

SIDS DOSSIER

LINEAR ALKYL BENZENE
SULFONATE (LAS)

CAS NOs. 1322-98-1
25155-30-0
26248-24-8
27636-75-5
68081-81-2
68411-30-3
69669-44-9
85117-50-6
90194-45-9
127184-52-5

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1. GENERAL INFORMATION

1.01 SUBSTANCE INFORMATION

- A. **CAS number** The information provided in this dossier refers to various individual compounds and mixtures of sulfonated linear alkyl benzenes which are identified by the following CAS numbers and names:

CAS No.	EINECS No.	Name
1322-98-1	215-347-5	Decylbenzene sulfonic acid, sodium salt
25155-30-0	246-680-4	Dodecylbenzene sulfonic acid, sodium salt
26248-24-8	247-536-3	Tridecylbenzene sulfonic acid, sodium salt
27636-75-5	248-583-2	Undecylbenzene sulfonic acid, sodium salt
68081-81-2	268-356-1	C ₁₀₋₁₆ Monoalkylbenzene sulfonic acid, sodium salt
68411-30-3	270-115-0	C ₁₀₋₁₃ Alkylbenzene sulfonic acid, sodium salt
69669-44-9	274-070-8	C ₁₀₋₁₄ Alkyl derivatives benzene sulfonic acid, sodium salt
85117-50-6	285-600-2	C ₁₀₋₁₄ Monoalkylbenzene sulfonic acid, sodium salt
90194-45-9	290-656-6	C ₁₀₋₁₃ Alkyl derivatives benzene sulfonic acid, sodium salt
127184-52-5		4-C ₁₀₋₁₃ -sec Alkyl derivatives benzene sulfonic acid, sodium salt

- B. **Name (IUPAC name)** See A.

- C. **Name (OECD name)** Linear alkylbenzene sulfonate (LAS)

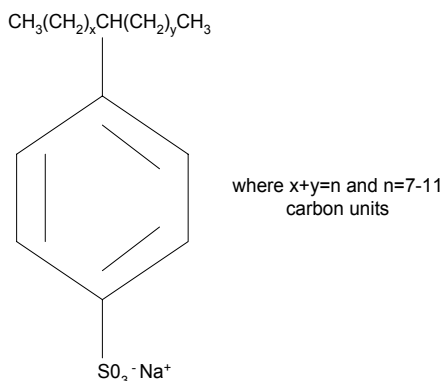
- D. **CAS Descriptor**

Relatively consistent mixture of homologues with predominately linear (currently >95% for most products) varying alkyl chain lengths (from C₁₀ to C₁₄) and phenyl isomers with attachment of the *para* sulfonate (sodium salt) benzene ring to the alkyl chain at non-terminal positions. This description applies to all of the CAS numbers listed in 1.01A as shown by the alkyl chain distribution in the table in 1.01G.

- E. **EINECS-Number** See A.

- F. **Molecular Formula** See G.

- G. **Structural Formula**



The linear alkyl carbon chain typically has 10 to 14 carbon units, with the approximate mole ratio varying somewhat regionally, as shown in the following table:

Region	<C ₁₀	C ₁₀	C ₁₁	C ₁₂	C ₁₃	C ₁₄	>C ₁₄	Range of Averages	Weighted Average*
Canada 68081-81-2	≤1	<16	19-39	20-50	5-27	<3	<1	11.8	11.8
Europe 25155-30-0 68081-81-2 68411-30-3 85117-50-6 90194-45-9 127184-52-5	≤1	8-20	19-39	20-50	5-27	<1-3	<1	11.6-11.8	11.7
Japan 68081-81-2 68411-30-3 69669-44-9	≤1	7-16	19-39	20-50	5-27	<1-3	<1	11.7-11.8	11.8
United States 1322-98-1** 25155-30-0 26248-24-8** 27636-75-5** 68081-81-2 69669-44-9 85117-50-6 90194-45-9	<2	1-25	7-50	20-50	5-45	<1-10	<1	11.3-12.6	11.7

*Weighted by production volume for each region.

**Manufacture of LAS under these CAS numbers has recently been discontinued.

The molecular weights depend on alkyl chain length and range from 338 (C_{11.3}) to 356 (C_{12.6}). As shown in the table, all the LAS category members (CAS numbers) have the alkyl chain distributions for the LAS category. All of the data in this assessment, except for homologue data identified as such, is on LAS category materials having the alkyl chain distribution shown in the table. The available information on the test substances is provided in the robust summary for each test. All results have been corrected for 100% activity.

Commercial LAS is exclusively manufactured as mixtures of C₁₀ to C₁₃ or C₁₄ alkyl chain homologues, having average alkyl chain lengths ranging from C_{11.3} to C_{12.6}, with the predominant materials having average alkyl chain lengths ranging from C_{11.7} to C_{11.8} (Table above). Each alkyl chain homologue consists of a mixture of all the possible sulfophenyl isomers except for the 1-phenyl isomer which is not found in the commercial material. The catalyst used to make the LAB determines the distribution of the phenyl isomers in commercial LAS with the proportion of the 2-phenyl isomers ranging from 18 to 28% (Valtorta et al., 2000). Consequently, commercial LAS consists of a mixture of 20 or more compounds, the 2-phenyl to 5-phenyl isomers of the C₁₀ homologue, the 2-phenyl to 6-phenyl isomers of the C₁₁ and C₁₂ homologues and the 2-phenyl to 7 phenyl isomers of the C₁₃ homologue, etc.

H. Substance Group Not applicable

I. Substance Remark Not applicable

J. Molecular Weight Range depending on alkyl chain length

1.02 OECD INFORMATION

A. Sponsor Country: United States of America

B. Lead Organization:

Name of Lead Organization: United States Environmental Protection Agency

Contact person: Mr. Oscar Hernandez

Address:

1200 Pennsylvania Avenue, N.W.

Washington, D.C. 20460

USA

Tel: (202) 564-7641

Email: hernandez.oscar@epa.gov

C. Name of responder

Name: John Heinze Ph.D., Consortium Manager

Address:

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Consortium Participants

Center for LAB Environmental and Technical Studies for Asia (CLETSA)

Cognis Deutschland GmbH&Co.KG

Colgate-Palmolive Company

Huntsman Corporation

Kao Corporation

Lion Corporation

Petresa International N.V.

Quimica Venoco, CA

YPF SA

Sasol North America

Stepan Company

TAYCA Corporation

The Dial Corporation

The Procter & Gamble Company

Unilever Household and Personal Care North America

Additional Participants

Mitsubishi Chemical Corporation

Nippon Petrochemicals Co., Ltd.

W.R. Grace & Company

1.03 CATEGORY JUSTIFICATION

The LAS molecule contains an aromatic ring sulfonated at the para position and attached to a linear alkyl chain at any position except the terminal carbons. The alkyl carbon chain typically has 10 to 14 carbon atoms and the linearity of the alkyl chains ranges from 87 to 98%. While commercial LAS consists of more than 20 individual components, the ratio of the various homologues and isomers, representing different alkyl chain lengths and aromatic ring positions along the linear alkyl chain, is relatively constant in currently produced products, with the weighted average carbon number of the alkyl chain based on production volume per region between 11.7-11.8. LAS is supported as a category because of the close consistency of the mixtures, their commercial uses, fate, and health and environmental effects. LAS is the primary cleaning agent used in many laundry detergents and cleaners at concentrations up to 25 percent in consumer products, and up to 30 percent in commercial products, with the exception of one reported product at 45% percent in concentrated solid form that is mechanically dispensed into diluted solution for dishwashing.

1.1 GENERAL SUBSTANCE INFORMATION

A. Type of Substance

element []; inorganic []; natural substance []; organic [X]; organometallic []; petroleum product []

B. Physical State (at 20°C and 1.013 hPa)

gaseous []; liquid []; solid [X] for pure substance

C. Purity

87-98%

Purity refers to the percent LAS, with iso-branched LAS and DATS considered to be impurities. The “activity” may also be stated, and represents the percent of active LAS in the solution (e.g., 50% active LAS is half strength LAS, the LAS of which will be 87-98% pure, with the remaining 50% consisting of water).

D. Manufacturing Process

LAS is manufactured from linear alkylbenzene (LAB) in self-contained, enclosed systems (see SIAR Annex, Format A, Section VI(1), for more information). LAB is produced by reacting paraffins with benzene and a catalyst and isolating the LAB by distillation. The LAB is then sulfonated, which in turn is then neutralized to sodium salts of LAS.

1.2 SYNONYMS

Linear alkylbenzene sulfonates
LAS
Sodium-n-alkyl (C₁₀₋₁₃) Benzene Sulfonate
Numerous trade names, e.g. Marlon A

1.3 IMPURITIES

Remarks: Dialkyltetralin Sulfonates (DATS) and single methyl-branched alkyl chain LAS (iso-LAS) make up minor components in commercial LAS. Concentrations range from <1 to 8% for DATS and <1 to 6% for iso-LAS, depending on the manufacturing process used. Recent market information (LAS SIDS Consortium, unpublished, 2005) indicates that less than 5% of the global LAS production contains high levels of DATS. Consequently, the average (based on production volume) linearity and purity of LAS worldwide is greater than 95%. The presence of DATS and iso-LAS did not significantly affect the biodegradation of LAS relative to the pure linear component, as both DATS and iso-LAS are biodegradable substances. This is discussed in detail in the summaries at section 3.5(x) and 3.5(y).

References: 1) Nielsen, A.M., Britton, L.N., Beall, C.E., McCormick, T.P. and Russell, G.L. 1997. Biodegradation of coproducts of commercial linear alkylbenzene sulfonate. Environ. Sci. Technol. 31L3397-3404.
2) Cavalli, L., Cassani, G., Lazzarin, M., Maraschin, C., Nucci, G., and Valtorta, L. 1996b. Iso-branching of linear alkylbenzene sulphonate (LAS). Tenside Surf. Det. 33:393-398.
3) LAS SIDS Consortium, unpublished, 2005.

1.4 ADDITIVES

Value: None
Remarks: No additives

1.5 QUANTITY

(a)
Remarks: Year 2000 data, as reported in a Colin A. Houston report, indicate an estimated volume of LAS consumption in North America (United States and Canada combined) as 390,000 metric tonnes.

Reference: Colin A. Houston. 2002. Surfactant Developments. Forecast to 2010. A Multiclient Study. Colin A. Houston & Associates, Inc., August 2002.

(b)
Remarks: In a market survey completed by ECOSOL in 2000, companies in Europe reported a total consumption of LAS of approximately 400,000 metric tonnes. This includes CAS numbers 1322-98-1, 25155-30-0, 68411-30-3, 85117-50-6 and 90194-45-9.

Reference: HERA. 2004. HERA-LAS Human and Environmental Risk Assessment: Linear Alkylbenzene Sulphonates, LAS. CAS No. 68411-30-3, Version 2.0, May 2004; available at www.heraproject.com.

(c)
Remarks: Total LAS production for the companies surveyed in the most recent year for which data are available (generally 2002) was approximately 430,000 metric tonnes. Almost half of this production (198,000 metric tonnes) occurred in North America (United States and Canada combined). Production in Europe, as reported by the member companies surveyed, was approximately 152,000 metric tonnes. Data cited above in (a) and (b)

for LAS consumption in the United States and Europe are viewed as the more reliable estimates, because all LAS producers are not included in the coalition member survey. Production in Japan, where all the LAS producers are members of the consortium, was 85,000 metric tonnes and is considered a reliable estimate.

Reference: Industry Coalition for the SIDS Assessment of LAS Survey conducted in 2002 (LAS SIDS Consortium Survey, 2002).

(d)

Remarks: More than 1 million tonnes per annum produced globally based on: (1) 364 to 415 ktonnes produced annually in U.S. from 1987 to 1991; (2) 400 ktonne produced in Western Europe and 2570 ktonnes world-wide in 1995; (3) 950 ktonnes produced in Europe, North America and Japan in 1994; (4) 2 million tonnes consumed world-wide in 1990; (5) 410 ktonne produced in Western Europe, 2.6 million tonnes worldwide in 1995.

Reference: (1) CHIMICA OGGI, Sept. 1998
(2) EU Risk Assessment Report for LAB, May 1997
(3) IPCS Environmental Health Criteria 169, WHO, 1996
(4) Nielsen et al. 1997
(5) Soap and Detergent Association, 1996

1.6 LABELLING AND CLASSIFICATION

Labelling

Remarks: None designated

Classification

Remarks: None designated

1.7 USE PATTERN

A. General

Type of Use:

main
industrial
use

Category:

Wide dispersive use
Personal and domestic use
Cleaning/Washing agent

Remarks: About 78-97% of the LAS consumption worldwide is in liquid and powder consumer and industrial laundry and fine fabric detergents. Another 2-10% of the LAS produced is used in consumer and industrial dishwashing liquids, with the remainder (1-5%) used in other consumer and industrial cleaners.

B. Uses in Consumer Products

<u>Function</u>	<u>Amount present</u>	<u>Physical state</u>
detergent	up to 25% of formulation	powder or liquid

Remarks: LAS is an anionic surfactant that lowers the surface tension of water, enabling soils and stains to loosen and release from fabrics and surfaces. LAS is the primary cleaning agent used in many liquid and powder laundry detergents and speciality household cleaners at concentrations up to 25 percent of the total formulation.

The following table shows the percentage of LAS that occurs in various types of consumer detergent products.

Consumer Product Type	Range of Percent Composition that is LAS		
	North America	Europe	Japan
Laundry Detergents			
- Powders	5-25%	5-25%	5-25%
- Liquids	1-25%	5-10%	5-25%
- Tablets	5-25%	10-25%	5-25%
Liquid Fine Fabric Detergents	-	-	1-5%
Bleaches	-	-	0.1-0.5%
Pre-Washes	-	-	5-10%
Fabric Conditioners (sheets)	0.1-0.5%	-	-
Dishwashing Detergents (liquids)	5-25%	10-25%	1-5%
General Cleaners (dilutable)	1-5%	1-5%	-
Hard Surface Cleaners	1-5%	0.1-0.5%	0.5-10%
Other Cleaners	-	-	0.1-0.5%
Face & Hand Soaps (bar)	1-5%	-	-

Reference: 1) Soap and Detergent Association 1996
2) Survey data for Industry Coalition for the SIDS Assessment of LAS. 2002.

