Silicone industry's experience with skin sensitisation test methods

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Presentation Outline

Introduction to the Silicone industry

Study outline

Results & discussion

Conclusion

Open questions

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Silicone industry

- The European Silicone Industry is organised under the umbrella of CES (Centre Europeen des Silicones) - a sector group of CEFIC.
- CES provides information on silicones from a health, safety and environmental perspective.
- The Si-industry uses validated alternative methods accepted by regulatory authorities without compromising hazard assessments.
- A number of product applications result in skin contact: Understanding and correctly identifying the potential skin sensitisation hazard of our substances is key to our obligation and commitment to the safe use of our products in the workplace and with consumers.

Skin sensitisation test methods

- The LLNA(OECD 429) has been adopted as a standalone method in 2002 and is the preferred test method for skin sensitisation testing under REACH.
- The LLNA represents a refined skin sensitisation method based on the „3R“ principle and provides valuable information on treshhold dose and dose response curve in comparison with the traditional OECD 406 test guidelines.
- Silicone materials were not part of the LLNA validation project.
- There were indications of apparent false positive findings with silicones tested in the LLNA.
- The Si-industry carried out a small scale - five substances - validation exercise in form of a research project.

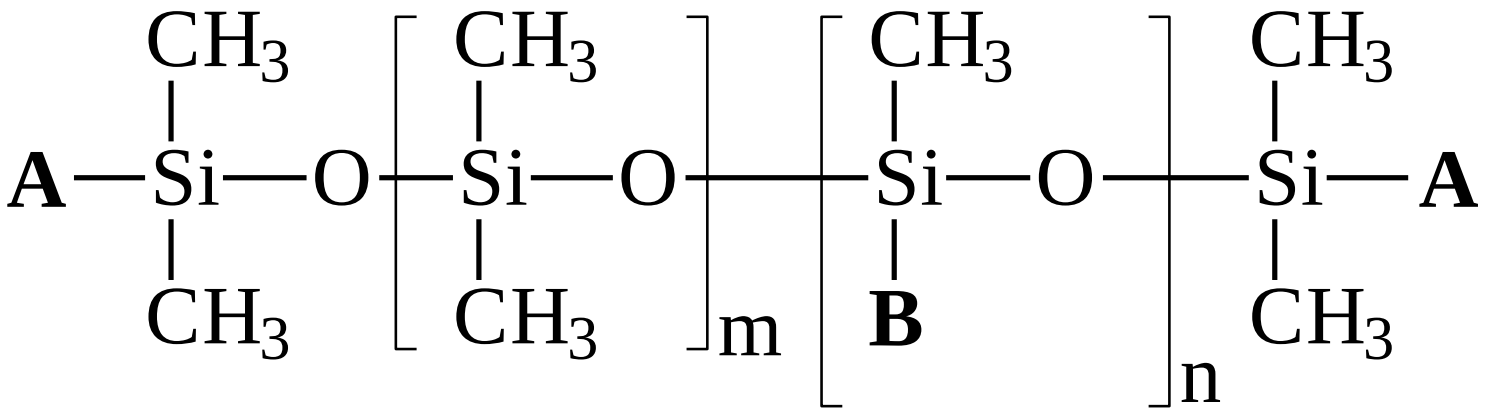
Study outline

- Assessment of the skin sensitisation potential of a total of 5 silicone materials in the
  - Guinea pig maximisation test
  - Mouse local lymph node assay
- Evaluation of discordant findings with regard to
  - Study reliability and methodological issues
  - Possible chemical factors
- Establishment of the weight of the evidence of the 5 silicone materials with regard to their potential to pose a skin sensitisation risk to humans

