracene

In a preliminary test, Chinese hamster V79 cells were treated with test substance at 13.6 to 1,740 µg/mL for 4 hours with metabolic activation and 4 and 24 hours without metabolic activation.

Metabolic Activation	*Test Substance Concentration (µg/mL)*	*Exposure Period*	*Expression Time*	*Selection Time*
Absent				
Test 1	108.8*, 217.5*, 435*, 870*, 1,305*, 1,740	4 h	7 days	7 days
Test 2	108.8*, 217.5*, 435*, 870*, 1,305*, 1,740	24 h	7 days	7 days
Present				
Test 1	108.8, 217.5*, 435*, 870*, 1,305*, 1,740*	4 h	7 days	7 days
Test 2	108.8, 217.5*, 435*, 870*, 1,305*, 1,740*	4 h	7 days	7 days

*Cultures selected for metaphase analysis

RESULTS

Metabolic Activation	*Test Substance Concentration (µg/mL) Resulting in:*			
	Cytotoxicity in Preliminary Test	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Absent				
Test 1	≥ 1,704	≥ 1,305	> 1,704	Negative
Test 2	≥ 1,704	≥ 1,305	> 1,704	Negative
Present				
Test 1	> 1,704	≥ 1,704	> 1,704	Negative
Test 2	Not conducted	≥ 1,704	> 1,704	Negative

Remarks – Results

In the preliminary toxicity test up to 1,704 µg/mL, the test substance induced evidence of toxicity (0.0% and 5.6% relative cloning efficiency at 4 and 24 hour exposures, tests 1 and 2 respectively) at the highest concentration tested (without metabolic activation).

No relevant and reproducible dose dependent increase of the mutation frequency was observed up to the maximum concentration, with or without metabolic activation.

The positive controls behaved as expected, confirming the validity of the test system.

CONCLUSION

The test substance was not clastogenic to Chinese hamster V79 cells treated *in vitro* under the conditions of the test.

TEST FACILITY

Harlan (2010)

B.15. Genotoxicity – *In Vivo* Mouse Micronucleus Test

TEST SUBSTANCE

Analogue chemical

METHOD

Similar to OECD TG 474 Mammalian Erythrocyte Micronucleus Test

Species/Strain

Mouse/CD-1

Route of Administration

Oral – gavage

Vehicle

Methylcellulose (1% aqueous solution)

Remarks – Method

Negative control: methylcellulose (1% aqueous solution)
Positive control: mitomycin C

In a preliminary dose range finding test, five groups of 4 mice (2M/2F) were treated with the test substance at 216-1,000 mg/kg bw. One high dose (1,000 mg/kg bw) female was sacrificed *in extremis* 5 hours after dosing. Piloerection was observed at all dose levels in all animals. In addition the following clinical symptoms were observed at 600 mg/kg bw and 1,000 mg/kg bw: coma, cyanosis, decreased and/or increased respiratory rate, gasping, hunched posture, lethargy, and loss of righting

reflex. Therefore, a concentration range of 216-600 mg/kg bw was chosen for the main test.

Group	*Number and Sex of Animals*	*Dose (mg/kg bw)*	*Sacrifice Time (hours)*
I (vehicle control)	5M/5F	0	24
II (vehicle control)	5M/5F	0	48
III (low dose)	5M/5F	150	24
IV(mid dose)	5M/5F	300	24
V (high dose)	5M/5F	600	24
VII (vehicle control)	5M/5F	600	48
VII (positive control, M)	5M/5F	12	24

M = mitomycin C

RESULTS

Doses Producing Toxicity	No unscheduled mortalities were observed during the study. One high dose female showed slight hunched posture, lethargy, piloerection and waddling at 1 and 4 hour observations and one high dose male showed slight piloerection at the 1 hour observation. A statistically significant reduction in the ratio of polychromatic to normochromatic erythrocytes (PCE/NCE) was obtained at the 48 hour sampling time for high dose animals. However the study authors state that this decrease in not thought to be indicative of bone marrow depression as the ratios were not unusually low when compared to the vehicle control at the 24 hour sampling time.
Genotoxic Effects	No increase in the incidence of micronucleated polychromatic erythrocytes was observed in the test groups.
Remarks – Results	The vehicle and positive controls performed as expected, confirming the validity of the test system.

CONCLUSION The test substance was not clastogenic under the conditions of this *in vivo* mouse micronucleus test.

TEST FACILITY Huntingdon (1994)

B.16. Reproductive/Developmental Toxicity Screening Test (dermal) – Rat

TEST SUBSTANCE Analogue chemical

METHOD OECD TG 421 Reproduction/Developmental Toxicity Screening Test (1995)

Species/Strain	Rat/Wistar Crl:WI(Han)
Route of Administration	Dermal – semi-occluded
Exposure Information	Exposure period – female: 2-week premating, 3-week mating period and the entire gestation period as well as approximately 2 weeks of the lactation period. Exposure period – male: 2-week premating and 3-week mating period
Vehicle	Corn oil
Remarks – Method	No significant protocol deviations.