C. Uses in Institutional and Industrial Products

The following table shows the percentage of LAS that occurs in various types of institutional and industrial detergent products.

Industrial Product Type	Range of Percent Composition that is LAS		
	North America	Europe	Japan
Laundry Detergents			
- Powders	5-25%	5-10%	5-10%
- Liquids	-	10-25%	-
Pre-Washes	-	10-25%	-
Dishwashing Detergents (liquids)	5-10%	25-30%	5-30%
General Cleaners			
- Dilutable	1-5%	-	-
- Spray	1-5%	-	-
Hard Surface Cleaners	-	-	1-10%
Disinfectants (liquids)	5-10%	-	-
Other Uses	25-30%*	10-25%	10-25%

* The only exception is a product containing 45% LAS that is a concentrated solid mechanically dispensed into diluted solution for dishwashing.

Reference: 1) Soap and Detergent Association 1996
2) Survey data for Industry Coalition for the SIDS Assessment of LAS. 2002.

1.8 OCCUPATIONAL EXPOSURE LIMIT VALUE

Exposure limit value

Type: None established by OSHA, ACGIH or NIOSH

Short term exposure limit value

Value: None established by OSHA, ACGIH or NIOSH

1.9 SOURCES OF EXPOSURE

Remarks: Exposure to industrial workers is limited because this is an enclosed manufacturing process designed to minimize losses and the potential for release. Worker exposure is possible during the detergent formulation stage by inhalation of powders or dermal contact of powders and liquids. However, good manufacturing design practices (e.g. enclosed production, exhaust ventilation, dust collection) and personal protective equipment (e.g. protection clothing, eyewear, and glove) in place of at facilities that manufacture liquid and dry (granular/powder) materials sufficiently mitigate worker exposure to LAS. No special engineering controls or additional personal protective equipment are uniquely specified for LAS.

LAS is used primarily in household laundry and dishwashing cleaning products. After use, LAS is discharged into the wastewater treatment system. The exposure of the general human population and of environmental organisms depends on the

application of LAS, the local sewage treatment practices, and on the characteristics of the receiving environment.

It is reasonable to consider that the tasks with the greatest exposure to the consumer are hand dishwashing and hand washing of clothing.

Reference: 1) EU Risk Assessment Report for LAB, May 1997.
2) IPCS Environmental Health Criteria 169, WHO, 1996.

1.10 ADDITIONAL REMARKS

A. Options for disposal

Remarks: Unused LAS may be recovered for reprocessing or disposed of by incineration or landfill or by flushing to sewage system; used material enters sewage system and is treated at WWTP. Spills may be recovered for reprocessing or disposal.

Reference: MSDS.

B. Other remarks

(a)

Remarks: The majority of LAS is disposed of in sewage during use as cleaning/washing agents.

Reference: Soap and Detergent Association 1996.

(b)

Methods: A study conducted for the Soap and Detergent Association (Battelle 1999) measured the under 10 micron fraction delivered from 6 consumer product spray nozzles. The six standard trigger sprayers (TS800) were manufactured by Calmar Dispensing System, Inc. The specified average output of the sprayers, based on water at 90 strokes per minute, is no less than 0.75 mL per stroke. The specified spray pattern is a nearly circular pattern with a diameter of no less than four inches at a distance of approximately eight inches. The six trigger sprayers were evaluated to determine emitted aerosol size distribution, output per stroke and spray pattern in order to avoid choosing a trigger sprayer with abnormal characteristics for the experiment. Size distribution of aerosols generated from the six sprayers was measured using a laser diffraction particle sizer (Mastersizer Model X, Malvern Instruments Ltd).

Remarks: The overall mean (n=30) is 0.11% particles under 10 microns and the standard deviation is 0.21. The very highest observation was 0.80% particles under 10 microns. This testing only captured the spray particles that are under 600 microns, so the actual mean respirable particle percent of total volume sprayed is less than 0.1%. The Battelle (1999) study also reported that for consumer spray products in normal use conditions, the peak breathing zone concentration under 10 microns ranged from 0.13-0.72 mg/m³. HERA (2004) reported that measurements of aerosol particles under 6.4 microns in size generated upon spraying with typical surface cleaning spray products resulted in a product concentration of 0.35 mg/m³. These estimates of exposure to respirable particles from consumer spray products indicate that inhalation is not a likely route of concern for human exposure (see SIAR Annex 1 for more information). Estimates of inhalation exposure apply to both consumer and commercial

products as both use the same type of spray nozzles (for spray cleaners) and the same type of equipment to make powder/granulated products. The human experience with eye irritation covers both manufacturing and use of consumer and commercial products.

- References:
- 1) Battelle, Inc. 1999. Measurement and characterization of aerosols generated from a consumer spray product-pilot study. Final report to the Soap and Detergent Association. Battelle Study No. N003043A. January.
 - 2) HERA. 2004. Linear Alkylbenzene Sulphonate, LAS. Human & Environmental Risk Assessment on ingredients of European household cleaning products. Version 2.0, May 2004.
<http://www.heraproject.com/RiskAssessment.cfm>

(c)

Methods: A comprehensive testing program was undertaken to evaluate consumer exposure to dust from powdered enzyme detergent use in comparison with worker exposure at factories. Airborne dust was collected in consumers' homes during normal use of laundry detergents. Consumer use of laundry products was then simulated in the laboratory to permit collection of sufficient samples for analysis of the amount of enzyme in detergent dust, and for detergent dust particle size distribution determinations and persistence measurements. Representative commercial products sold by Procter & Gamble were tested. Air sampling was carried out using an electrostatic precipitator using a battery powered source and was conducted continuously from the time each housewife began to pour laundry product for use until she left the laundry area. The entry orifice of the sampling device was located at a point spatially equivalent to the direction and distance from the housewife's nose from the point of dust generation. Laboratory simulation of consumer practices was based on extensive consumer habits developed by a variety of conventional techniques.

Remarks: The results of the in-home studies indicate that detergents contribute only 5% of the dust present during the time detergents are dispensed for laundering, with the rest of the dust believed to be mainly lint. Virtually all detergent dust (95%) settled in less than 2 minutes. On average, there is 0.27 µg detergent dust exposure per cup of product used for double-pour machine laundering. Based on this amount, HERA (2004) calculated the amount of LAS exposure from laundry detergent use. Up to 22% (0.06 µg/use) of the detergent dust can be expected to be LAS. Assuming a worst case exposure (all dust is inhaled and laundry is done 3 times a day), the exposure to LAS of an average adult is estimated to be 0.003 µg/kg bw/day. This amount does not contribute significantly to the total exposure of LAS as compared to the amount from inhalation of aerosols from cleaning sprays, which is approximately 10-fold higher (0.04 µg/kg bw/day).

- References:
- 1) Hendricks, M.H. 1970. Measurement of enzyme laundry detergent product dust levels and characteristics in consumer use. J. Am. Oil Chem. Soc. 47:207-211.
 - 2) HERA. 2004. Linear Alkylbenzene Sulphonate, LAS. Human & Environmental Risk Assessment on ingredients of European household cleaning products. Version 2.0, May 2004.
<http://www.heraproject.com/RiskAssessment.cfm>

2. PHYSICAL-CHEMICAL DATA

2.1 MELTING POINT

(a)

Value: 198.5°C

Decomposition: Onset at 444°C (47% weight loss at 500°C)

Method: Thermal analysis was performed on the Netzsch DSC 204C and TG209C with N₂ atmosphere.

GLP: Yes No ?

Test Substance: C₁₀₋₁₄ Monoalkylbenzene sulfonic acid, sodium salt (CAS #85117-50-6); mean molecular weight = 348, average alkyl chain length = C_{12.0}. The test material is a commercial product, contains 85% active matter and is a coarse, cream-colored powder at 25°C.

Remarks: This measured melting point value is significantly lower than the EPI Suite estimated values for other LAS materials. Note that the activity represents the total percent active materials (LAS, iso-LAS and DATS) in the test substance. The nonactive material in a powdered sample of LAS is likely sodium sulfate and other salts which have very high melting points (e.g., sodium sulfate = 884°C) and would not interfere with the measurement of the LAS melting point.

Reference: Huntsman. 2002. Report of melting point analysis for NANSA HS 85/5. Cover memo from A. Ashworth to K.B. Sellstrom dated April 12, 2002.

Reliability: 2 Valid with restrictions

(b)

Value: 274°C

Decomposition: Not identified

Method: Estimation: EPI Suite (Mean or Weighted MP)

GLP: Yes No ?

Test Substance: C₁₀ LAS (CAS #1322-98-1)

Remarks: Structure modeled is the pure C₁₀ sodium salt homologue, 2-phenyl isomer, not the commercial material.

Reference: USEPA. 2000. EPI Suite v3.10.

Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(c)

Value: 279°C

Decomposition: Not identified

Method: Estimation: EPI Suite (Mean or Weighted MP)

GLP: Yes No ?

Test Substance: C₁₁ LAS (CAS #27636-75-5)

Remarks: Structure modeled is the pure C₁₁ sodium salt homologue, 2-phenyl isomer, not the commercial material.

Reference: USEPA. 2000. EPI Suite v3.10.

Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(d)

Value: 284°C

Decomposition: Not identified

Method: Estimation: EPI Suite (Mean or Weighted MP)

GLP: Yes No ?

Test Substance: C₁₂ LAS (CAS #25155-30-0)
Remarks: Structure modeled is the pure C₁₂ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(e)
Value: 290°C
Decomposition: Not identified
Method: Estimation: EPI Suite (Mean or Weighted MP)
GLP: Yes No ?
Test Substance: C₁₃ LAS (CAS #26248-24-8)
Remarks: Structure modeled is the pure C₁₃ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

2.2 BOILING POINT

(a)
Decomposition: Onset at 444°C (47% weight loss at 500°C)
Method: Thermal analysis was performed on the Netzsch DSC 204C and TG209C with N₂ atmosphere.
GLP: Yes No ?
Test Substance: C₁₀₋₁₄ monoalkylbenzene sulfonic acid, sodium salt (CAS #85117-50-6); mean molecular weight = 348, average alkyl chain length = C_{12.0}. The test material is a commercial product, contains 85% active matter and is a coarse, cream-colored powder at 25°C.
Remarks: Note that the activity represents the total percent active materials (LAS, iso-LAS and DATS) in the test substance. The nonactive material in a powdered sample of LAS is likely sodium sulfate and other salts which have very high melting points (e.g., sodium sulfate = 884°C) and would not interfere with the measurement of the LAS melting point.
Reference: Huntsman. 2002. Report of melting point analysis for NANSA HS 85/5. Cover memo from A. Ashworth to K.B. Sellstrom dated April 12, 2002.
Reliability: 2 Valid with restrictions

(b)
Value: 630°C
Method: Estimation: EPI Suite (Adapted Stein & Brown method)
GLP: Yes No ?
Test Substance: C₁₀ LAS (CAS #1322-98-1)
Remarks: Structure modeled is the pure C₁₀ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(c)
Value: 642°C

Method: Estimation: EPI Suite (Adapted Stein & Brown method)
GLP: Yes No ?
Test Substance: C₁₁ LAS (CAS #27636-75-5)
Remarks: Structure modeled is the pure C₁₁ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(d)
Value: 654°C
Method: Estimation: EPI Suite (Adapted Stein & Brown method)
GLP: Yes No ?
Test Substance: C₁₂ LAS (CAS #25155-30-0)
Remarks: Structure modeled is the pure C₁₂ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(e)
Value: 665°C
Method: Estimation: EPI Suite (Adapted Stein & Brown method)
GLP: Yes No ?
Test Substance: C₁₃ LAS (CAS #26248-24-8)
Remarks: Structure modeled is the pure C₁₃ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

2.3 DENSITY

(a)
Type: Bulk density ; Density ; Relative Density
Value: 1.06 g/cm³
Temperature: 20°C
GLP: Yes No ?
Test Substance: Marlon A 390 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}
Remarks: Source cited in IUCLID is Sicherheitsdatenblatt "Marlon A 390; Huels Ag. vom 03.12.93
Reference: Cited in IUCLID Data Sheet for CAS #68411-30-3.
Reliability: 4 Not assignable. Original report not available for review.

(b)
Type: Bulk density ; Density ; Relative Density
Value: 0.45 g/cm³ (450 kg/m³)
Temperature: 20°C
GLP: Yes No ?
Test Substance: C₁₀₋₁₄ monoalkylbenzene sulfonic acid, sodium salt (CAS #85117-50-6); mean molecular weight = 348, average alkyl chain length = C_{12.0}. The test material is 85% active matter and is a coarse, cream-colored powder at 25°C.

Reference: Huntsman 2002. Report on the melting point analysis for NANSA HS 85/S. Attached technical bulletin dated 04-01-1994. Cover memo from A. Ashworth to K.B. Sellstrom dated April 12, 2002.
Reliability: 4 Not assignable. Original report not available for review.

(c)

Type: Bulk density [X]; Density []; Relative Density []
Value: ca. 550 kg/m³
GLP: Yes [] No [X] ? []
Test Substance: Marlon A 390 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}
Remarks: Source cited in IUCLID is Sicherheitsdatenblatt "Marlon A 390; Huels Ag. vom 03.12.93
Reference: Cited in IUCLID Data Sheet for CAS #68411-30-3.
Reliability: 4 Not assignable. Original report not available for review.

