Soil risk assessment of the fragrance ingredient, Helvetolide ®

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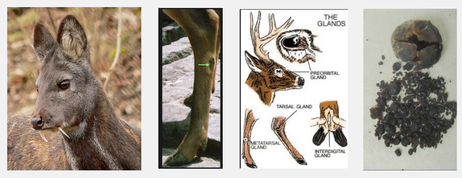
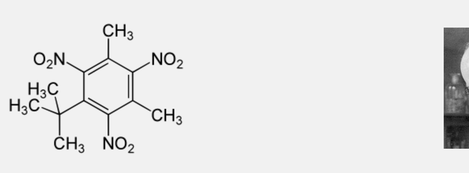
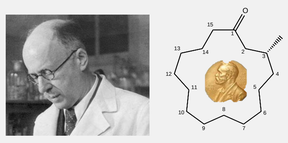
Introduction from banned natural musks and synthetic nitromusks to biodegradable synthetic alicyclic and macrocyclic musks



For millenia **musk deer w**as hunted in Eurasia for the apocrine glands. The granules in a weak tincture exude a warm, sweet, earthy and woody smell. Since 1979 the CITES convention protects deers from hunting.

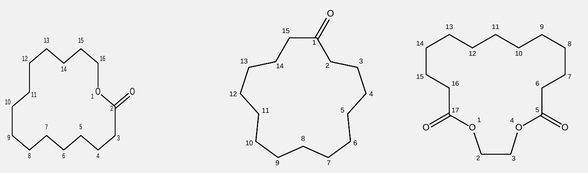


1888- **Nitromusks** were discovered by accident trying to find safe explosives such as TNT … with strong musk-smelling substances being produced, and exploited until their ban in 1981 due to poor biodegradability and bioaccumulation in organisms

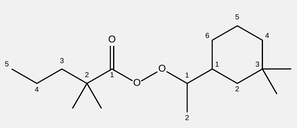


1920's- The chemical structure of **muscone**, responsible for the odour in musk was discovered by Firmenich chemist Prof. L. Ruzicka (Nobel prize chemistry in 1939).

1927- macrocyclic lactone found in plants, and later synthesized in the laboratory, and brought to market as **exaltolide**.



Macrocyclic alternatives are mostly readily biodegradable and not bioaccumulative despite their relatively high log Kow values. However, some of the earlier developed alicyclic musks are only partially biodegradable according to screening biodegradation tests.



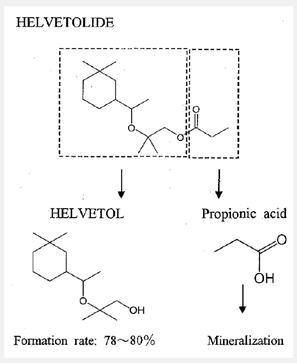
Helvetolide ®

1990- 4th generation of musks, first alicyclic musk produced on a commercial scale by Firmenich (fruity pear musky smell)

Helvetolide® Environmental profile

Persistency

- Not readily biodegradable (17% OECD 301D, 21% 301B)
- Not Persistent in OECD 301C: helvetolide C6 almost completely degraded into:
  - One identified metabolite (helvetol) and
  - propionic acid, which mineralises



- Helvetol, the principal degradation product is **not readily biodegradable** (OECD 301F) "potentially P" according to PBT criteria

Bioaccumulation

- Potential for bioaccumulation of parent and metabolite : log P measure of 4.68 (Helvetolide) and 4.33 (Helvetol)
- BCF experimental on **helvetol** C6 (OECD 305, GLP Japan 2015)

Exposure level (mg/L)	Measure of main (minor) isomer	BCF experimental
High 0.07	0.0677 mg/L (0.00233)	10-22
Low 0.007	0.00677 mg/L (0.000233)	<DL

- Helvetol main product of Helvetolide® is confirmed to be **not bioaccumulative** according to PBT criteria

Aquatic toxicity

	Experimental results		Acute EC50/LC50	Chronic NOEC/EC10
Helvetolide®	Algae	*S. capricornutum*	>1.1 mg/L	≥ 1.1 mg/L
	Invertebrates	*D. magna*	3.3 mg/L	0.20 mg/L
	Fish	*O. mykiss*	3.6 mg/L	No data

	ECOSAR & iSafeRat® predictions (experimental tests on-going)	Acute EC50/LC50	Chronic NOEC/EC10
Helvetol	Algae	1.73-2.0 mg/L	0.74-0.73 mg/L
	Invertebrates	0.93-2.4 mg/L	0.17-0.20 mg/L
	Fish	1.31-1.9 mg/L	0.17-0.34 mg/L

- Helvetolide® and helvetol are **not toxic** according to PBT criteria

Soil Hazard category (ECHA R.7c, 2014)

Helvetolide (the registered substance):

- Not highly adsorptive (log Koc = 4.3 <5),
- Not highly persistent, OR
- Not very toxic to aquatic organisms

→ **Soil Hazard Category 1**

but Helvetolide not relevant in soil !