RESULTS

Group	*Number and Sex of Animals*	*Dose (mg/kg bw/day)*	*Mortality*
0	10M/10F	0	0/20
1	10M/10F	100	0/20
2	10M/10F	300	0/20
3	10M/10F	1,000	0/20

Mortality and Time to Death
No unscheduled mortalities were observed during the study.

Effects on Parental (P) animals:
Male mating index was 100% in all groups. One sperm positive high dose female and two sperm positive control females did not become pregnant. The male fertility indices were 80%, 100%, 100% and 90% in control, low, mid and high dose groups.

The gestation indices were 87.5% for control and 100% for low, mid and high dose groups.

The mean number of implantation sites was 10.4, 10.8, 12.3 and 11.6 implants/dam in control, low, mid and high dose groups, respectively. The mean number of F1 pups delivered were 9.5, 10.0, 11.7 and 10.6 pups/dam in control, low, mid and high dose groups, respectively. Therefore, there were no indications for dose related intrauterine embryo or foetal lethality.

The live birth indices were 87.5% in control group and 100% in low, mid and high dose groups.

Effects on 1st Filial Generation (F1)
One out of 76 and 1/95 pups from control and high dose groups respectively, were found dead. Six stillborn (control) and 1 stillborn (mid dose) pups were observed.

The mean number of delivered F1 pups per dam and the rates of live born, stillborn, cannibalised and dead F1 pups were similar in all groups.

The viability indices during lactation (PND 0 - 4) were 89.4%, 98.2%, 100% and 98.9% in control, low, mid and high dose groups, respectively.

Remarks – Results
No adverse skin reactions were observed during the study.

CONCLUSION
The NOAEL for systemic and reproductive/developmental toxicity was established by the study authors as 1,000 mg/kg bw/day in this study, based on the absence of treatment related adverse effects up to the highest dose tested.

TEST FACILITY	BASF (2015f)

B.17. Reproductive Toxicity, Extended One-Generation Study (diet) – Rat

TEST SUBSTANCE	Analogue chemical
METHOD	OECD TG 443 Extended One-Generation Reproductive Toxicity Study (2011)
Species/Strain	Rat/Wistar,Crl:WI(Han),
Route of Administration	Oral – diet
Exposure Information	<u>Parental (F0) animals</u> Females: 10 weeks premating, 2 weeks mating, gestation and lactation (up to postnatal day 21 or 22) Males: 10 weeks premating, 2 weeks mating and 4 weeks post mating <u>F1 Generation</u> Cohort A: post weaning (~ 62 days) Cohort B: post weaning (~ 67 days)
Remarks – Method	As dams' food intake increases during lactation, females were given only 50% of the test substance during this period. The study authors stated this dietary adjustment was based on historical body weight and food consumption data on dams. No significant protocol deviations

RESULTS

F0 generation parental animals

Group	*Number and Sex of Animals*	*Dose (mg/kg bw/day)*	*Mortality*
1	25M/25F	0	0/50
2	25M/25F	~ 90 (1,000 ppm)	0/50
3	25M/25F	~ 359 (4,000 ppm)	0/50
4	25M/25F	~ 1,113 (12,500 ppm)	0/50

F1 rearing animals, cohort 1A

Group	*Number and Sex of Animals*	*Dose (mg/kg bw/day)*	*Mortality*
5	20M/20F	0	0/40
6	20M/20F	~ 90 (1,000 ppm)	0/40
7	20M/20F	~ 359 (4,000 ppm)	0/40
8	20M/20F	~ 1,113 (12,500 ppm)	0/40

F1 rearing animals, cohort 1B

Group	*Number and Sex of Animals*	*Dose (mg/kg bw/day)*	*Mortality*
9	25M/25F	0	0/50
10	25M/25F	~ 90 (1,000 ppm)	0/50
11	25M/25F	~ 359 (4,000 ppm)	0/50
12	25M/25F	~ 1,113 (12,500 ppm)	0/50

Mortality and Time to Death

No unscheduled mortalities were observed during the study.

Effects on Parental (F0) animals:

Statistically significant reduction in food consumption was observed in high-dose males (during premating days 0 - 3) (about 14% below control), however, the average food consumption of these animals during premating (day 0-73) was quite comparable to the control.

Statistically significant reduction in food consumption was observed in high dose females during premating days 28 – 38 (up to 16% below control), during GD 0 – 6 (up to 8% below control) and during several days of the lactation period (up to 9% below control). Food consumption was more consistent during lactation resulting in an overall reduced food consumption of 7% (below control) in these females.

Food consumption of the low and mid dose F0 males and females were comparable to the control values.