2.4 VAPOR PRESSURE

(a)

Value: 3 x 10⁻¹³ Pa
Method: Calculation
Test Substance: C₁₂ LAS (CAS #25155-30-0)
Remarks: Cites estimates calculated by Lyman (see References).
Reference: 1) HERA. 2002. HERA-LAS Human and Environmental Risk Assessment: Linear Alkylbenzene Sulphonates, LAS. CAS No. 68411-30-3, Draft #6, May 2002.
2) Lyman, W.J. 1985. Environmental exposure from chemicals, V.I, p.31, Neely, W.B., and Blau, G.E., editors. CRC Press. Boca Raton.
Reliability: 4 Not assignable. Original report not available for review.

(b)

Value: 2.88 x 10⁻¹² Pa
Temperature: 25°C
Method: Estimation: EPI Suite (Modified Grain Method)
GLP: Yes [] No [X] ? []
Test Substance: C₁₀ LAS (CAS #1322-98-1)
Remarks: Structure modeled is the pure C₁₀ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v.3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(c)

Value: 1.22 x 10⁻¹² Pa
Temperature: 25°C
Method: Estimation: EPI Suite (Modified Grain Method)
GLP: Yes [] No [X] ? []
Test Substance: C₁₁ LAS (CAS #27636-75-5)
Remarks: Structure modeled is the pure C₁₁ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v.3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(d)
Value: 5.13×10^{-13} Pa
Temperature: 25°C
Method: Estimation: EPI Suite (Modified Grain Method)
GLP: Yes No ?
Test Substance: C₁₂ LAS (CAS #25155-30-0)
Remarks: Structure modeled is the pure C₁₂ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v.3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(e)
Value: 2.16×10^{-13} Pa
Temperature: 25°C
Method: Estimation: EPI Suite (Modified Grain Method)
GLP: Yes No ?
Test Substance: C₁₃ LAS (CAS #26248-24-8)
Remarks: Structure modeled is the pure C₁₃ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v.3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

2.5 PARTITION COEFFICIENT $\log_{10}P_{ow}$ ($\log_{10}K_{ow}$)

(a)
Log Pow: 3.32
Method: Calculation
GLP: Yes No ?
Test Substance: C_{11.6} LAS
Remarks: Calculated for C_{11.6} LAS using the QSAR method of Leo and Hansch (1979) as modified by Roberts (1991) for surfactant structures. This takes into account the various phenyl positions along the linear alkyl chain. See the Roberts (1991) summary at 2.5(g) for a full description of the method modifications.
Reference: 1) HERA. 2002. HERA-LAS Human and Environmental Risk Assessment: Linear Alkylbenzene Sulphonates, LAS. CAS No. 68411-30-3, Draft #6, May 2002.
2) Leo, A.J. and Hansch, C. 1979. Substituent Constants for Correlation Analysis in Chemistry and Biology. J. Wiley & Sons, N.Y.
3) Roberts, D.W. 1991. QSAR issues in aquatic toxicity of surfactants. Sci. Total Environ. 109/110:557-568.
Reliability: 2 Valid with restrictions. These results are considered reliable because a standard calculation technique was employed.

(b)
Log Pow: 1.94
Method: Estimation: EPI Suite
GLP: Yes No ?
Test Substance: C₁₀ Decylbenzene sulfonic acid, sodium salt (CAS #1322-98-1)

Remarks: Structure modeled is the pure C₁₀ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(c)

Log Pow: 2.43
Method: Estimation: EPI Suite
GLP: Yes No ?
Test Substance: C₁₁ LAS (CAS #27636-75-5)
Remarks: Structure modeled is the pure C₁₁ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(d)

Log Pow: 2.92
Method: Estimation: EPI Suite
GLP: Yes No ?
Test Substance: C₁₂ LAS (CAS #25155-30-0)
Remarks: Structure modeled is the pure C₁₂ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(e)

Log Pow: 3.42
Method: Estimation: EPI Suite
GLP: Yes No ?
Test Substance: C₁₃ LAS (CAS #26248-24-8)
Remarks: Structure modeled is the pure C₁₃ sodium salt homologue, 2-phenyl isomer, not the commercial material.
Reference: USEPA. 2000. EPI Suite v3.10.
Reliability: 2 Valid with restrictions. Standard EPA estimation software.

(f)

Remarks: In its review of LAS and related compounds, IPCS notes that while the octanol-water partition coefficient can be calculated in practice, it is impossible to measure P_{ow} for surface-active compounds like LAS. This has been confirmed by Roberts (2000).
Reference: 1) IPCS. 1996. Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates and Related Compounds. World Health Organization, Geneva, Switzerland.
2) Roberts, D.W. 2000. Use of octanol/water partition coefficient as hydrophobicity parameters in surfactant science. 5th World CESIO Congress 2:1517-1524, May-June 2000, Firenze, Italy.

(g)

Methods: Acute lethal toxicity data for a range of anionic and non-ionic surfactants were analyzed with the objective of determining whether QSARs can be developed relating toxicity to calculated log P values. Approaches to dealing with the deficiencies in the Leo and Hansch (1979) fragment

method for calculating log P of surfactants (related to mixtures and phenyl isomer position) were developed and applied to the general narcosis QSAR of Könemann (1981) as represented by Equation 1:

$$\text{Log } (1/\text{LC}_{50}) = 0.87\log P + 1.13 \quad (\text{EQ } 1)$$

(for 14-d LC_{50} tests on guppies; $n = 50$, $r = 0.998$, $s = 0.237$)

Mixtures

Two approaches were taken in this paper to address mixtures. In the first approach, P was calculated for each component individually then multiplied by the mole fraction and summed to give a weighted average log P. Alternatively, when only the overall average composition was known, log P was calculated for the average structure.

Phenyl isomer position

Since the fragment method gives values for log P that are independent of branch (i.e., phenyl isomer) position, a position-dependent branch factor (PDBF) was defined. Branching results in a decrease in the number of water molecules required to solvate the hydrocarbon chain by allowing water molecules to be shared between the two branches. Where both branches are long the water sharing effect should continue, although to a decreasing extent with increasing distance from the branching position, as long as the branches can be paired. To model this, a water sharing function $\log (CP + 1)$ was defined in which CP is found by pairing off carbon atoms along the two branches up to the terminus of the shorter branch. Regression analysis correlating $\log (1/\text{LC}_{50})$ to goldfish with a combination of ALP [representing log P calculated without a branch factor] and $\log (CP + 1)$ [representing the water sharing function]:

$$\text{Log } (1/\text{LC}_{50}) = 0.78\text{ALP} - 1.13 \log (CP + 1) + 2.06 \quad (\text{EQ } 2)$$

(for LC_{50} tests on guppies; $n = 20$, $r = 0.997$, $s = 0.041$)

Further, dividing the first two terms on the right of EQ 2 by 0.78 gave, assuming the role of the second term to be solely that of the branching factor, the following equation:

$$\text{Log } P = \text{ALP} - 1.44 \log (CP + 1) \quad (\text{EQ } 3)$$

Thus, the PDBF was defined as $-1.44 \log (CP + 1)$. Further details on this method are described in Roberts (1989).

Log P values calculated using EQ 3 were found to give good correlations with published river sediment sorption partition coefficients for LAS compounds, supporting the applicability and validity of the PDBF. Log P values calculated using EQ 3 were also used successfully in regression of toxicity data for pure LAS homologues and isomers to *Daphnia magna* and *Gammarus pulex*. The basic equation is similar to EQ 1 and has the general form:

$$\text{Log } (1/\text{LC}_{50}) = a\log P + b \quad (\text{EQ } 4)$$

with the values for a, b and regression data shown in the following table:

	<i>Daphnia</i> (H)	<i>Daphnia</i> (S)	<i>Gammarus</i> (H)	<i>Gammarus</i> (S)
Value of a	0.7	0.64	0.76	0.71
Value of b	2.23	2.44	2.46	2.27
Regression data				
n	9	12	9	11
r	0.987	0.955	0.966	0.950
s	0.07	0.15	0.13	0.16
F	263	103	98	83

Notes: H = hard water (250 mg/L CaCO₃); S = soft water (25 mg/L CaCO₃). Strongly negative outliers omitted.

- Results:** The log P coefficients and intercepts for goldfish, *Daphnia*, and *Gammarus* are all intermediate between those of Könemann's QSAR equation (EQ 1), suggesting that a narcosis (possibly polar) mechanism applies to LAS acute toxicity. Agreement between observed and calculated toxicities for LAS is good. The fact that QSARs derived from compounds whose log P values are calculated with the PDBF give good predictions for unbranched compounds supports the validity of the PDBF for the type of branching encountered in LAS.
- Remarks:** The analyses presented in this paper indicates that the problems of calculating log P for surfactants can be overcome. The case for the applicability of the PDBF appears compelling, although it is based on indirect evidence. In terms of acute aquatic toxicity, anionic surfactants like LAS do not seem greatly different from unreactive non-surfactant organic chemicals. Anionic surfactants of various types have log (1/LC₅₀) values which are well predicted on the basis of their calculated log P values by QSAR equations resembling those associated with the polar narcosis mechanism.
- Reference:**
- 1) Roberts, D.W. 1991. QSAR issues in aquatic toxicity of surfactants. *The Science of the Total Environment* 10/110:557-568.
 - 2) Roberts, D.W. 1989. Aquatic toxicity of linear alkyl benzene sulphonates (LAS) – A QSAR Approach. *Comunicaciones presentadas a las Jornadas del Comité Español de la Detergencia* 20:35-43. Also in J.E. Turner, M.W. England, T.W. Schulz and N. J. Kwaak (Eds) *QSAR 88. Proc. Third Int. Workshop on Quantitative Structure-Activity Relationships in Environmental Toxicology* 22-26 May 1988, Knoxville, TN, pp. 91-98. Available from NTIS.
 - 3) Könemann, H. 1981. Quantitative structure-activity relationships in fish toxicity studies: Part I. Relationships for 50 industrial pollutants. *Toxicology* 19:209-221.
 - 4) Leo, A.J. and Hansch, C. 1979. *Substituent Constants for Correlation Analysis in Chemistry and Biology*, New York.
- Reliability:** 2 Valid with restrictions. Well documented QSAR analysis.

2.6 WATER SOLUBILITY

A. Solubility

(a)

Value: CMC = 0.1 g/L (C₁₂ LAS)

Description: Miscible in water

Method: Surface tension of aqueous solutions of NaLAS was measured by an automated Lauda TE IC by the duNouy ring method. All solutions were prepared with water passed through a Nanopure water filter system and with sufficient salt to equalize the ionic strength. Solubility of commercial NaLAS was determined by slowly lowering the temperature in a Fisher Isotemp incubator over several days and recording the temperature at which the solutions first turned cloudy (cloud point). Clear points were determined by raising the temperature slowly over several days and recording when the solution cleared. The solubility of the C₁₂ narrow-distribution phenyl isomers was determined by heating a saturated solution to 70°C for 0.5-3 h and then cooling to 5°C. After equilibrating to room temperature overnight, the cloudy mixture was centrifuged, and the clear supernatant drawn off, weighed, and dehydrated to determine percentage solids. The percentage dissolved was then calculated from the weight loss to the supernatant.

GLP: Yes No ?

Test Substance: C₁₂ LAS and commercial C₁₁₋₁₃ LAS (sodium salts)

Remarks: Surface activity increased with increasing average alkyl chain length. Some decrease in surface tension was observed with increasing phenyl isomer number, but the range of phenyl isomer distribution of commercial products is not large enough to significantly alter the surface tension vs. log concentration plot. Increasing the average alkyl chain length of LAS decreases solubility (i.e., increases cloud point). The following table shows the critical micelle concentration (CMC) for various chain lengths LAS tested.

Chain length, isomer composition, dialkyltetralin sulfonate content	CMC (g/L)
C ₁₁ high 2-phenyl, low dialkyltetralinsulfonate	0.120
C ₁₁ high 2-phenyl, high dialkyltetralinsulfonate	0.120
C ₁₁ low 2-phenyl, low dialkyltetralinsulfonate	0.120
C ₁₂ low 2-phenyl, low dialkyltetralinsulfonate	0.105
C ₁₃ low 2-phenyl, low dialkyltetralinsulfonate	0.038

Reference: Smith, D.L. 1997. Impact of composition on the performance of sodium linear alkylbenzenesulfonate (NaLAS). JAOCS 74:837-845.

Reliability: 2 Valid with restrictions

(b)

Value: >250 g/L

Description: Miscible in water

GLP: Yes No ?

Test Substance: Various LASs made from four commercial LABs, average alkyl chain length = 11.6

Remarks: The study shows that 25% solutions (250 g/L) of various LASs have cloud clear points (i.e., form clear solutions at rising temperatures) at temperatures of 2-21°C, depending on LAS composition. Cloud and clear points decrease dramatically with increasing 2-phenyl isomer composition.

The results demonstrate that 25% solutions of LAS are soluble at room temperature.

Reference: Cohen, L., Vergara, R., Moreno, A. and Berna, J.L. 1995. Influence of 2-phenyl alkane and telralin content on solubility and viscosity of linear alkylbenzene sulfonate. *JAOCS* 72:115-122.

Reliability: 2 Valid with restrictions

(c)

Value: ca. 250 g/L

Temperature: 20°C

Description: Miscible in water

GLP: Yes No ?

Test Substance: Marlon A 390 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = 11.6

Remarks: Miscible with water at 20°C. Depending on the concentration, clear solutions (up to ~25% w/w) or inhomogeneous, viscous pastes were obtained. Sources cited are two reports by Huels AG dated 1988 and 1993.

Reference: Cited in IUCLID Data Sheet for CAS #68411-30-3.

Reliability: 4 Not assignable. Original report not available for review.

B. pH Value, pKa Value

(a)

pH Value: 10.0 ± 1.0 (1% solution)

GLP: Yes No ?

Remarks: C₁₀₋₁₄ monoalkylbenzene sulfonic acid, sodium salt (CAS #85117-50-6); mean molecular weight = 348, average alkyl chain length = C_{12.0}

Reference: Huntsman 2002. Report on the melting point analysis for NANSA HS 85/S. Attached technical bulletin dated 04-01-1994. Cover memo from A. Ashworth to K.B. Sellstrom dated April 12, 2002.