Helvetol (the degradation product of the registered substance):

- Not highly adsorptive (log Kow = 4,33, predicted log Koc= 2.19-2.81),
- Potentially highly persistent, **BUT**
- Not very toxic to aquatic organisms

→ **Soil Hazard Category 3**

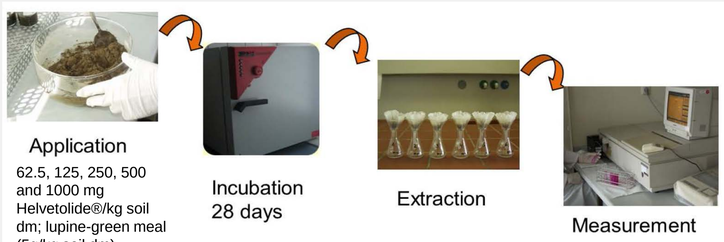
Despite the expected absence of the Parent substance in sludge and soil, long term soil toxicity tests on the registered substance, helvetolide were requested by ECHA.

The industry task force took the decision to **add further analysis** on the concentrations of the expected major and persistent breakdown product, **helvetol** in soil throughout the **earthworm reproduction test.**

Long-term soil toxicity tests on Helvetolide ®

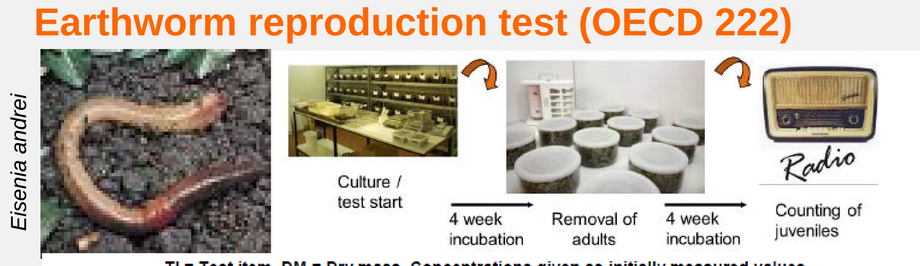
Soil microbial activity

(OECD 216, Nitrogen transformation)



Parameter		EC_{10}	EC_{20}	EC_{50}
		[mg test item/kg dw]		
Nitrogen transformation	Value	161	200	304
	95 %-cl	(122 – 190)	(164 – 227)	(274 – 338)

Earthworm reproduction test (OECD 222)



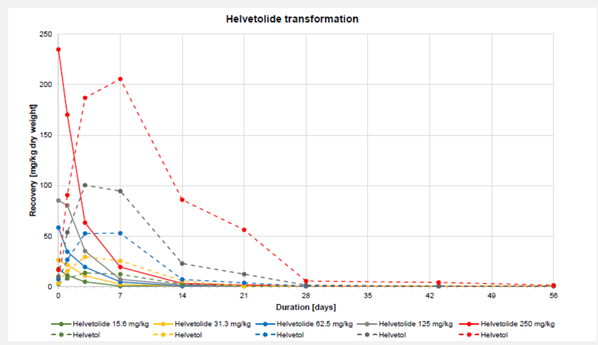
TI = Test item. DM = Dry mass. Concentrations given as initially measured values.

Parameter		EC_{50}	EC_{20}	EC_{10}	LOEC	NOEC
		[mg test item/kg]				
Survival rate	Value	> 234.9	> 234.9	> 234.9	> 234.9	≥ 234.9
	95 %-cl	(n.d.)	(n.d.)	(n.d.)	-	-
Weight change	Value	> 234.9	> 234.9	170.1	> 234.9	≥ 234.9
	95 %-cl	(n.d.)	(n.d.)	(n.d.)	-	-
Reproduction	Value	221.0	95.8	61.8	58.5	26.4
	95 %-cl	(188.1 – 274.3)	(75.0 – 113.8)	(42.5 – 78.3)	-	-

n.d.: not determined.