Bodyweights and body weight change of all F0 males and females were essentially comparable to the control values.

The male and female mating indices were 100% in all treatment groups. Two low, one mid and three high dose males and females did not produce F1 pups. The male and female fertility indices were 100%, 92%, 96% and 88% for control, low, mid and high dose males respectively. No adverse histopathological findings were observed in the infertile animals.

The gestation index was 100% in all test groups.

The number of implantation sites were 12.7, 12.9, 13.0 and 11.5 implants/dam in control, low, mid and high dose groups, respectively, therefore implantation was not considered to be affected by the test substance. Furthermore, the post-implantation loss did not show any statistically significant differences (5.8%, 4.1%, 8.6% and 6.1% in control, low, mid and high doses, respectively), and the mean number of F1 pups delivered per dam remained unaffected (12.0, 12.4, 11.9 and 11.0 pups/dam in control, low, mid and high doses, respectively).

The live indices were 100%, 96%, 100% and 99% in control, low, mid and high dose groups, respectively.

Statistically significant reduction in live born pups and several stillborn pups were observed in the low dose group. As this was due to a single dam having seven still born pups, the study authors stated this effect was not dose related and considered to be spontaneous in nature.

The following statistically significant clinical chemistry, haematology, urinalysis effects were observed:

- Reduction in prothrombin time and urine pH values in mid and high dose males
- Reduction in relative monocyte counts in low, mid and high dose males
- Increased blood urea level in high dose males
- Increased inorganic phosphate levels in low dose males
- Increased epithelial and granular casts in urine sediments, in combination with the histopathologic finding of α-2-u-globulinuria in the kidneys of mid and high dose males. The α-2-u-globulinuria is regarded as a rat specific effect with no relevance for humans.
- Increased percentage of abnormal sperms in the cauda epididymidis of high dose males

The study authors stated that as most of these values were within the historical control range, these changes were considered be incidental and not treatment-related.

The following statistically significant effects in organs were observed:

- Increased mean absolute and relative adrenal gland and liver weights in high dose males
- Increased mean absolute and relative seminal vesicle in mid (8% increase compare to control males) and high (14% increase compare to control males) dose males
- increased mean absolute and relative adrenal gland and thyroid gland weights in high dose (14% increase compare to control females) females, (slight increase [statistically not significant] in total thyroxine and thyroid stimulating hormone was observed).
- increased mean absolute kidney and spleen weights in high dose males

Adrenal glands
Five high dose males showed slight (grade 1) diffuse cortical hyperplasia in the adrenal gland. This finding was characterised by a minimal diffuse increase in the number of cortical cells of the zona fasciculata, resulting in an increased cortical thickness.

Liver
Increased incidence of foci of cellular alteration in males (1 in low, 1 in mid and 6 in high doses) and females (2 in control, 7 in low, 4 in mid and 17 in high dose) was observed. Most of the foci were single and small (up to 8 -15 cells) and of basophilic tigroid type. In addition, two males and two females of the high dose group showed additional single eosinophilic foci.

The adrenal glands and liver findings were considered by the study authors as incidental and not dose related.

Effects on 1st Filial Generation (F1)
Statistically significant reduction in bodyweight in high dose female pups at PND 21 (7% below control) and in both sexes at PND 14 – 21 (9% below control for both sexes) was observed. In addition, after weaning the body weight gain in the high dose adolescents continued to be lower compared to the control (7-10%). The study authors claimed this effect was due to the direct exposure of the offspring to the test substance through the diet rather indication of developmental toxicity.

The pup viability indices during early lactation (PND 0 - 4) were 99%, 99%, 99% and 98% in control, low, mid and high doses, respectively. The lactation indices on PND 4 - 21 were 100%, 99%, 100% and 100% in control, low, mid and high doses, respectively.

The following statistically significant changes were observed:

- reduction in glucose level in low (13.4% reduction compare to control males), mid (13.8% reduction compare to control males) and high (16.5% reduction compare to control males) dose males
- reduction in total bilirubin in mid and high dose males
- increase in blood urea level in mid and high dose males
- reduction in urine pH values and increased incidence of transitional epithelial cells and granular and epithelial casts in the urine sediments in high dose males. Histopathology showed α-2-u-globulinuria (rat specific effect)

Sexual maturity
High dose female pups showed statistically significant increase in the mean number of days required to reach vaginal opening and this was 31.5, 31.7, 31.9 and 32.7 for control, low, mid and high dose pups, respectively.

The study authors stated that the later onset of puberty in high dose pups is most likely a consequence of a general developmental delay and not a specific effect on the timing of puberty.

First preputial separation in male F1 pup was observed on PND 38 and the last was on PND 52. High dose male group showed statistically significant increase in preputial separation and the mean number of days to reach the criterion was 41.6, 42.1, 42.4 and 43.2 for control, low, mid and high dose groups, respectively. The study authors stated the increase in high dose males was within the historical control value.