Reliability: 4 Not assignable. Original report not available for review.

(b)

pKa Value: <1 for aromatic sulfonic acids such as benzosulfonic acid

Reference: Hodgman, C.D. 1961. Handbook of Chemistry and Physics, 43rd edition. The Chemical Rubber Publishing Company, Cleveland, Ohio.

Reliability: 4 Not assignable. Original data not available for review.

2.7 FLASH POINT (*liquids*)

Remarks: Not applicable.

2.8 AUTO FLAMMABILITY (*solid/gases*)

Remarks: Not applicable.

2.9 FLAMMABILITY

Remarks: Not applicable.

2.10 EXPLOSIVE PROPERTIES

Remarks: Not applicable.

2.11 OXIDISING PROPERTIES

Remarks: Not applicable.

2.12 OXIDATION: REDUCTION POTENTIAL

Remarks: Not applicable.

2.13 ADDITIONAL DATA

A. Henry's law constant

Value: 6.37×10^{-3} Pa m³/mole

Method: Estimation: EPI Suite

GLP: Yes No ?

Test Substance: LAS (CAS #25155-30-0)

Remarks: Calculated using the bond method detailed in Meylan and Howard (1991) and the input parameters resident in the EPI Suite database. Assumes 25°C, molecular weight 348.48. Data reported in EPI Suite as 6.29×10^{-8} atm-m³/mole and converted to Pa m³/mole.

Reference: 1) USEPA. 2000. EPI Suite v3.10.
2) Meylan, W.M. and Howard, P.H. 1991. Bond contribution method for estimating Henry's law constant. Environ. Toxicol. Chem. 10:1283-1293.

Reliability: 2 Valid with restrictions. Standard EPA estimation software.

3. ENVIRONMENTAL FATE AND PATHWAYS

3.1 STABILITY

3.1.1 PHOTODEGRADATION

(a)

Type: Air ; Water ; Soil ; Other

Light source: Sunlight ; Xenon lamp ; Other **Mercury vapor lamp**

Light spectrum: 200-350 nm

Concentration: Initial LAS concentration 60 to 182 mg/l

Temperature: 28°C

Direct photolysis
Degradation: >95 % (weight/weight) after 20 minute (exposure time)

Indirect Photolysis
Degradation: Rapid photodegradation

Method: A series of photodegradation studies were conducted. Aqueous solution of LAS (pH 6.75) were passed through an irradiated tubular flow reactor. Reaction rates were obtained for both non-sensitized conditions and when ferric perchlorate (0.04 to 3.15×10^{-4} g-mole/L) was used as a sensitizer. A Hanovia 1200-watt mercury-vapor lamp was the source of radiation. The LAS concentration was determined by the methylene blue method. Appropriate controls were used.

GLP: Yes No ?

Test substance: LAS; activity: 95% (CAS #25155-30-0)

Remarks: Complete conversion of LAS to intermediates at an average residence time as low as 1 minute. The maximum conversion to CO₂ was obtained at a residence time of 20 minutes and corresponded to 7 moles CO₂ per mole of LAS. Reaction rate increases by two orders of magnitude in presence of ferric perchlorate. Half order kinetics with respect to light intensity and LAS concentration explained the data for nonsensitized conditions. An appropriate rate equation could be derived by assuming a second-order deactivation of light-activated LAS molecules. The sensitized reaction was believed to occur by abstraction of hydrogen atoms from LAS by hydroxyl radicals. Hydroxyl radicals presumably are produced by an electron-transfer reaction involving light-activated ferric ions. The mechanism is complex; over-all kinetics indicated a first-order effect of (Fe⁺³), 1.2 order in light intensity, and maxima in the rate for intermediate LAS and O₂ concentrations.

Reference: Matsuura, T. and Smith, J.M. 1970. Kinetics of photodecomposition of dodecyl benzene sulfonate. Ing. Eng. Chem. Fund. 9:252-260.

Reliability: 2 Valid with restrictions

(b)

Type: Air ; Water ; Soil ; Other

Light source: Sunlight ; Xenon lamp ; Other

Spectrum: >330 nm

Concentration: 50 mg/L

Temperature: 25°C; during photolysis the solution temperature reached 35-40°C.

Indirect Photolysis:
Type of sensitizer: TiO₂ suspension
Degradation: Rapid photodegradation of LAS (<1 to 2 hours)

Method: A study was conducted to determine the photodegradation of LAS in aqueous TiO₂ dispersions. Experiments were carried out with 25 mL solutions containing LAS surfactant with TiO₂. Some experiments used open vessels (37 mL Pyrex glass reaction vessels) under aerobic conditions. Others used vessels sealed with a rubber septum, the solution purged with argon and a fixed volume of oxygen injected. Spectrophotometric analysis was performed at regular intervals.

GLP: Yes No ?

Test substance: LAS (CAS #25155-30-0)

Remarks: The reaction involves fast decomposition of the aromatic ring followed by slower oxidation of the aliphatic chain.

Reference: Hidaka, H., Kubata, H., Gratzel, M., Serpone, N. and Pelizzetti, E. 1985. Photodegradation of surfactants. I. Degradation of sodium dodecyl sulfonate in aqueous semiconductor dispersions. *Nouveau J. Chemie* 9:67-69.

Reliability: 2 Valid with restrictions

(c)

Type: Air ; Water ; Soil ; Other

Light source: Sunlight ; Xenon lamp ; Other **Mercury lamp**

Light spectrum: 400-580 nm

Spectrum: 223 nm

Concentration: 100 mg/L H₂O

Temperature: 20°C

Indirect Photolysis:

Type of sensitizer: Humic substances

Results: Photodegradation of LAS was reduced by humic substances by a factor of 2 or more. The aliphatic side chains are degraded first, followed by aromatic ring cleavages. Degradation follows first order kinetics both with and without the presence of humics.

Method: The effects of humics on the photolytic degradation of LAS was studied. Soil humic substances were extracted by a cationic exchange resin/water suspension from a humic podzol. Water-soluble synthetic humic substances were prepared by autoxidation of pyrogallol in alkaline solution. Aqueous solutions of 15 mg/L humic substance and 100 mg/L LAS were irradiated with a mercury lamp. Photometric measurements were performed with a spectrophotometer for recording the changes caused by photolysis at definite times at 223 nm for LAS.

GLP: Yes No ?

Test substance: LAS (CAS #25155-30-0)

Remarks: The presence of humic substances delays photodegradation of LAS, primarily because they act as UV-absorbers. The reaction between humics and LAS is dominated by electrostatic repulsion because of the negatively charged components at the given pH. The hydrophobic interaction between humics and LAS is relatively weak compared to the electrostatic repulsion. Possibly the sulfonic groups from LAS may be bound by metal bridges to humic surfaces. The study used humic substance with a relatively high proportion of aromatic carbon; whereas a lower proportion is more typical in natural environments. Therefore, the difference in photolysis rate is likely to be less pronounced.

Reference: Hermann, R., Gerke, J. and Ziechmann, W. 1997. Photodegradation of the surfactants LAS and dodecylpyridinium-chloride as affected by humic substances. *Water, Air, and Soil Pollution* 98:45-55.
Reliability: 2 Valid with restrictions

3.1.2 STABILITY IN WATER

Type: Abiotic (hydrolysis) ; biotic (sediment)
Results: LAS is stable in water.
GLP: Yes No ?
Test substance: C₁₀₋₁₃ alkylbenzene sulfonic acid, sodium salt (CAS #68411-30-3)
Remarks: LAS can be decomposed at extreme conditions such as elevated temperatures in the presence of inorganic acids such as phosphoric, sulphuric and hydrochloric acid, e.g.: 60-70% sulphuric acid at 140 - 190 degree C or with concentrated HCl in a sealed container at 150 - 200 degree C. Information as cited in IUCLID Data Sheet for CAS #68411-30-3 and in an analytical textbook.
Reference: Cross, J. and Dekker, M. (ed.). 1977. Anionic surfactants: Chemical analysis. Vol.8. Pp. 111-115.
Reliability: 4 Not assignable. Original studies not available for review.

3.1.3 STABILITY IN SOIL

(a)
Type: Laboratory
Radiolabel: Yes No ?
Concentration: 27.2 mg/kg (Ecosystem Section I) and 16.2 mg/kg (Ecosystem Section II) (initial amounts in dry soil); 0.44 mg/kg (I) and 0.19 (II) (at end of trials)
Temperature: Room temperature
Dissipation Time: DT₅₀ = 13-26 days
Method: Soil cores taken from two ecosystems were collected and placed in a climate controlled "plant metabolism box". Ecosystem Section I consisted of a heavy clay-like soil. Ecosystem Section II consisted of loose, sandy soil. Radiolabeled LAS (a defined mixture) absorbed to digested sludge was incorporated into the soils, after which the soils were planted with either grass, bush beans and radishes (Section I) or potatoes (Section II). The test systems were maintained under a defined standard climate (i.e., an average day in June in Northern Germany) for the vegetative period (76 and 106 days, respectively for Sections I and II). At the end of the growing season samples were collected from plants and soil and subjected to radioanalysis.
GLP: Yes No ?
Test Substance: LAS. The authors state that they tested a defined mixture of LAS, but do not report the composition in this paper.
Remarks: Corresponding to Ecosystem Sections I and II, 63.6% and 72.3% of initial radioactivity went to the atmosphere (primarily as CO₂), 26.8% and 18.3% were detected in soil cores, 6.6% and 5.9% were present in biomass, and 0.99% and 1.4% leached out with percolated water. The study shows that LAS adsorbed to digested sludge is relatively rapidly converted to CO₂ and, to a lesser extent, polar organic secondary products

in the upper soil layers. LAS and the secondary products are strongly adsorbed to the topsoil. LAS introduced into the topsoil by repeated application of sludge did not accumulate in the soil. Growth of crops is not impaired; the use of LAS-containing sludge had no adverse effect on the biomass yield (crop yield) under regulated use conditions.

Reference: Figge, K. and Schoberl, P. 1989. LAS and the application of sewage sludge in agriculture. *Tenside Surf. Det.* 26:122-128.

Reliability: 2 Valid with restrictions

(b)

Type : Field trial ; Laboratory ; Other

Radiolabel: Yes No ?

Concentration: 0.05 mg/kg

Soil temperature: Probably room temperature

Soil humidity: 80% of water holding capacity

Soil classification: DIN19863 ; NF X31-107 ; USDA ; Other mainly U.S. Soil Conservation Service soil type designation, by Howard Laboratories, Dayton, Ohio

Organic Carbon: 1.4 - 50%

Soil pH: 4.9 - 8.4

Cation exchange capacity: 0.15 meq/100 g soil dry weight

Microbial biomass: Activity in dpm/h/gdw soil varied from 11,327 to 51,683

Dissipation time: DT₅₀ : 1.1 - 3.7 days

Method: other: described in reference

GLP: Yes No ?

Test substance: C₁₃ LAS (CAS #26248-24-8); activity 98%

Remarks: Half-lives ranged from 1.1 to 3.7 days (mineralization). First order dissipation rate constants ranged from 0.14 - 0.63/day. Mineralization occurred without a lag-period in every soil tested. Cycles of wetting and drying in the lab prior to testing resulted in more rapid and extensive mineralization. Community microbial activity did not correlate with rate or extent of mineralization. Microbial communities have indigenous ability to degrade low LAS concentrations, the ability is present in a wide array of soil types from various locations.

1. Soil Alpine, sand, pH 5.5, CEC 0.15, TOC 50 (mg/g)
2. Soil Bonnell, sandy loam, pH 8.4, CEC 29, TOC 31.6
3. Soil Brashear, silt loam, pH 7.7, CEC 37, TOC 24.9
4. Soil Eden, silt loam, pH 7.4, CEC 33, TOC 45.1
5. Soil Eden, loamy sand, pH 6.1, CEC 41, TOC 19.6
6. FL soil, sand, pH 4.9, CEC 6.3, TOC 13.8
7. GA soil, loamy sand, pH 4.9, CEC 15.5, TOC 1.4
8. Soil Huntington, loam, pH 7.3, CEC 24, TOC 20.1
9. Soil Lakin, loam, pH 6.5, CEC 24, TOC 34.6
10. Soil Pate, silt loam, pH 7.7, CEC 46, TOC 48.0.

Reference: Knaebel, D.B., Federle, T.W. and Vestal, J.R. 1990. Mineralisation of linear alkylbenzene sulfonate (LAS) and linear alcohol ethoxylate (LAE) in 11 contrasting soils. *Envir. Toxicol. Chem.* 9:981-988.