Lowest EC10

Chemical analyses in soil



Rapid loss of Helvetolide confirmed in soil (disappearance after 7-14 days). Helvetol is formed as the parent degrades and disappears within 28d at the lowest concentrations.

Preliminary results of additional analyses conducted at Firmenich* on soil samples, show that helvetal is formed but not detected anymore at the end of the test.

Soil risk assessment for Helvetolide ®

PEC Predicted Environmental Concentration

The PECs soil based on 100 tons are :

$PEC_{agricultural\ soil}$ =0.277 mg/kg (wide dispersive use)

$PEC_{agricultural\ soil}$ =0.533 µg/kg (PEC regional)

$PNEC_{soil}$ Predicted No Effect Concentration

2 chronic soil toxicity values are available. An Application Factor of 50 is applied to the lowest EC10

$PNEC_{soil}$ = 61.8 / 50 = 1.236 mg / Kg dw

Risk Characterisation Ratio $_{soil}$=PEC/PNEC

The RCR_{soil} for Helvetolide® =0.277/1.236= 0.224

Conclusion

- Helvetolide® is degraded to helvetol when passing through a sewage treatment plant, thus helvetol will be present in the sludge, that may be used to amend agricultural soil
- A more realistic soil risk assessment would be to base the results on helvetol. Nevertheless, since helvetol was also present in the earthworm test, it can be concluded that the toxicity of helvetol to earthworms has also been included in this study.
- The analytical measures of helvetolide and helvetol confirm that both parent and degradation product, will be completely and rapidly lost from the test system and that parent and degradation product(s) will not pose a long term risk to soil organisms.

SETAC EUROPE 29th ANNUAL MEETING
26–30May 2019 I Helsinki, Finland
One Environment. One Health. Sustainable Societies.

Poster WE153 May 29th 2019 : SESSION Advances in Soil Ecotoxicology and Risk assessment – Impact, Ecotoxicity tests, Monitoring and Risk assessment of soil stressors

* Analyses being conduced at Firmenich R&D Analytical Laboratory by Lucie Baroux

Session: Advances in Soil Ecotoxicology and Risk Assessment - Impact, Ecotoxicity Tests, and Concepts for a Retrospective Environmental Risk Assessment (P)
Poster, Exhibition Hall, ID WE153
Wednesday May 30th, 2019, 8:30 AM

Soil risk assessment of the fragrance ingredient, Helvetolide®

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Alicyclic musks and macrocyclic musks are fragrances that were developed in the 1970's as alternatives to the nitromusks and polycyclic musks which were already controversial due to their persistence and bioaccumulation potential. The olfactory characteristics of musks - warm, sensual, animalic and natural - are of great value to perfumers and consumers and as such musks are added in cosmetics as well as in laundry products. Alicyclic musks and macrocyclic alternatives are mostly readily biodegradable and have been shown not to be bioaccumulative despite their relatively high log K_{ow} values. However, some of the earlier developed alicyclic musks are only partially biodegradable according to screening biodegradation tests. We report here the environmental profile of Helvetolide® and focus on the soil risk assessment, whereby the chronic effects on soil microorganisms and worm have been tested.

The inhibitory effects of Helvetolide® on nitrogen transformation activity of soil microorganisms was assessed following the OECD TG 216 resulting in a 28d-EC_{10} value of 161 mg/Kg dw. The effects of Helvetolide® on soil worm reproduction were assessed using *Eisenia andrei* following the OECD TG 222 up to 250 mg Helvetolide®/kg dry mass soil. Soil samples were taken at multiple times for chemical analyses of Helvetolide® and of the transformation product helvetol. Rapid loss of the Helvetolide® was observed within the first week of the test, while most of the helvetol was eliminated after several weeks. The 56d-EC_{10} value based on number of offsprings hatched from the cocoons was determined to be 86 mg/kg dw based on nominal concentration of Helvetolide®. These results are used to derive the PNEC soil and compared with the PNEC soil derived by using the equilibrium partitioning method. Finally the equation of PEC soil/PNEC soil ratio below 1 confirms that Helvetolide® can be used safely and does not pose any risk to soil organisms. In addition, we demonstrate that the degradation product, helvetol is less bioaccumulable and less toxic than the parent molecule Helvetolide®.