F1 rearing animals Cohort A
Statistically significant reduction in body weights of the high dose males towards the end of the study (days 49 – 56, about 7% below control) was observed. The body weights of the high dose females were below control values (final weight about 5%), but were not statistically significant. A statistically significant overall reduction in body weight gain was also observed between day 0 (post weaning) and 56 in high dose males (10% below control) and females (8% below control). The study authors claimed this effect was due to the direct exposure of the offspring to the test substance through the diet rather than representing developmental toxicity.

Increased mean absolute and relative adrenal gland weights was observed in high (16% increase compare to control males) dose males and females (13% increase compare to control females), and histopathology showed minimal diffuse cortical hyperplasia in the adrenal glands in 2 high dose males. The study authors stated this finding was considered to be incidental and not dose related.

F1 rearing animals Cohort B
Statistically significant reduction in the body weights of the high dose males on days 28 – 56 (7% below control) and high-dose females on days 42 – 49 and on day 63 (6% below control) was observed. A statistically significant overall reduction in body weight gain (7% below control) was also observed between day 0 (post weaning) and 63 in high dose males and females. The study authors claimed this effect was due to the direct exposure of the offspring to the test substance through the diet rather than representing developmental toxicity.

Statistically significant increase in absolute and relative mean adrenal glands in high dose (24% increase compare to control males) males and high (20% increase compare to control females) dose females was observed.

Remarks – Results
Lower pup body weights in the high dose pups shortly before weaning as well as a delay of puberty in females were considered to be caused by systemic toxicity of the test substance after direct exposure through the diet and do not represent developmental toxicity.

CONCLUSION
The NOAEL for systemic toxicity was established by the study authors as 359 mg/kg bw/day based on decreased food consumption in F0 parental animals and decreased bodyweight/bodyweight gain in adolescent and adult F1 offspring at 1,113 mg/kg bw/day.

The NOAEL for reproductive and developmental toxicity was established by the study authors as 1,113 mg/kg bw/day in this study.

TEST FACILITY BASF (2018b)

B.18. Prenatal Developmental Toxicity (dermal) – Rat

TEST SUBSTANCE	Analogue chemical
METHOD	OECDE TG 414 Proposal for Updating Guideline, Prenatal Developmental Toxicity Study (2001)
Species/Strain	Rats/Wistar (Crl:WI[Han])
Route of Administration	Dermal – semi-occluded
Exposure Information	Exposure days: 14 days (gestation days (GD) 6-19) Duration of exposure: 6 hours/day
Vehicle	Corn oil
Remarks – Method	In a prenatal developmental toxicity study, the test substance was applied to pregnant rats from implantation to one day prior to the expected day of

parturition (GD 6-19) for the evaluation of its potential maternal and prenatal developmental toxicity.

RESULTS

Group	*Number of Animals*	*Dose (mg/kg bw/day)*	*Mortality*
1	25F	0	0/25
2	25F	100	0/25
3	25F	300	0/25
4	25F	1,000	0/25

Mortality and Time to Death

No unscheduled mortalities were observed during the study.

Effects on Dams

Vaginal haemorrhage was observed in one low, three mid and one high dose females on GD 15-17 either before the daily dermal application or after the daily 6-hour exposure time. The study authors claimed that this finding may due to the handling of animals and semi-occlusive dressing rather test substance related.

The conception rate was 96% in control and high dose groups and 100% in low and mid dose groups.

Reproductive parameters, such as conception rate, mean number of corpora lutea, mean number of implantations, as well as pre- and post-implantation loss, were similar among all groups.

Effects on Foetus

One high dose male foetus (out of 236 foetuses) showed limb hyperextension. The study authors stated this single finding was considered to be spontaneous in nature.

One skeletal malformation affecting the forelimb was observed in high dose group. The study authors stated the single finding can be found in the historical data in a comparable frequency.

Skeletal variations of different bone structures (super numeracy thoracic vertebrata and wavy rib) were observed in all groups, with or without effects on corresponding cartilages. The study authors stated the effect was comparable to the historical control data.

Statistically significant increase in supernumerary thoracic vertebra in mid dose foetus and statistically significant increase in wavy rib in high dose foetus were observed. These findings were marginally above the historical control data.

Isolated cartilage findings related to skull, sternum and ribs without impact on the respective bony structures were observed in control (90.7%), low (87.1%), mid (84.6%) and high (92.9%) dose foetus.

Foetal weight and sex distribution of the foetuses were similar among the groups.

CONCLUSION

The NOAEL for maternal and prenatal developmental toxicity was established by the study authors as 1,000 mg/kg bw/day in this study..