Reliability: 2 Valid with restrictions

(c)

Type : Field trial ; Laboratory ; Other

Radiolabel: Yes No ?
 Dissipation time: DT₅₀ : 7 - 22 days
 Results: In fields not recently spread with sludge, the concentrations of LAS found in the sludge amended soil were generally less than 1 mg/kg. This represents an estimated loss of LAS from soil of >98%. In fields recently spread, the concentrations in soil are in the range of <0.2 to 20 mg/kg, representing losses of LAS between 70 and 99% of the estimated total cumulative load. The authors conclude that overall the data indicate that an adequate safety margin exists between the concentrations of LAS in sludge-amended soils and those likely to affect the growth of crop plants.
 Method: The disappearance of LAS from sludge-amended soils was investigated from 51 fields on 24 farms in the Thames Water Authority, U.K. Annual sludge spreading averaged 6 ton/ha. Application of sludge was made by subsurface injection, surface spreading onto arable land with or without ploughing, or surface spreading onto pasture land. Regular sampling was conducted for up to 122 days. LAS concentrations in the soil were analyzed with HPLC.
 GLP: Yes No ?
 Test substance: Commercial LAS as present in primary sludge or anaerobically digested sludge from WWTPs in the United Kingdom.
 Remarks: Half-lives compare well with those for ultimate degradation in lab soil tests (with 14-C-evolution), indicating that the degradation of LAS does not lead to the formation of significant levels of break-down intermediates in soil. The homologue distribution of LAS in soil suggests that removal represents biodegradation rather than leaching.
 Reference: 1) Holt, M.S., Matthijs, E. and Waters, J. 1989. The concentrations and fate of linear alkylbenzene sulphonate in sludge amended soils. *Wat. Res.* 23:749-759.
 2) Waters, J., Holt, M.S., Matthijs, E. 1989. Fate of LAS in sludge amended soils. *Tenside Surfactants Detergents* 26(2):129-135.
 Reliability: 2 Valid with restrictions
 (d)
 Type: Laboratory
 Radiolabel: Yes No ?
 Concentration: 8 to 488 mg/kg
 Soil Composition: Coarse sand 67%, fine sand 16%, silt 8.6%, clay 6.2% and humus 2.7%
 Organic Carbon: 1.5%
 Method: LAS mixed with sewage sludge was applied to sandy agricultural soil and incubated for up to 8 weeks. Various microbial soil parameters were measured (see Section 4.4). LAS was quantified after methanol extraction using HPLC.
 GLP: Yes No ?
 Test Substance: C₁₀₋₁₃ LAS obtained as an aqueous sodium salt solution with a LAS content of 16.1% (w/w), NA-LAS average molecular weight = 342 g/mol, distribution: C₁₀ 14%, C₁₁ 34%, C₁₂ 31%, and C₁₃ 21%.
 Results: For nominal concentrations of 8 to 62 mg/kg, the depletion of LAS after 2 weeks was more than 73%. At 488 mg/kg, only 15% depletion occurred. It is possible that this high LAS level may have inhibited microbial activity or caused a prolonged log phase to occur.
 Reference: Elsgaard, L. Petersen, S.O. and Debosz, K. 2001b. Effects and risk assessment of linear alkylbenzene sulfonates in agricultural soil. 2.

Effects on soil microbiology as influenced by sewage sludge and incubation time. Environmental Toxicology and Chemistry. 20:1664-1672.

Reliability: 2 Valid with restrictions

(e)

Type : Field trial ; Laboratory ; Other
Radiolabel: Yes No ?
Soil Content: Clay 1.8 - 4%, Silt 7.6 - 18.5 %, Sand 77.1 - 95.5%
Organic Carbon: Ranged from 0.9 - 1.79%
Soil pH: 5.2 – 6.8
Dissipation time: DT₅₀: 3 days (lysimeters)
DT₅₀: 7 days (field trials)
Method: Sewage sludge containing LAS was added to four cultivated sandy soils with low amounts of organic matter in field trials and lysimeter studies. The field trial lasted one year. For the lysimeter studies, undisturbed soil columns were taken from the corresponding field sites.
GLP: Yes No ?
Test substance: Marlon A350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = 11.6; activity: 50%
Remarks: LAS are mobile in all four soils tested. They were detected down to a depth of 30-40 cm after being applied at the surface. After one week, a concentration of 500 mg/kg was measured in the 0-5 cm layer, which corresponds to 23% of the the total LAS. In the 5-10 cm layer, the concentration was measured at 9.5 mg/kg (approximately 4%). Very little reached the 30-40 cm layer, although a value significantly above the limit of detection (0.050 mg/kg) was determined. No leaching was observed out of the soil columns and disappearance of LAS in the field trials was attributed to rapid biodegradation in the soil (approximately 99% of the total LAS was biodegraded after 42 days). The half-life was determined to be 3 days. There was a noticeable shift to shorter alkyl chain length homologues in the percolating water (i.e., longer alkyl chains were retained more strongly). These shorter chain length homologues have a lower toxicity.
Reference: Kuchler, T. and Schnaak, W. 1997. Behaviour of linear alkylbenzene sulfonates (LAS) in sandy soils with low amounts of organic matter. Chemosphere. 35:153-167.
Reliability: 2 Valid with restrictions

(f)

Type : Field trial ; Laboratory ; Other
Radiolabel: Yes No ?
Concentration: 250 mg/kg
Soil temperature: 20°C
Soil humidity: 35 g water/100g soil dry weight
Soil pH: 6.9 - 7.1
Dissipation time: DT₅₀ : 15.8 - 25.7 day
Method: Assays were conducted in flow-through microcosms with 15 g (dry wt) of soil adjusted to a 35% moisture level. The test substances were ¹⁴C ring labelled LAS pure homologues (C₁₀, C₁₁, C₁₂, C₁₃, C₁₄) which were premixed with digester sludge and added to flasks at an initial concentration of 2.5, 25 or 250 mg/kg. ¹⁴CO₂ evolution was determined

by LSC. The first-order biodegradation rate constant was estimated using non-linear regression techniques. The study was conducted in both sandy loam and loamy sand soil types.

GLP: Yes No ?
Test substance: Pure LAS homologues C₁₀₋₁₄ tested concurrently.
Results: The biodegradation rates followed first order kinetics over a wide range of concentrations and chain lengths. Half-lives (mineralization) were reported as C₁₀ 21 days; C₁₁ 25.7 d; C₁₂ 23.1 d; C₁₃ 18.2 d; C₁₄ 17.8 d in sandy loam; C₁₀ 16.5 d and C₁₄ 15.8 d in loamy sand. The degradation rate constants were C₁₀ 0.033/d; C₁₁ 0.027/d, C₁₂ 0.030/d, C₁₃ 0.038/d, C₁₄ 0.039/d in sandy loam; C₁₀ 0.042/d C₁₄ 0.044/d in loamy sand. Mineralisation efficiency averaged 65%. The remaining radiolabel was incorporated into microbial biomass or soil humic material.
Remarks: Half-lives for mineralization of the benzene ring, the rate-limiting step for LAS degradation, ranged from 18 to 26 days.
Reference: Ward, T.E. and Larson, R.J. 1989. Biodegradation kinetics of LAS in sludge-amended agricultural soils. *Ecotox. and Environ. Safety* 17:119-130.
Reliability: 2 Valid with restrictions

(g)

Type: Field trial ; Laboratory ; Other **Greenhouse pot**
Radiolabel: Yes No ?
Concentration: 3.7 to 5.1 g/kg dry wt
Soil composition: Clay 3.7 %, Silt 3.1 %, Fine Sand 19.5 %, Coarse Sand 71.4 %
Organic Carbon: 1.3%
Soil pH: 5.9
Cation exchange capacity: 10.7 cm/kg
Microbial biomass: not stated
Method: Sewage sludge was incorporated into a sandy soil to give a range of very low to very high applications (0.4 to 90 mg/ha dry weight). LAS was added as water solutions to this mixture. The soil was transferred to pots and sown with barley, rape, or carrot and allowed to grow for 19, 85, and 30 days, respectively in a greenhouse. Plant-free controls were also established. Samples were collected of the soil and analyzed for total LAS and for individual homologues.

GLP: Yes No ?
Test substance: LAS C₁₀₋₁₃, Approximate composition at start of study: C₁₀ 3%, C₁₁ 22%, C₁₂ 40%, C₁₃ 35%
Results: LAS was not taken up by plants and its degradation in soil increased by the presence of crop plants with concentration decreasing in rape from 27 mg/kg (dry soil) to 0.7 - 1.4 mg/kg (dry soil) at harvesting after 30 days. During degradation, the relative fraction of homologues C₁₀, C₁₁, and C₁₂ decreased, while C₁₃ increased.
Reference: Mortensen, G.K., Elsgaard, H., Ambus, P., Jensen, E.S., and Gron, C. 2001. Influence of plant growth on degradation of linear alkylbenzene sulfonate in sludge-amended soil. *J. Environ. Qual.* 30:1266-1270.
Reliability: 2 Valid with restrictions

(h)

Type of Measurement: Background ; At contaminated site ; Other
Medium: Sludge modified soils

Method: LAS biodegradation and its kinetic parameters were studied in a land filling operation using sludges (15%) blended with soil (85%) at a site in Spain. Once the soil was blended with the sludges, grab samples were taken in three different zones of the plot. All samples were blended every day in order to have a daily composite sample. Immediately after sampling, all samples were frozen and then sieved to a 2 mm particle size. The sampling was conducted on 10 different days over a period of 62 days. LAS determination in sludge-modified soils was carried out by HPLC-UV. Degradation was determined based on LAS concentrations measured on the 10 sampling days.

Results: Average LAS concentrations on the 10 sampling days were 155, 55.6, 28.0, 30.2, 35.2, 28.2, 31.6, 35.1, 19.3 and 16.7 for days 0, 6, 15, 20, 27, 34, 41, 48, 55 and 62, respectively. The biodegradation level reached after 62 days was 89.2%. Assuming first order kinetics, the half-life is 19.3 days.

Remarks: LAS adsorbed or precipitated on anaerobic sludges is biodegraded during soil amendment operations by commonly occurring micro organisms. The table shows a shift in the percent homologue distribution on days 0 and 62, demonstrating that higher molecular weight homologues exhibit stronger soil adsorption.

% phenyl homologue	Day 0	Day 62
C ₁₀	3.7	1.2
C ₁₁	29.4	18.7
C ₁₂	41.4	48.9
C ₁₃	25.5	31.2

Reference: de Ferrer, J., Moreno, A., Vaquero, M.T. and Comellas, L. 1997. Monitoring of LAS in direct discharge situations. *Tenside Surf. Det.* 34:278-283.

Reliability: 2 Valid with restrictions

3.2 MONITORING DATA (ENVIRONMENTAL)

(a)
Type of Measurements: Background []; At contaminated site []; Other [X] **Mississippi River**
Medium: Surface Water
Results: LAS was detected in 16% of the 323 mainstem samples collected during the upstream sampling cruises at concentrations ranging from 0.1 to 10.3 µg/L, and in 15% of the 39 tributary samples at concentrations ranging from 0.1 to 2.8 µg/L. LAS was detected in 21% of the 38 mainstem composite samples collected during the downstream cruises at concentrations ranging from 0.1 to 2.8 µg/L and was not detected in any of the 16 tributary composite samples. LAS was detected in 85% of the 34 samples collected from the Thebes time-series site at concentrations ranging from 0.4 to 28.2 µg/L.

Remarks: The 2,800 km reach of the Mississippi River between Minneapolis and New Orleans was examined for the occurrence of LAS. River water was sampled in the summer and fall of 1991 and in the spring of 1992 during upstream and downstream sampling cruises. LAS was analyzed using solid-phase extraction and gas chromatography/mass spectrophotometry.

The range of average chain length for all dissolved LAS was 10.2-12.0, with an average of 11.1. The removal of the higher LAS homologues and external isomers indicates that sorption and biodegradation are the principle processes affecting dissolved LAS.

Reference: Tabor, C.F. and Barber, L.B. 1996. Fate of linear alkylbenzene sulfonate in the Mississippi River. Environ. Sci. Technol. 30:161-171.

Reliability: 2 Valid with restrictions

(b)

Type of Measurements: Background []; At contaminated site []; Other [X] **Mississippi**

Medium: Sediment

Results: LAS was present on all bottom sediments (33 locations) at concentrations ranging from 0.01 to 20 mg/kg dry matter. It should be noted that all concentrations were <0.1 mg/kg with the exception of the one extremely high level of 20 mg/kg. The 20 mg/kg sample was found at Pig's Eye Slough, the canal carrying the Minneapolis STP effluent to the the Mississippi River. All concentrations are dry weight.

Remarks: The 2,800 km reach of the Mississippi River between Minneapolis and New Orleans was examined for the occurrence of LAS. Bottom sediment was sampled in the summer and fall of 1991 and in the spring of 1992. Composite samples were collected at 25 locations during the downstream leg of each cruise. These samples consisted of 5-7 individual sediment samples on each of 2-3 transects in each pool. In addition, grab samples were taken at 8 other locations during the downstream cruise. LAS was analyzed using solid-phase extraction and gas chromatography/mass spectrophotometry. The average chain length for sorbed LAS ranged from 10.7 to 12.5, with an average of 11.5. Sorbed LAS appears to degrade slowly.

Reference: Tabor, C.F. and Barber, L.B. 1996. Fate of linear alkylbenzene sulfonate in the Mississippi River. Environ. Sci. Technol. 30:161-171.

Reliability: 2 Valid with restrictions

(c)

Type of Measurements: Background []; At contaminated site []; Other [X] **Rivers in USA**

Medium: Influent, effluents, and surface water

Results: Average LAS influent concentrations ranged from 4.2-5.7 mg/L among the various types of treatment plants. LAS removal rates averaged 99.3% for activated sludge (n = 15), 98.0-98.5% for lagoon/oxidation ditch (n = 14), 96.2% for rotating biological contact (n = 9) and 77.4% for trickling filters (n = 12). Concentrations of LAS below the mixing zone of wastewater treatment plants were generally below 50 µg/L, even though the samples were collected under low flow (i.e., low dilution) conditions. The mean surface water concentrations ranged from <10 to 330 µg/L, with mean values of 42 to 46 µg/L. The highest concentration was observed in a low (less than 3-fold) dilution irrigation canal below a trickling filter plant. All other values were <180 µg/L, with more than 80% of the sites below 50 µg/L.

Remarks: Surface water samples were collected in rivers at 50 locations in 11 states below wastewater treatment plants. Alkyl chain lengths of LAS averaged 12.0 carbon units in most environmental compartments, with the exception of sludge solids and river sediments, in which an enrichment of longer chain lengths was observed. Since several of the wastewater treatment

plants included in this study have dilution factors less than 3, these values include worst case estimates.

Reference: McAvoy, D.C., Eckhoff, W.S. and Rapaport, R.A. 1993. Fate of linear alkylbenzene sulfonate in the environment. Environ. Toxicol. Chem. 12:977-987.