Test substances

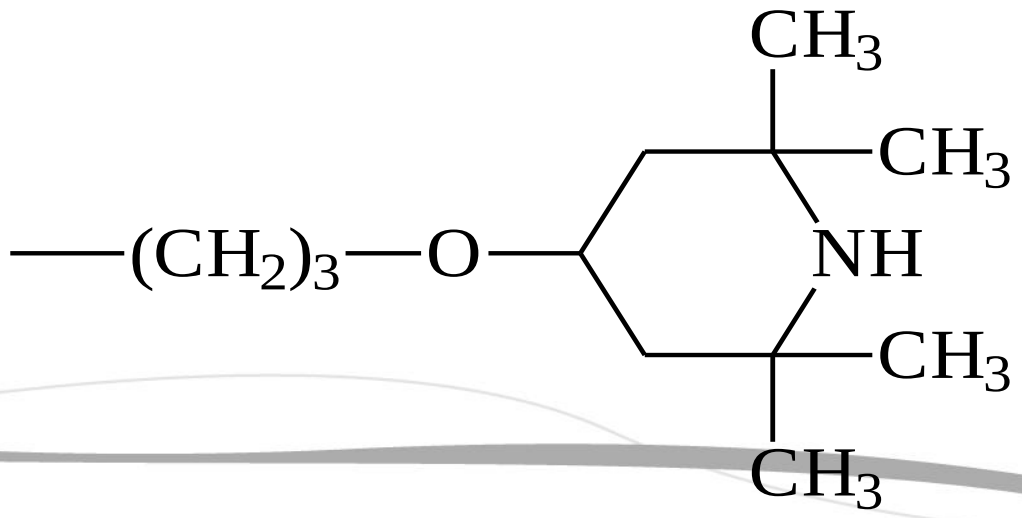
- Propylpipyridyl functionalized silicones (“PFS”)
  - Complex condensation product composed of a large methylated siloxanes ((CH3)2SiO units) chain and a propylpipyridyl group at an approximate mole ratio of 1:96
  - 4 variants with differences in viscosity and amine content
  - Polymer content > 97%
  - MW = 80,000-165,000 g/mol;
- One polyaminofunctional Siloxane (“PAEAMPS”)
  - Composed of a mixture of methoxy-terminated and hydroxyl-terminated Poly[3-((2-aminoethyl)amino)propyl]methyl-(dimethyl)siloxane
  - Polymer content > 98%;
  - MW = 80,000-165,000 g/mol

Propylpipyridyl functionalized silicones (PFS)

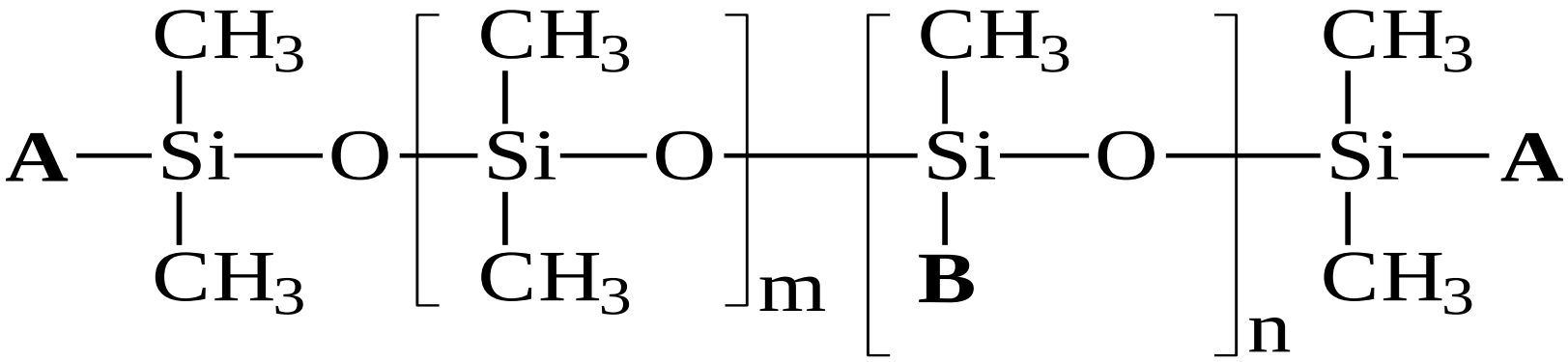


A = CH_3

B =



Polyaminofunctional Siloxane (PAEAMPS)



A = OH

A = OMe

B = $(CH_2)_3$-NH-$(CH_2)_2$-NH_2

Study design LLNA studies

	PFS 45	PFS 45LC	PFS 50	PFS 50LC	PAEAMPS
No. of Animals Preliminary Per dose/Control Positive Control	4 4/4 4	4 4/4 4	4 4/4 4	4 4/4 4	5 5/5 5
Induction pre-test	5; 10; 25; 50% AOO (4:1)	5; 10; 25; 50% AOO (4:1)	5; 10; 25; 50% MEK	2.5; 5; 10; 100% PG	50; 100% AOO (3:1)
Induction main test	2.5; 5; 10; 25; 50% AOO (4:1)	2.5; 5; 10; 25; 50% AOO (4:1)	0.5; 1; 2.5; 5; 10; 25% MEK	0.5; 1; 2.5; 5; 10; 25% MEK	5; 10; 25; 50% AOO (3:1)
Ear thickness measurements	Yes; Days 1,2,3, and 6	Yes; Days 1,2,3, and 6	Yes; Days 1,2,3, and 6	Yes; Days 1,2,3, and 6	Yes; Days 1,2,3, and 6
Positive control	HCA	HCA	HCA	HCA	p-phenylenediamine