TEST FACILITY BASF (2015g)

B.19. *In vitro* Dermal Penetration Study

TEST SUBSTANCE Analogue chemical

METHOD OECD TG 428, Skin Absorption: *in vitro* Method

Remarks – Method Diffusion of ^{14}C-pyranol into and through rat skin was assessed by a single topical application of 8,500 µg/cm² (neat) and 902 µg/cm² (as a solution

in corn oil) test substance to split thickness rat skin preparations mounted on Franz-type diffusion cells.

RESULTS

Group	*Dose (µg/cm²)*	*Non-absorbed dose*	*Absorbed dose*	*Dose onskin*	*Total recovery*	*Dermal absorption**
		[% of applied dose]				
1	8,500	83.54	14.8	3.12	101.47	**17.92**
2	902	55.48	36.27	7.51	99.25	**43.78**

*absorbed dose + dose onskin

Remarks – Results

Under these test conditions, 14.8% and 36.27% of the applied dose of ^{14}C-pyranol were recovered as absorbed dose for groups 1 and 2, respectively.

The estimated dermal absorption was 17.92% and 43.78% of the applied dose for groups 1 and 2, respectively.

Absorption rates were 127.08 and 37.99 [µg/(cm²*h)] and related permeability constants were 13.38 and 37.10 [*10^{-5} cm/h] for groups 1 and 2, respectively.

Mean absorption lag times were 1.27 hours and 0.42 hours for groups 1 and 2, respectively, and demonstrate the presence of a functional barrier in the skin samples used.

CONCLUSION

There was evidence that the test substance was absorbed through rat skin under the conditions of the test.

TEST FACILITY

BASF (2013b)

APPENDIX C: ENVIRONMENTAL FATE AND ECOTOXICOLOGICAL INVESTIGATIONS

C.1. Environmental Fate

C.1.1. Ready Biodegradability

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 301 B Ready Biodegradability: CO2 Evolution Test
Inoculum	Activated sludge
Exposure Period	28 days
Auxiliary Solvent	None
Analytical Monitoring	TOC
Remarks – Method	Aniline was used as a reference substance. A toxicity control was also conducted.

RESULTS

Test Substance		*Aniline*		*Toxicity control*	
Day	*% Degradation*	*Day*	*% Degradation*	*Day*	*% Degradation*
4	7	4	27	4	4
7	14	7	53	7	30
11	24	11	66	11	42
14	38	14	75	14	54
18	61	18	86	18	68
28	85	28	102	28	80

Remarks – Results	All validity criteria were met. The inorganic carbon content in the test solutions at the start of the test was < 1 mg/L, the difference in extremes at the end of the test was 5%, the CO_2 evolution of the inoculum blank was 24 mg CO_2/L The toxicity control reached the pass level by day 7 and is therefore not considered inhibitory to the inoculum. Degree of biodegradation at the end of the ten-day window (day 5 – 15) was approximately 47%.
CONCLUSION	The test substance is not readily biodegradable.
TEST FACILITY	BASF (2015h)

C.1.2. Inherent Biodegradability

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 302 C Inherent Biodegradability: Modified MITI Test (II)
Inoculum	Activated sludge
Exposure Period	28 Days
Auxiliary Solvent	None
Analytical Monitoring	BOD and HPLC
Remarks – Method	Sodium benzoate was used as a reference substance. Toxicity and abiotic controls were also conducted in parallel.

RESULTS

Test Substance		*Sodium benzoate*		*Toxicity control*	
Day	*% Degradation*	*Day*	*% Degradation*	*Day*	*% Degradation*
2	0*	2	14	2	9
4	0*	4	69	4	16
14	3	14	85	14	58
21	9	21	88	21	64
28	9	28	94	28	64

*negative values were corrected to 0.

Remarks – Results	The reference substance reached the day 7 and 14 pass levels, therefore the test is considered to be valid. HPLC analysis showed that there was 0% residual amount of the test substance after 28 days, while the residual amount in the abiotic control was 90%. This implies that the test substance is degraded into intermediate products which do not produce CO_2 when formed.
CONCLUSION	The test substance is not ultimately biodegradable but is degraded into simpler organic compounds.
TEST FACILITY	BSAL (2019b)

C.2. Ecotoxicological Investigations

C.2.1. Acute Toxicity to Fish

TEST SUBSTANCE	Notified chemical
METHOD	Equivalent to OECD TG 203 Fish, Acute Toxicity Test – semi static
Species	*Gobiocypris rarus* (Rare minnow)
Exposure Period	96 hours
Auxiliary Solvent	None
Water Hardness	142 mg $CaCO_3$/L
Analytical Monitoring	GC
Remarks – Method	The species of fish used is not in the recommended fish species list in the guideline for this study. No major protocol deviations. Based on a range finding study, test concentrations (detailed below) were prepared from dilution of a stock solution. Test solutions were renewed after 48 hours. A reference test was conducted, less than one month prior to the definitive study using potassium dichromate.