Reliability: 2 Valid with restrictions

(d)

Type of Measurement: Background []; At contaminated site []; Other [X] **Rivers in U.S.A.**

Medium: sediment

Results: Below outfall of trickling filter treatment plant: 190 ± 95 mg/kg dry matter
< 5 miles downstream: 11.9 ± 9.5 mg/kg dry matter;
> 5 miles downstream: 5.3 ± 4.7 mg/kg dry matter

Remarks: Monitoring studies for LAS in river sediment in Rapid Creek, USA below the outfall of a trickling filter sewage treatment plant. Compiled by Procter & Gamble between 1973 and 1986.

Reference: Rapaport, R.A. and Eckhoff, W.S. 1990. Monitoring linear alkylbenzene sulfonate in the environment: 1973-1986. Environ. Toxicol. Chem. 9:1245-1257.

Reliability: 2 Valid with restrictions

(e)

Type of Measurement: Background [X]; At contaminated site []; Other [X]

Medium: Sewage treatment influents & effluents; rivers and sediments.

Results: The removal from four activated sludge and five trickling filter wastewater treatment facilities averaged 99.5% & 82.9% for LAS and 99.1% and 97.3% for LAS intermediate, respectively, for the activated sludge and trickling filter facilities.

Remarks: LAS concentrations in receiving waters downstream of four activated sludge treatment plants ranged from 0.002 to 0.081 mg/L. LAS concentrations in receiving waters downstream of five trickling filter treatment plants ranged from 0.004 to 0.094 mg/L. Upstream LAS concentrations ranged from <0.001 to 0.110 mg/L and <0.001 to 0.005 mg/L for the activated sludge and trickling filter treatment plants, respectively.

Reference: Trehy, M.L., Gledhill, W.E., Mieux, J.P., Adamove, J.E., Nielsen, A.M., Perkins, H.O. and Eckhoff, W.S. 1996. Environmental monitoring for linear alkylbenzene sulfonates, dialkyltetralin sulfonates and their biodegradation intermediates. Environmental Toxicology and Chemistry 15:233-240.

Reliability: 2 Valid with restrictions

(f)

Type of Measurement: Background []; At contaminated site []; Other [X] **concentrations in sludge from waste water treatment plants**

Medium: sludge

Results: (A) Conc. In sludge 0.7 and 0.4 g LAS/kg of dry matter (data from 2 plants)
(B) Conc. In sludge 0.5 and 0.1 g LAS/kg of dry matter (data from 2 plants)
(C) Conc. In sludge 30.2; 12.8; 11.4; 7.0 and 7.5 g LAS/kg of dry matter (from 5 plants)

Remarks: (A) Plants with aeration/settling system

(B) Plants with activated sludge & aerobic digestion of sludges system

(C) Plants with activated sludge & anaerobic digestion of sludges.

The concentrations in sludge correlate with water hardness. The higher the water hardness, the greater the amount of calcium-precipitated LAS in the sludge. For example, the water hardness that resulted in the 30.2 g/kg value was >500 mg/L as calcium carbonate. This level of hardness is very high and is 2-3 times higher than the more typical range of 200-300 mg/L as calcium carbonate. The amount of LAS physically removed to the sludge during primary settling versus biodegraded can be seen in the following table.

Treatment Plant (all from category C)	Physical Removal	LAS Biodegradation	Water Hardness (as mg/L CaCO₃)
Alicante	35%	68.2%	>500
Sevilla S.E.	30%	75.6%	220
Sevilla N.	27%	87%	315
La China (Madrid)	16%	91%	<100
Viveros (Madrid)	15%	91.2%	<100

Reference: Berna, J.L., de Ferrer, J., Moreno, A., Prats, D., Ruiz Bevia, F. 1989. The fate of LAS in the environment. *Tenside Surfactants Detergents* 26(2):101-107.
 Reliability: 2 Valid with restrictions

(g)

Type of Measurement: Background []; At contaminated site []; Other [X] **STP effluent in the UK**

Medium: Final effluent

Results: Final effluent concentration of LAS from four trickling filter STPs ranged from 40 to 430 µg/L (mean 80 to 300 µg/L).

Remarks: Removal from trickling filter STPs averaged 92.9%

Reference: Holt, M.S., Daniel, M., Buckland, H. and Fox, K.K. 2000. Monitoring studies in the UK designed for validation of the Geo-Referenced Exposure Assessment Tool for European Rivers (GREAT-ER), 5th World CESIO Congress. V.2:1358-1369, Firenze, Italy.

Reliability: 2 Valid with restrictions

(h)

Type of Measurement: Background []; At contaminated site []; Other [X] **Lambro River (Italy)**

Results: Mean background LAS concentration downstream from the STP was 28 µg/L.

Remarks: A two year water quality monitoring program was conducted in the river Lambro (northern Italy) during the period March 1997 to May 1998. Prior to 1998, 40% of the local waste water was discharged untreated directly into the river. The plants were undersized activated sludge plants. From April to September 1997, grab samples were collected approximately once a month from 19 stations along the main channel of the river, its two main tributaries, and at two STPs located within the monitoring area. From November 1997 to May 1998, the sampling protocol was modified and grab samples were replaced by 24-hour composite samples (one sample shot every 20 minutes) collected using automatic samplers, twice a month

at four sites downstream of the Merone plant, at the STP overflow, and occasionally one site upstream of Merone. Additional studies were also performed in conjunction with the overall monitoring program.

Reference: Gandolfi, C., Facchi, A., Whelan, M.J., Cassarri, G., Tartari, G. and Marcomini, A. 2000. Validation of the GREAT-ER model in the River Lambro catchment. 5th World CESIO Congress. V.2:1370-1379.

Reliability: 2 Valid with restrictions

(i)

Type of Measurement: Background []; At contaminated site []; Other [X] **Tiber River**

Medium: Sewage treatment plant activated sludge and surface water

Results: The average LAS concentrations in the Roma Nord activated sludge sewage treatment plant were 4.6 mg/L (influent), 0.068 mg/L (effluent), and 6000 mg/kg dry matter (final sludge), which correspond to an overall 98.5% LAS removal rate in the plant. In the receiving waters of the Tiber River LAS was 9.7 µg/L in the aqueous phase and 1.8 mg/kg dry matter in the sediment. Total daily LAS entering the treatment plant amount to 1150 kg. About 1.5% (17 kg) leave the treatment plant through the final effluent and 234 kg (about 20% of the influent LAS) are removed by the digested sludge.

Remarks: Samples were collected over several days in June 1993. It is important to note that LAS environmental fingerprints in effluent and surface waters differ from the composition of the commercial material. The relative ratio of the various homologues detected in the aquatic environmental samples is as follows: C₁₀:C₁₁:C₁₂:C₁₃ = 45:30:23:2 with an average carbon number of 10.8. That is due to the alkyl chain switch to shorter homologues in water as a consequence of both biodegradation in the water phase, which is faster for the higher homologues, and of adsorption into sediments, suspended solids, and the sludge, which is more pronounced for higher homologues. The LAS homologue distribution in sludge is approximately in the mole ratio C₁₀:C₁₁:C₁₂:C₁₃ = 7:24:39:30 with an average carbon number of 11.9, as a consequence of a preferential adsorption of higher homologues.

Reference: DiCorcia, A., Samperi, R., Belloni, A., Marcomini, A., Zanette, M., Lemr, K. and Cavalli, L. 1994. LAS pilot study at the "Roma-Nord" sewage treatment plant and in the Tiber river. *La Rivista Italiana Delle Sostanze Grasse*. LXXI:467-475.

Reliability: 2 Valid with restrictions

(j)

Type of Measurement: Background []; At contaminated site []; Other [X] **Rivers in five European Countries**

Medium: Sewage treatment plant activated sludge, river surface water, sediment

Results: A very high average LAS total removal of 99.2% (98.5-99.9%) in sewage treatment was found. LAS concentrations in river water below activated sludge treatment plants in five European countries ranged from <2.1 to 47 µg/L.

Method: As part of a pilot study, LAS monitoring was conducted at five activated sludge sewage treatment plants located in Germany, the UK, the Netherlands, Spain and Italy. Samples were collected over 7-day monitoring periods. Daily flow-related composites of raw sewage and treated effluent were taken. On at least one day, samples were taken at 2

or 3 hour intervals to assess diurnal variations in LAS concentration. Sludge samples were also taken. Samples of river water and sediment were also collected from sites above and below the sewage effluent outfalls. Samples consisted of either grab or composite samples, as well as interval sampling to determine diurnal changes in LAS concentration. Sediment samples were collected from the top layer (0-5 cm) of the river bed in four of 5 pilot study locations. LAS determinations were made with validated trace enrichment-HPLC procedures employing a specific fluorescence detector.

Remarks: LAS concentrations in raw sewage ranged from 4.0 to 15.1 mg/L. Only low concentrations of LAS were discharged to the receiving waters. The range of mean effluent concentrations was 0.009-0.140 mg/L. The mean concentration of LAS in river sediments below effluent discharges ranged from 0.49-5.3 µg/g. Below treatment plants, LAS levels in sediments were very similar (and sometimes lower) than levels above treatment plants. Based on these observations, the authors suggest that LAS is bioeliminated in river sediments. LAS levels in digested sludge from Spain and Italy ranged from 6.0 to 9.4 g/kg dry weight. Differences in the main operating characteristics at the five sites (e.g., treatment type, plant size, sludge retention time, hydraulic retention time, temperature) were not found to greatly influence the removal of LAS.

Reference: Waters, J. and Feijtel, T.C.J. 1995. AIS/CESIO Environmental Surfactant monitoring programme: Outcome of five national pilot studies on linear alkylbenzene sulphonate (LAS). *Chemosphere* 30:1939-1956.

Reliability: 2 Valid with restrictions

(k)

Type of Measurement: Background []; At contaminated site []; Other [X] **Red Beck, a small Yorkshire stream**

Medium: Sewage treatment effluents

Results: The results show an LAS concentration, corrected for dilution, of 0.07 mg/L (uncorrected 0.033 mg/L) at Sunny Bank (the furthest downstream station, approximately 4.8 km from the out fall) after 6 hours of travel time. The calculated half-life was in the 2-3 h range, indicating kinetics faster than that of laboratory biodegradation studies in river waters.

Method: LAS and water quality parameters have been measured at seven sites downstream of the effluent discharge point of a trickling filter treatment plant (Shibden Head Sewage Treatment Works, Yorkshire, UK). This study was carried out specifically to measure in-stream removal kinetics of LAS. Time of travel was measured by detection of a fluorescent dye (Rhodamine WT) added to the effluent. Increase in flow as the river proceeds through the catchment was determined by flow measurements and boron dilution rate. Nine sampling stations were selected. The study began with the injection of Rhodamine WT dye to the final effluent. The concentration profile of the dye pulse was established from plots of fluorescence intensity versus time, which allowed the measurement of LAS concentration in the same stream volumes of water as the flow moved downstream. LAS concentrations were corrected for increased flow of the stream (and dilution of LAS) by inputs from side streams. Water samples were collected using automatic samples (time proportional or time proportional centroid composites) and/or grab samples and analyzed for water quality parameters and LAS. Boron was

used as a reference substance for measuring the increasing stream flow as boron is highly soluble in water and non-degradable. LAS was analyzed as per the method of Holt et al. 1995. Briefly, LAS was extracted from the samples by solid phase extraction on C18 cartridges, eluted with methanol, evaporated under nitrogen to dryness, reconstituted in 1 mL methanol, and analyzed by reverse phase HPLC on a C18 column with fluorescence detection.

Remarks: The study indicates that an LAS removal half-life of 2-3 hours will be appropriate for small shallow streams, which have an LAS concentration between 50-250 µg/L, for use in the GREAT-ER model calibration exercise.

Reference: Fox, K., Holt, M., Daniel, M., Buckland, H., and Guymmer, I. 2000. Removal of linear alkylbenzene sulfonate from a small Yorkshire stream. Contribution to GREAT-ER project #7. *Sci. Total Environ.* 251:265-275.

Reliability: 2 Valid with restrictions

(l)

Type of Measurement: Background []; At contaminated site []; Other [X] **Dutch surface water**

Medium: Surface water downstream of activated sludge STP outfalls just after the mixing zone.

Results: The mean LAS concentration in surface waters just downstream of the mixing zone was 14.2 µg/L, with a range mostly between <2 to 47 µg/L.

Remarks: Mean derived from a total of 23 records taken from the joint NVZ/VROM monitoring program of sewage treatment plants around the Netherlands. Samples were collected during three consecutive days from seven different sewage treatment plants.

Reference: 1) Feijtel, T.C.J. and van de Plassche, E.J. 1995. Environmental Risk Characterization of 4 Major Surfactants used in the Netherlands. RIVM Report No. 679101 025.
2) Matthijs, E., Holt, M.S., Kiewist, A., and Rijs, G.B. 1999. Environmental monitoring for LAS, AE, AES, AS, and soap. *Environ. Toxicol. Chem.* 18:2634-2644.

Reliability: 2 Valid with restrictions

(m)

Type of Measurement: Background []; At contaminated site []; Other [X] **Rivers in Japan**

Medium: Surface water

Results: Measured LAS concentrations from March 1998 to September 2002 ranged from below the detection limit (< 4 µg/L) to 81µg/L. The 95th percentile values ranged from below detection (< 4 µg/L) to 48.7 µg/L. The following table shows the maximum, median and 95th percentile LAS concentrations along with the number of samples in which LAS was detected.

Site #	# Data Points	LAS Concentration (µg/L)		
		Maximum	Median	95 th Percentile
1	18	7.0	<4.0	7.0
2	2	12.0	8.0	11.6

3	18	24.0	9.5	16.4
4	18	50.0	6.0	44.1
5	18	81.0	9.5	48.7
6	2	<4.0	<4.0	<4.0
7	14	17.0	5.0	13.8
All Sites	90	81.0	6.0	33.1

Methods: River water samples were collected from seven sites on four urban rivers in Japan (Tamagawa, Edogawa, Arakawa, and Yodogawa Rivers), as summarized in the following table.