Results LLNA studies

	PFS 45	PFS 45LC	PFS 50	PFS 50LC	PAEAMPS
Vehicle	AOO (4:1)	AOO (4:1)	MEK	Propylene Glycol	AOO (3:1)
Concentration	2.5/5/10/25/50%	2.5/5/10/25/50%	0.5/1/2.5/5/10%	0.5/1/2.5/5/10%	5/10/25/50%
Stimulation Index (SI) per dose level	1.1/0.9/1.6/3.68/ 12.2	1.0/0.9/1.7/1.7/ 5.4	1.8/1.8/2.5/6.4/ 22.3	1.2/1.2/1.1/1.3/1 .5	1.6/3.5/18.9/ 22.8
Calc.EC3	20.1%	30%	2.8%	Not applicable	8.7%
SI Control	8.4 (25% HCA)	5.8 (25% HCA)	33.9 (25% HCA)	19.5 (25% HCA)	12.1 (1% PPD)
Increased ear thickness (%)	3/-2/6/2/13	3/4/7/5/10	5/0/3/3/6	0/-1/1/-1/1	23/27/64/142
Clinical signs of irritation	**Slight irritation**	**Very slight irritation**	**No**	**No**	**Yes** (25%/50%)
Report conclusion	**Sensitizer**	Sensitizer	**Sensitizer**	**Not a Sensitizer**	**Sensitizer** Response potentially influenced by irritation
Klimisch Rating	1	2	1	1	2

Study design GPMT studies

	PFS 45	PFS 45LC	PFS 50	PFS 50LC	PAEAMPS
No. of Animals Preliminary Treatment/Control Positive Control	4 20/10 10/5	4 20/10 10/5	4 20/10 10/5	4 20/10 10/5	5 10/5 10/5
Induction Injection	10% w/w Corn oil	10% w/w Corn oil	5% w/w Corn oil	10% w/w Corn oil	5% w/w Olive oil
Pre-treatment	10% SLS	10% SLS	No	No	No
Induction Patch	100% Olive oil	100% Acetone	25% Olive oil	10% Olive oil	75% Olive oil
Challenge patch	100% Olive oil	50% Acetone	25% Olive oil	100% Olive oil	15% Olive oil
Positive control	Yes Mercapto benzo-thiazole	Yes Mercaptobenzo-thiazole	Yes Mercaptobenzo-thiazole	Yes Mercaptobenzo-thiazole	Yes Mercaptobenzo-thiazole

Results GPMT studies

	PFS 45	PFS 45LC	PFS50	PFS 50LC	PAEAMPS
Skin Grades Challenge					
24 hr - Treated	4/20	0/20	0/20	0/20	0/10
24 hr - Control	3/10	0/10	0/10	0/10	0/5
48-hr – Treated	2/20	0/20	0/20	0/20	0/10
48-hr – Control	2/10	0/10	0/10	0/10	0/5
Positive control					
Treated (48ht)	5/10	5/10	5/10	5/10	10/10
Control (48hr)	0/5	0/5	0/5	0/5	0/5
Conclusion	**Not a sensitizer**	**Not a sensitizer**	**Not a sensitizer**	**Not a sensitizer**	**Not a sensitizer**
Klimisch Rating	1?	1	1	1	1

Results summary: GPMT versus LLNA

- All 5 silicone materials were negative in the GPMT studies
- Of the 5 LLNA's
  - One study negative (PFS 50LC)
  - One study positive but strongly influenced by irritation (PAEMPS)
  - Three studies weakly positive (PFS 45; PFS 45LC; PFS 50), but S.I. values above 3 = criteria for C&L

Factors considered in WoE Analysis

- Absence of occupational allergic contact dermatitis in workers with daily skin exposures over a period > 10 years
- Safe use in shampoo formulations at levels ≤ 2% without any evidence of in-market issues
- Complete absence of skin sensitisation response in the GPMT at high induction/challenge concentrations
- Excessive level of irritation in PAEMPS study suggests LLNA to be a false positive
- Generally weak response and absence of clear dose-response in the LLNA with PFS materials
- Absence of any functional groups in the structures known to be associated with skin sensitisation response
- Low dermal penetration potential due to high molecular weight

Conclusions

- WoE suggests none of the examined silicone materials represent a skin sensitisation risk to humans under normal conditions of use
- LLNA produced questionable results for this class of materials and may therefore not be considered as the first choice for skin sensitisation testing.
- Future sensitisation testing strategy for other silicone materials will need to be considered on a case by case basis

Open Questions

- Do and if so, how do weak or non-irritating substances lead to non-specific cell activiation resulting in a positive outcome in the LLNA?
- How much data is sufficient to account as „scientific justification“ versus generating more data , thereby compromising animal welfare ?
- How can new substances be evaluated for their potential skin sensitisation hazard in the future, if little information/data elements are available to go through a WoE approach?

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- Further information about the Silicone industry, their members and on other regional silicone industry associations can be found at : www.silicones-europe.com

- Acknowledgement to Thomas Petry / ToxMinds for the review of the data.

- Thank you for your attention !

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