RESULTS

Concentration (mg/L)		*Number of Fish*	*Mortality*				
Nominal	*Actual*		*3 h*	*24 h*	*48 h*	*72 h*	*96 h*
Control	-	7	0	0	0	0	0
42	38.1	7	0	0	0	0	0
66	50.4	7	0	0	0	0	0
71	67.1	7	0	0	0	0	1
92	91.0	7	0	7	7	7	7
120	115	7	0	7	7	7	7

LC50	74.9 mg/L at 96 hours
NOEC	38.1 mg/L at 96 hours
Remarks – Results	All validity criteria were met. The dissolved oxygen content was maintained at 61.3 – 98.9% of the air saturation value and the

concentration of the test substance was analysed. LC50 values were calculated using the Spearman-Karber method based on the measured test concentrations.

The results from the reference study showed an LC50 of 257 mg/L, which is consistent with previous results.

CONCLUSION The test substance is harmful to fish.

TEST FACILITY BSAL (2019c)

C.2.2. Acute Toxicity to Aquatic Invertebrates

TEST SUBSTANCE Notified Chemical

METHOD OECD TG 202 Daphnia sp. Acute Immobilisation Test and Reproduction Test – Static

Species	*Daphnia magna*
Exposure Period	48 hours
Auxiliary Solvent	None
Water Hardness	163 mg $CaCO_3$/L
Analytical Monitoring	LC-MS/MS
Remarks – Method	Based on a range finding study, test concentrations (detailed below) were prepared from dilution of a stock solution. A reference test was conducted, less than one month prior to the definitive study using potassium dichromate.

RESULTS

Concentration (mg/L)		*Number of D. magna*	*Number Immobilised*	
Nominal	*Actual (Geometric mean)*		*24 h*	*48 h*
Control	-	20	0	0
3.13	0.715	20	0	0
6.25	2.01	20	0	0
12.5	3.15	20	0	1
25	6.87	20	0	0
50	23.8	20	9	12
100	59.3	20	15	20

EC50	22.3 mg/L at 48 hours
EC10	15.4 mg/L at 48 hours
Remarks – Results	There was a significant drop in the concentration of the test substance over the duration of the test. Separate stability studies were conducted to investigate the drop in concentration which determined that the test substance is stable over 48 hours. No effects were observed which could explain the decrease in test concentration. Therefore, a geometric mean was calculated from the measured values at the beginning and end of the definitive study. The EC50 and EC10 values were calculated by sigmoidal dose-response regression. All other validity criteria were met. Dissolved oxygen was maintained at ≥ 7.09 mg/L, pH was maintained between 7.67 and 7.98 and temperature was maintained at 20.3 °C. The reference test showed potassium dichromate had a 24h EC50 of 2.0 mg/L, which is within the expected range (0.6 – 2.1 mg/L).

CONCLUSION The test substance is harmful to aquatic invertebrates.

TEST FACILITY	Noack (2015a)

C.2.3. Algal Growth Inhibition Test

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 201 Alga, Growth Inhibition Test
Species	*Pseudochneriella subcaptitata*
Exposure Period	72 hours
Concentration Range	Nominal: 7.25 - 120 mg/L Actual: 8.3 - 100 mg/L
Auxiliary Solvent	None
Analytical Monitoring	LC-MS/MS
Remarks – Method	Based on a range finding study, test concentrations were prepared from dilution of a stock solution. A reference test was also conducted, less than one month prior to the definitive study using potassium dichromate.

RESULTS

Growth rate	
ErC50 (mg/L)	*NOEC (mg/L)*
74.6	17.1

Remarks – Results	All validity criteria were met. The control cell density increased by a factor of 256, the mean coefficient of variation for section-by-section specific growth was 20.2% and the coefficient of variation for the average specific growth rates was 2.73%. The reference study indicated an ErC50 for potassium dichromate of 0.61, which is consistent with previous results.
CONCLUSION	The test substance is harmful to algal growth.
TEST FACILITY	Noack (2015b)

C.2.4. Inhibition of Microbial Activity

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 209 Activated Sludge, Respiration Inhibition Test
Inoculum	
Exposure Period	3 hours
Concentration Range	Nominal: 6.25 - 1000 mg/L
Remarks – Method	Test concentrations were prepared by direct addition of the test substance into test vessels. 3,5-Dichlorophenol was used as a reference substance.
RESULTS	
IC50	> 1,000 mg/L
IC10	580 mg/L
Remarks – Results	The reference test showed 3,5 dichlorophenol IC50 of 7.0 mg/L, which is within the expected range of 2 – 25 mg/L. All validity criteria were met. The oxygen uptake of the controls was 32 mg/ g×h and the coefficient of variation between replicates was 4.2%.