Site #	River Name	Site Name	Water Area Category	Description	BOD in 1999 Median/75 th percentile (mg/L)
1	Tamagawa	Hamura-seki	A	Upstream. Drinking water intake site.	0.5/0.5
2	Tamagawa	Harabashi	B	Midstream. Just below municipal wastewater treatment plant effluent discharge	2.1/2.3
3	Tamagawa	Denen-chofu-seki	B	Midstream	1.6/1.7
4	Edogawa	Kanamachi	A	Downstream. Drinking water intake site	1.4/1.7
5	Arakawa	Chisui-bashi	B	Midstream	4.6/5.5
6	Arakawa	Sasame-bashi	C	Downstream. Just below municipal wastewater treatment plant effluent discharge	2.5/3.1
7	Yodogawa	Hirakata-oohashi	B	Midstream. Drinking water intake site	1.6/1.9

Grab samples were collected four times a year (summer, autumn, winter, and spring) at each sampling location. River water was characterized at each sampling occasion for BOD, TOC, SS, pH, Cl, NH₄ and MBAS. LAS was complexed with MBAS, extracted, passed through a cation-exchange column, and the concentration of C₁₀₋₁₃ LAS measured using HPLC. Populations in the catchments of the four rivers are relatively dense and municipal wastewater treatment coverage rates are middle to high (i.e., from 60-70% to over 90% coverage). The seven sites cover upstream (Site 1), midstream (2, 3, 5, 7), and downstream (4, 6), and water area categories ranging from A to C. Two of the sites (2, 6) are just below municipal wastewater treatment plant effluent discharges. Three sites (1, 4, 7) are near drinking water intake sites.

Reference: Nishiyama, N., Yamamoto, A., and Takei, T. 2003. 37th Annual meeting of the Japan Society of Water Environment, Kumamoto, Japan. Japan Soap and Detergent Association (JSDA). Annual reports of environmental issues (Years 1999, 2000, 2001, 2002).

Reliability: 2 Valid with restrictions

(n)

Type of Measurement: Background []; At contaminated site []; Other [X] **Europe STP sludge**
Medium: Anaerobic sludge

Results: Typical values are between 3.4 and 9.4 g/kg dry matter, with a mean of 5.6 g/kg.

Remarks: Mean based on a total of 16 records. The LAS homologue distribution in sludge is approximately in the mole ratio C₁₀:C₁₁:C₁₂:C₁₃ = 7:24:39:30,

with an average carbon number of 11.9, as a consequence of a preferential adsorption of higher homologues. The possible differences of LAS concentration in wet sludge, freshly produced at a STP from that of dry sludge, aged and dried before its use in agriculture, should be taken into account. It was found (Carlsen et al. 2002) that the LAS concentration in the bulk dry sludge could drop up to 74% compared to that of the wet sludge. Carlsen et al. (2002) also found that the average LAS concentration in soil core samples taken in a cultivated field spread with medium amounts of sludge was 1.12 mg/kg in the 0-10 cm depth. Lower LAS concentrations were recorded at depths from 10-50 cm.

Reference:	1) Carlsen, L., Metzton, M.B. and Kjelsmark, J. 2002. Linear alkylbenzene sulfonates (LAS) in the terrestrial environment. <i>The Science of the Total Environment</i> . 290:225-230. 2) Feijtel, T.C.J., Matthijs, E., Rottiers, A., Rijs, G.B.J., Kiewiet, A. and de Nijs, A. 1995. AIS/CESIO environmental surfactant monitoring program. Part 1: LAS monitoring study in “de Meer” STP and receiving river “Leidsche Rijn”. <i>Chemosphere</i> . 30:1053-1066. 3) Holt, M.S., Water, J., Comber, M.H.I., Armitage, R., Morris, G. and Nebery, C. 1995. AIS/CESIO environmental surfactant monitoring programme. SDIA sewage treatment pilot study on LAS. <i>Wat. Res.</i> 29:2063-2070. 4) Sanchez Leal, J., Garcia, M.T., Tomas, R., de Ferrer, J. and Bengoechea, C. 1994. Linear alkylbenzene sulfonate removal. <i>Tenside Surf. Det.</i> 31:253-256.
Reliability:	2 Valid with restrictions
(o)	
Type of Measurement:	Background [<input type="checkbox"/>]; At contaminated site [<input type="checkbox"/>]; Other [X] concentrations in sludge-amended agricultural soils
Medium:	soil
Method:	An extensive monitoring study was performed on sludge-amended soils in the Thames Water Authority area, UK, by a Soap and Detergent Industry Association task force. A total of 51 fields from 24 farms were used, with sites representing a range of soil types, frequency and level of sludge applications, and agricultural uses. A total of 35 of the fields were pasture lands and 16 fields were arable land. The majority of fields were surface spread with primary and anaerobically digested sludge containing approximately 4.5% dry solids.
Results:	Forty two farm sites that had received their most recent sludge application prior to 1987 (the study year) were monitored to establish the levels of LAS in sludge amended soils. Thirty five (83%) of the samples contained LAS levels below 1 mg/kg soil (mean = 0.7 mg/kg). The seven other sites contained LAS levels between 1.1 and 2.5 mg/kg soil. Nine sites received sludge applications in January to May 1987. Sampling was conducted in May 1987 and resulted in LAS concentrations ranging from <0.2 to 19.8 mg/kg. The highest LAS concentrations in soil (19.8, 10.6, 7.8, and 4.5 mg/kg) were recorded within days after the sludge application.
Remarks:	Soil concentrations dropped significantly in less than two months (e.g., the 19.8 mg/kg value dropped to 2.1 mg/kg in 55 days). The average chain length of the LAS found in soil samples was C _{11.7} .
Reference:	Waters, J., Holt, M.S., Matthijs, E. 1989. Fate of LAS in sludge amended soils. <i>Tenside Surfactants Detergents</i> 26(2):129-135.

Reliability: 2 Valid with restrictions

(p)

Type of Measurement: Background []; At contaminated site []; Other [X] **concentrations in sludge-amended agricultural soils**

Medium: sludge and soil

Method: This paper collects the literature on the concentrations of LAS in sludge-amended soil resulting from the use of sewage sludge applications on agricultural fields. Data were compiled from a variety of original reference sources.

Results: The concentrations of LAS in sludge are shown in the following table:

Sludge Description	Concentration (mg/kg dw)	Reference *
Anaerobic	2,900 - 11,900	McEvoy and Giger 1985
Anaerobic	4,660 - 1,540	Rapaport et al. 1987
Aerobic	182 - 432	Matthijs and De Henau 1987
Anaerobic	1,330 - 9,930	Matthijs and De Henau 1987
Anaerobic	5,500	Marcomini 1988
Aerobic	100 - 500	Berna et al. 1989
Anaerobic	7,000 - 30,200	Berna et al. 1989
Aerobic	152 ± 120	McAvoy et al. 1993
Anaerobic	10,460 ± 5,170	McAvoy et al. 1993
Anaerobic	11,500 - 14,000	Cavalli et al. 1993
Anaerobic	12,100 - 17,800	Prats et al. 1993
Anaerobic	6,000 ± 1,200	Di Corcia et al. 1994
Primary	3,400 - 5,930	Feijtel et al. 1995
Aerobic	205	Feijtel et al. 1995
Aerobic	11 - <500	VKI 1997
Anaerobic	1,000 - 16,100	VKI 1997

* See article for full citations.

The concentrations of LAS in sludge-amended soil are shown in the following table.

Initial Concentration (mg/kg dw)	Typical Concentration * (mg/kg dw)	Reference**
16, 27	--	Figge et al. 1989
45	5	Marcomini et al. 1989
16,53	0.3	Berna et al. 1989
Max. 66	0-20	Waters et al. 1989
Max. 145	0-8	Holt et al. 1992
Max. 250	1-7	Ward et al. 1989
22.4	0.7, 3.1	Prats et al. 1993

* Typical values after a test period

** See article for full citations

Remarks: The range of LAS concentrations in sludge rarely exceeds 30,000 mg/kg dw. The range of LAS concentrations in sludge-amended soil also is low.

Reference: Cavalli, L., and Valtorta, L. Surfactants in sludge-amended soil. Tenside Surfactants Detergents 36:22-28.

Reliability: 2 Valid with restrictions

(q)

Type of Measurement: Background []; At contaminated site []; Other [X] **Coastal waters and harbor sediments with municipal and industrial discharge**

Medium: marine sediments

Test Substance: The chemical standard was a commercial LAS with a low dialkyltetralinsulfonates content (<0.5%) in a single standard mixture with proportional composition of the homologues C₁₀ 3.9%, C₁₁ 37.4%, C₁₂ 35.4% and C₁₃ 23.1% (Petroquímica Española S.A.)

Method: Sediment samples were taken from 23 sites along the Mediterranean coast of Spain and from three sites on the Atlantic coast. Water samples were taken at 14 sites on the Mediterranean coast. Samples underwent standard sample preparation and were analyzed using HPLC/MS. Samples were analyzed for individual LAS homologues (C₁₀, C₁₁, C₁₂, C₁₃) and other nonionic surfactants and their degradation products.

Results: In seawater the concentrations of LAS across all the samples ranged from 2.4 to 92 µg/L and in marine sediment the concentrations of total LAS ranged from 0.1 to 238 mg/kg dry weight. Concentrations were higher in the sediments than in the water column, with the highest concentrations found in sediments collected in the proximity to the outflow of untreated urban wastewaters. The average carbon chain length of LAS ranged from 10.6 to 11.6 in water and 12.0 to 12.8 in sediment.

Remarks: High concentrations of nonionic surfactants and their degradation products have been shown to accumulate in sediments, which seem to act as a sink for LAS in studied areas. The relative concentrations of the lower homologues C₁₀ and C₁₁ LAS in water samples are higher than the typical laundry detergent, mainly due to the partial removal and/or enrichment of these species during transportation of the wastewater in the sewage system because of the higher degradation and adsorption tendency of the longer alkyl chain homologues. The longer chain lengths are preferentially sorbed to particulate matter because of their high lipophilicity, thus explaining the increase in relative concentration of C₁₂ and C₁₃ homologues in the sediment samples. When interpreting this study, it is important to note that hot spots such as described are not representative of European coastal sediments. Little or no macrofauna lives in such sediment, probably due to multistressor pressure.

Reference: Petrovic, M., Fernandez-Alba, A.R., Borrull, F., Marce, R.M., Mazo, E.G., and Barcelo, D. 2002. Occurrence and distribution of nonionic surfactants, their degradation products, and linear alkylbenzene sulfonates in coastal waters and sediments in Spain. *Environmental Toxicology and Chemistry* 21:37-46.

Reliability: 2 Valid with restrictions

(r)

Type of Measurement: Background []; At contaminated site []; Other [X] **Coastal sediments in Bay of Cadiz, Spain**

Medium: Estuarine and marine sediments

Method: Sediment samples were collected from seven stations (5 in the Bay of Cadiz, 2 in the Barbate River), representing a range of low, moderate, and high levels of chemical contamination. Samples were collected using a 0.025m² Van Veen grab sampler during winter and summer in the same year. LAS was measured using specific HPLC analytical techniques. Fourteen heavy metal contaminants were also measured. Concurrent

sediment toxicity tests were also conducted in which the rate of burial for clams (*Ruditapes philippinarum*) was measured over 48 hours and the survival of amphipods (*Microdeutopus gryllotalpa*) was measured over 10 days of exposure to whole sediments.

Results: LAS concentrations in the sediment ranged from 1.2-26.7 mg/kg dw in the summer and 1.2-62.1 mg/kg dw in the winter. Five of the 7 stations had LAS concentrations < 2.6 mg/kg dw.

Remarks: No mortality was observed in the clam toxicity studies. Clam burial was fastest in the uncontaminated sites (e.g., ET_{50} = 0.01-0.76 hours in winter), intermediate in moderately polluted sites (e.g., ET_{50} = 0.92-1.29 hours in winter), and slowest in highly polluted sites (e.g., ET_{50} > 48 hours in winter). The highest survival in the amphipod studies was in the uncontaminated site (85% survival) and the lowest survival was in the highly polluted site (16% survival). LAS is the only organic component of untreated sewage discharges considered, and sediment contaminants also included high levels of Ag and Pb.

Reference: DelValls, T.A., Forja, J.M. and Gomez-Parra, A. 2002. Seasonality of contamination, toxicity, and quality values in sediments from littoral ecosystems in the Gulf of Cadiz (SW Spain). *Chemosphere*. 46:1033-1043.

Reliability: 2 Valid with restrictions

(s)

Type of Measurement: Background []; At contaminated site []; Other[X]; **Coastal sediments in Denmark**

Medium: marine sediments

Method: Two core samples of 1 meter length were taken in the Baltic Sea, one in the inner Stockholm archipelago, and one north of Gotland, in the autumn of 2000. Sediment samples were also taken from five locations in Haderslev Fiord on December 20, 2000. On April 3, 2001, samples were taken from five locations each in Vejle and Kolding Fiords. All samples were analyzed for total alkylbenzene sulfonates (LAS), and if possible, branched (branched dodecylbenzene sulfonates (BDS)) and linear alkylbenzene sulfonates separately. In addition, soap and volatile solids were analyzed separately.

Results: Danish marine sediments are not generally contaminated with LAS. The levels of health sediments are near or below detection limits. The concentration of LAS in soft sediments of the Baltic Sea along the Swedish coast is <0.5-1 mg/kg dry wt. Soap can be detected in high concentrations (1,000-2,000 mg/kg dry wt.) in sediments, where LAS could not be detected (<0.05 mg/kg dry wt.). Sediments from a Danish shipping port (Haderslev Fiord), as previously reported by Danish EPA, were found to contain relatively high concentrations of LAS (2-20 mg/kg dry wt.) as well as extremely high soap concentrations (3,000-10,000 mg/kg dry wt.) and many other pollutants.

Remarks: The authors conclude that the environmental problem in Haderslev Fiord is not LAS or soap, but the fact that the whole sediment consists of stinking sludge formed by past discharges of untreated sewage and today's overflow of sewage from emergency spillways in the municipal sewage system. The presence of BDS in the sediments demonstrates that this a historical problem.