CONCLUSION	The test substance is not harmful to bacterial respiration.
TEST FACILITY	BASF (2015i)

BIBLIOGRAPHY

ACI (2010) Consumer Product Ingredient Safety, Exposure and Risk Screening Methods for Consumer Product Ingredients, 2nd Edition, American Cleaning Institute, Washington DC.

BASF (2013a) Pyranol Repeated Dose 28-day Toxicity Study in Wistar Rats Including a Recovery Period of 14 Days Administration by Gavage (Project No. 30C0639/09C109, May, 2013). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier).

BASF (2013b) ^{14}C-Pyranol Study of Penetration through Rat Skin *In Vitro* (Project No: 10B0466/12B019, July, 2013). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier).

BASF (2015a) Water Solubility of "pyranyl acetate pure" (Study No. 15L00047, April, 2015). Ludwigshafen, Germany, Competence Center Analytics, BASF SE (Unpublished report submitted by the notifier).

BASF (2015b) Hydrolysis as a Function of pH of "pyranyl acetate pure" (Study No. 15L00024, June, 2015). Ludwigshafen, Germany, Competence Center Analytics, BASF SE (Unpublished report submitted by the notifier).

BASF (2015c) Partition Coefficient n-Octanol/Water (Pow) of "pyranyl acetate pure" (Study No. 15L00048, April, 2015). Ludwigshafen, Germany, Competence Center Analytics, BASF SE (Unpublished report submitted by the notifier).

BASF (2015d) Test Report (Flash Point and Autoignition Temperature (Study No: 15/0146, February, 2015). Ludwigshafen, Germany, BASF SE (Unpublished study submitted by the notifier)

BASF (2015e) Pyranol Repeated-Dose 90-Day Dermal Toxicity Study in Wistar Rat (Project No. 53C0639/09S134, February, 2015). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier)

BASF (2015f) Pyranol Reproduction/Developmental Toxicity Screening Test in Wistar Rats Dermal Administration (Project No.: 82R0639/09R137, March, 2015). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier).

BASF (2015g) Pyranol Prenatal Developmental Toxicity Study in Wistar Rats Dermal Application (Project No.: 33R0639/09R142, April, 2015). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier).

BASF (2015h) Pyranyl Acetate Pure Determination of the Ready Biodegradability in the CO2-Evolution Test (Study No. 22G0182/15G278, September, 2015). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished report submitted by the notifier).

BASF (2015i) Pyranyl Acetate Pure Determination of the Inhibition of Oxygen Consumption in the Activated Sludge Respiration Inhibition Test (Study No. 08G0182/15G279, August, 2015). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished report submitted by the notifier).

BASF (2017a) Pyranyl Acetate Pure *In vitro* Skin Irritation and Corrosion Turnkey Testing Strategy (Project No: 69V0182/15A243, April, 2017). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier).

BASF (2017b) Pyranyl Acetate Pure *In Vitro* Eye Irritation Turnkey Testing Strategy (Project No.: 71V0182/15A244, April, 2017). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier).

BASF (2017c) Pyranyl Acetate Pure *In Vitro* Skin Sensitization Turnkey Testing Strategy (Project No.: 67V0182/15A242, November, 2017). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier).

BASF (2017d) Pyranyl Acetate Pure Salmonella typhimurium / Escherichia coli Reverse Mutation Assay (Project No.: 40M0182/15M184, June, 2017). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier).

BASF (2018a) Pyranyl Acetate Pure Murine Local Lymph Node Assay (LLNA) (Project No.: 58V0182/15A255, March, 2018). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier).

BASF (2018b) Pyranol Modified Extended One-generation Reproduction Toxicity Study in Wistar Rats Administration via the Diet (Project No: 03R0639/09R138, July, 2018). Ludwigshafen, Germany, Experimental Toxicology and Ecology, BASF SE (Unpublished study submitted by the notifier).

Bioassay (2017a) Pyranyl Acetate Pure Acute Oral Toxicity Study in Rats (Project No.: 16-BF-OT125, April, 2017). Heidelberg, Germany, Labor für biologische Analytik GmbH, Bioassay (Unpublished study submitted by the notifier).

Bioassay (2017b) Pyranyl Acetate Pure Acute Dermal Toxicity Study in Rats (Project No.: 16-BF-DT126, April, 2017). Heidelberg, Germany, Labor für biologische Analytik GmbH, Bioassay (Unpublished study submitted by the notifier).

BSAL (2019a) Adsorption/Desorption of Pyranyl Acetate (Study No. R1802222105A, October, 2019). Shanghai, China, Bioassay and Safety Assessment Lab Shanghai Academy of Public Measurement (Unpublished report submitted by the notifier).

BSAL (2019b) Inherent Biodegradability by the Modified MITI test (II) of Pyranyl Acetate (Study No. R1802222105C, May, 2019). Shanghai, China, Bioassay and Safety Assessment Lab Shanghai Academy of Public Measurement (Unpublished report submitted by the notifier).

BSAL (2019c) Acute toxicity of Pyranyl Acetate to Chinese Rare Minnow (Gobiocypris rarus) (Study No. R1802222105B, May, 2019). Shanghai, China, Bioassay and Safety Assessment Lab Shanghai Academy of Public Measurement (Unpublished report submitted by the notifier).

Cadby, PA, Troy, WR, Vey, MG (2002) Consumer Exposure to Fragrance: Providing Estimates for Safety Evaluation, Regulatory Toxicology and Pharmacology, **36**:246-252.

Earnest, CW Jr. (2009) A Two-Zone Model to Predict Inhalation Exposure to Toxic Chemicals in Cleaning Products, MSCEng thesis, The University of Texas at Austin.

enHealth (2012) Australian Exposure Factor Guide, companion document to: Environmental Health Risk Assessment: Guidelines For Assessing Human Health Risks from Environmental Hazards, EnHealth, Commonwealth of Australia.

Hard, GC, Rodgers, IS, Baetcke, KP, Richards, WL, McGaughy, RE, Valcovic, LR (1993) Hazard Evaluation of Chemicals That Cause Accumulation of α2u-Globulin Hyaline Droplet Nephropathy, and Tubule Neoplasia in the Kidneys of Male Rats. Environmental Health Perspectives, **99**:313 – 349.

Harlan (2010) Gene Mutation Assay in Chinese Hamster V79 Cells *In Vitro* (V79 / HPRT) with Pyranol (Study No: 1299000, May, 2010). Rossdorf, Germany, Harlan Cytotest Cell Research GmbH (Unpublished study submitted by the notifier).

Huntingdon (1994) Florosa Mouse Micronucleus Test (Study No: KC940415, December, 1994). Huntingdon, Cambridgeshire, UK, Huntingdon Research Centre Ltd (Unpublished study submitted by the notifier)

Loretz, L, Api, AM, Barraj, L, Burdick, J, Davis, DA, Dressler, W, Gilberti, E, Jarrett, G, Mann, S, Pan, YHL, Re, T, Renskers, K, Scrafford, C, Vater, S (2006) Exposure Data for Personal Care Products : Hairspray, spray perfume, liquid foundation, shampoo, body wash, and solid antiperspirant, Food and Chemical Toxicology, **44:**2008-2018.

Noack (2015a) Pyranyl Acetate Pure Acute Immobilisation Test to Daphnia magna Semi-static, 48 h, Limit-Test (Study No. 140926BZ/ DAI16715, October, 2015). Sarstedt, Germany, DR. U. Noack-Laboratorien (Unpublished report submitted by the notifier).

Noack (2015b) Pyranyl Acetate Pure Acute Alga, Growth Inhibition Test with Pseudokirchneriella subcapitata, 72 hours (Study No. 140926BZ/ SPO16715, October, 2015). Sarstedt, Germany, DR. U. Noack-Laboratorien (Unpublished report submitted by the notifier).

OECD (2016) Guidance Document on the Reporting of Defined Approaches and Individual Information Sources to be used within Integrated Approaches to Testing and Assessment (IATA) for Skin Sensitisation, Series on Testing and Assessment No. 256, Organisation for Economic Co-Operation and Development (OECD).

Rothe, H, Fautz, R, Gerber, E, Neumann, L, Rettinger, K, Schuh, W, Gronewold, C (2011) Special Aspects of Cosmetic Spray Evaluations: Principles on Inhalation Risk Assessment, Toxicology Letters, **205**:97-104.

Safepharm (1988) ST 01 C 88: OECD 473 Metaphase Analysis in Human Lymphocytes *In Vitro* (Project No: 161/1, May, 1988). Derby, UK, Safepharm Laboratories Limited (Unpublished study submitted by the notifier).

SCCS (2012) The SCCS's Notes of Guidance for the Testing of Cosmetic Substances and their Safety Evaluation (8th revision) European Commission – Scientific Committee on Consumer Safety.

Steiling, W, Bascompta, M, Carthew, P, Catalano, G, Corea, N, D'Haese, A, Jackson, P, Kromidas, L, Meurice, P, Rothe, H, Singal, M (2014) Principle Considerations for the Risk Assessment of Sprayed Consumer Products, Toxicology Letters, **227**:41-49.

SWA (2012) Code of Practice: Managing Risks of Hazardous Chemicals in the Workplace, Safe Work Australia, https://www.safeworkaustralia.gov.au/doc/model-code-practice-managing-risks-hazardous-chemicals-workplace.

United Nations (2009) Globally Harmonised System of Classification and Labelling of Chemicals (GHS), 3rd revised edition. United Nations Economic Commission for Europe (UN/ECE), http://www.unece.org/trans/danger/publi/ghs/ghs_rev03/03files_e.html.