Reference: Folke, J., Cassani, G., de Ferrer, J., Lopez, I., Karlsson, M.O., and Willumsen, B. 2003. Linear alkylbenzene sulphonates, branched dodecylbenzene sulfonates and soap analyzed in marine sediments from the Baltic proper and Little Belt. *Tenside Surf. Det.* 40:17-24.

Reliability: 2 Valid with restrictions

(t)

Type of Measurement: Background []; At contaminated site []; Other [X] **Coastal sediments receiving untreated urban effluents**

Medium: marine sediments

Test Substance: LAS C₁₀₋₁₄ (sum of all homologues)

Method: There were three objectives to the study: 1) determine LAS levels in coastal sediments in areas receiving discharges of untreated urban effluents; 2) determine LAS distribution between the solid phase and the interstitial water; and 3) determine the presence of SPCs in the sediment column at depths below the aerobic oxidation/reduction interface. The study was carried out in a salt marsh in the south part of the Bay of Cadiz in the southwest of Spain. Samples were taken at three stations in an area where LAS levels are very high due to untreated urban effluents. Ten cores of sediment were collected at each station, frozen, cut into 1 cm thick sections and analyzed using standard HPLC/FL preparation, analysis, and cleanup techniques. Analyses were conducted to determine both LAS concentrations and concentrations of long-chain sulfophenyl carboxylic acids (SPCs) resulting from LAS biodegradation.

Results: The vertical profile of LAS concentrations in the sediment and interstitial waters showed a sharp reduction with depth, whereas the long chain SPCs (6-13 carbon atoms) was greatest at 10-14 cm depth where the interstitial water becomes anoxic. Surface (0-8 cm) sediment concentrations (dry weight) for total LAS and SPCs are shown below.

Station	Location	Total LAS (mg/kg)	Total SPCs (µg/kg)
B	Close to discharge point	138.6	924.6
C	Distant, strong tidal current	16.4	224.2
A	Distant, weaker tidal current	0.8	70.6

The partition coefficients between the solid phase of the sediment versus the interstitial water are very different for LAS and for its degradation intermediates. For LAS, the organic carbon-based partition coefficient values were between 2.4×10^3 and 6.6×10^5 L/kg for the homologues C₁₀ and C₁₃, respectively. For the longer chain SPCs, the partition coefficients are several orders of magnitude lower as a consequence as their lower hydrophobicity.

Remarks: The LAS concentration in the upper sediment layer (0-8 cm) decreased with distance from the point of effluent discharge. The concentration of LAS in the sediment was up to 1000 times greater than that in the interstitial water. For SPC, the concentrations in the sediment and interstitial water were similar to each other.

Reference: Leon, V.M., Gonzalez-Mazo, E., Pajares, J.M.F., and Gomez-Parra, A. 2001. Vertical distribution profiles of linear alkylbenzene sulfonates and their long-chain intermediate degradation products in coastal marine sediments. *Environmental Toxicology and Chemistry* 20:2171-2178.

Reliability: 2 Valid with restrictions

(u)

Type of Measurement: Background []; At contaminated site []; Other [X] **Predicted marine water concentrations in the North Sea**

Medium: Estuarine and marine surface water

Method: LAS environmental concentrations were predicted from per capita LAS use (2.5 g LAS/cap/day in Western Europe), water treatment statistics, and population estimates for Western Europe.

Results: The predicted LAS concentration range in the estuaries around the North Sea are 0.9-9 µg/L, which is in agreement with field monitoring data (1-9 µg/L) from the western Scheldt estuary.

Remarks: A risk assessment of LAS in marine sediments was initiated in 2002 and is expected to be available in 2004.

Reference: Temara, A., Carr, G., Webb, S., Versteeg, D. and Feijtel, T. 2001. Marine risk assessment: linear alkylbenzene sulfonate (LAS) in the North Sea. *Marine Poll. Bulletin*. 42:635-642.

Reliability: 2 Valid with restrictions

(v)

Type of Measurement: Background []; At contaminated site []; Other [X] **Coastal waters and sediments in the Elbe estuary**

Medium: Surface water and marine sediments

Test Substance: LAS

Method: Water samples (100-L) were collected from a depth of 5 m at several locations in the Elbe estuary of the German Bight of the North Sea. Sediment samples were collected at the same locations. All samples were extracted and analyzed for LAS and other compounds (e.g. nonylphenols) present in detergents.

Results: The maximum concentration of LAS in surface waters was 0.03 µg/L and occurred in marinas in the Elbe estuary. Sediment LAS concentration ranged from 39-109 µg/kg dry weight.

Remarks: The LAS found in marina sediment probably originated from the discharges of municipal wastewater treatment plants.

Reference: Bester, K., Theobald, N., and Schroeder, H.Fr. 2001. Nonylphenols, nonylphenol-ethoxylates, linear alkylbenzenesulfonates (LAS) and bis (4-chlorophenyl)-sulfone in the German Bight of the North Sea. *Chemosphere*. 45:817-826.

Reliability: 2 Valid with restrictions

3.3 TRANSPORT AND DISTRIBUTION BETWEEN ENVIRONMENTAL COMPARTMENTS INCLUDING ESTIMATED ENVIRONMENTAL CONCENTRATIONS AND DISTRIBUTION PATHWAYS

3.3.1 TRANSPORT

(a)

Type: partition coefficient
Media: sludge
Method: Monitoring data were collected in a pilot-scale municipal activated sludge treatment plant. The plant consisted of a completely mixed aeration tank (490L) and a secondary settler (280L). The plant was operated at C₁₂ LAS influent concentrations between 2 and 12 mg/L and at sludge retention times of 10 and 27 days. At least every other day 24-h samples of 2.5L influent and 2.5L effluent were collected in PE bottles and total (sum of adsorbed and dissolved) LAS concentrations were determined using an HPLC method adapted from Feijtel et al. 1995. At least once a day 200 mL grab samples were taken from the aeration tank and return sludge and transferred into PE centrifuge tubes for determination of dissolved and adsorbed LAS. The sludge samples were immediately centrifuged for 15 min at 3500 rpm. The supernatant was transferred into PE bottles and preserved by 3% formalin and stored for a maximum of 10 days at 4°C until further analysis. Representative aliquots of pre-settled influent, final effluent, or supernatant of the centrifuged sludge samples were passed over 6 mL preconditioned C18 SPE columns at a rate not exceeding 10 mL/min. The SPE columns were washed with 2 mL methanol/water and eluted with 5 mL of methanol. The eluate was then passed through strong anion exchange (SAX) columns, washed, eluted and subsequently evaporated to dryness under a gentle flow of nitrogen gas. The dry residue was dissolved in 2-5 mL of HPLC mobile phase. The HPLC was operated according to specifications. Identification of the different LAS alkyl homologues and quantification were made against a commercial LAS mixture (Marlon A390). A sorption-isotherm and the kinetics of adsorption and desorption of LAS to activated sludge were determined in batch experiments. Three different biodegradation tests were also carried out (an OECD 301F ready biodegradation test; a batch activated sludge [BAS] test; and a “by-pass” test developed to mimic condition of the pilot scale activated sludge plant). Only the sorption results are presented here.

Results: K_p (C₁₂ LAS): 3210 L/kg (log K_p = 3.5)
K_p (commercial C_{11.6} LAS mixture): 2,500 L/kg (log K_p = 3.4)

Test Substance: C₁₂ LAS

Remarks: Sorption equilibrium was achieved rapidly, within 5-10 minutes. Desorption was less pronounced, but still reached rapid equilibration. The sludge-water partition coefficient K_p of 3210 L/kg volatile suspended solids is reported. Applying the same QSAR for the commercial C_{11.6} LAS mixture results in a value of log K_p = 3.4 (i.e., K_p = 2500 L/kg), consistent with Feijtel et al. 1999 (see section 3.3.1(b)). In the other experiments conducted in this study, only 2-8% was present as dissolved C₁₂ LAS, with the remaining 92-98% adsorbed to the sludge. Despite this high degree of sorption, more than 99% of the LAS load was removed by biodegradation, showing that the adsorbed fraction as well as the soluble fraction of LAS is readily available for biodegradation.

Reference: Temmink, H. and Klapwijk, B. 2004. Fate of LAS in activated sludge plants. *Water Research* 38:903-912.

Reliability: 2 Valid with restrictions

(b)
Type: partition coefficient
Media: sludge

Method: QSAR analysis
Results: K_p (C_{11} LAS): 1000 L/kg ($\log K_p = 3.0$)
 K_p (C_{12} LAS): 3162 L/kg ($\log K_p = 3.5$)
 K_p (commercial $C_{11.6}$ LAS mixture): 2,512 L/kg
Test Substance: Pure C_{11} and C_{12} LAS; and commercial $C_{11.6}$ LAS
Remarks: The K_p values for C_{11} and C_{12} LAS are reported in this study as experimentally determined by Games et al. (1982), although they actually appear to be as reported in Games (1982; see 3.3.1 (d) for summary). The K_p for the commercial $C_{11.6}$ LAS mixture is calculated by Feijtel et al. using the reported C_{11} and C_{12} values. Also cites as a model input the $\log K_{oc}$ from Traina et al. 1995 (see 3.3.1(b) for summary). These values are consistent with the experimental results of Temmink and Kapwijk (see 3.3.1(a)).
Reference: Feijtel, T.C.J., Struijs, J., and Matthijs, E. 1999. Exposure modelling of detergent surfactants – Prediction of 90th-percentile concentrations in The Netherlands. *Environ. Toxicol. Chem.* 18:2645-2652.
Reliability: 4 Not assignable (see 3.3.1(e) for Games 1982)

(c)

Type: organic carbon partition coefficient
Media: dissolved humic substances
Method: The association of C_{10} , C_{12} and C_{14} LAS with natural and specimen-grade dissolved humic substances (DHS) was measured with fluorescence quenching and with ultracentrifugation techniques. Water-soluble organic carbon (Carlisle-WSOC) was obtained and extracted from 0-0.1 m depth of a Carlisle muck in northwest Ohio. The acid-soluble fraction (Carlisle-HA) was suspended, centrifuged, and dialyzed to remove Cl^- and to reduce the polyvalent cation content. The specimen-grade humic acid was purchased from Aldrich Chemical Company (Aldrich-HA). Suwanee River humic acid (SRHA) was obtained from the International Humic Substances Society. The fluorescence quenching followed the method of Guathier et al. (1986). Aliquots of the humic acid were placed in borosilicate bottles containing either NaCl, $CaCl_2$, or concentrated synthetic river water, capped and incubated at 25°C for 18-24 hours, after which an aliquot of C_{10} , C_{12} or C_{14} LAS was added and the solutions equilibrated at 25°C for 2 hours. All treatments were prepared in triplicate. After 2 hours, 3 mL of each sample solution was placed into cuvettes and the fluorescence of LAS measured with a Perkin Elmer LS 5B spectrofluorometer. Ultraviolet absorption measurements were made at 230 and 288 nm with a Beckman DU 6 UV-vis spectrophotometer. For the second independent analytical method, a batch ultracentrifugation method was developed. Aliquots of C_{12} LAS were added to polyallomer ultracentrifuge tubes containing the humic acid in a background electrolyte of either NaCl or $CaCl_2$ and allowed to equilibrate at 25°C. All treatments were prepared in triplicate. After 2 hours, the samples were centrifuged at 141,000 g for 6 hours at 25°C. The supernatants were decanted and saved for analysis. The walls of the centrifuge tubes were extracted with CH_3OH , which was saved for analysis. The concentration of C_{12} LAS in the supernatants and in the CH_3OH extracts was determined by exciting the solutions in cuvettes at 230 nm and the emission intensity recorded at 288 nm with a Perkin Elmer LS-5B spectrofluorometer. The quantity of C_{12} LAS associated with DHS was calculated from the

difference in the initial and final solution concentrations, following the correction for the quantity of C₁₂ LAS sorbed to the walls of the centrifuge tubes.

Results: The average Log K_{oc} values over the four DHS materials (Aldrich-HA, Carlisle-WSOC, Carlisle-HA, and SRHA) for each LAS chain length tested are:

Log K_{oc} (C₁₀ LAS): 4.02 L/kg

Log K_{oc} (C₁₂ LAS): 4.83 L/kg

Log K_{oc} (C₁₄ LAS): 5.49 L/kg

The data for Ca-saturated Aldrich-HA was linear over the entire concentration range of DHS, whereas some curvatura was present in the data from the Ca-saturated SRHA, and considerable deviation from linearity was apparent in many of the Na-saturated DHS solutions.

Test Substance: C₁₀ LAS (98% purity), C₁₂ LAS (93% purity), and C₁₄ LAS (88% purity), each synthesized at Procter and Gamble. Uniformly ¹⁴C-ring labeled C₁₀-, C₁₂- and C₁₄-LAS were obtained from New England Nuclear and were 93.8, 96.3 and 92.5% pure, with specific activities of 26.6, 68.2, and 34.3 μCi/mg, respectively.

Remarks: Good agreement was obtained with both of the analytical methods, indicating that both techniques can be used to quantify the effects of DHS on speciation of LAS in natural waters with certain limitations. LAS-DHS partition coefficients increased with increasing length of the alkyl chain in the LAS. These data indicate the significance of nonpolar forces in LAS-organic matter interactions. Good agreement was found between the partition coefficients obtained from the two analytical techniques and those calculated from the response of uptake and depuration studies conducted with fathead minnows.

Reference: Traina, S.J., McAvoy, D.C. and Versteeg, D.J. 1996. Association of LAS with dissolved humic substances and its effect on bioavailability. *Env. Sci. Technol.* 30:1300-1309.

Reliability: 2 Valid with restrictions

(d)

Type: Adsorption ; Desorption ; Volatility ; Other

Media: water - activated sludge

Method: estimation of K_d with Freundlich equation

Results: The K_d of commercial LAS was between 660 and 5200 L/kg (6 citations), dependant on organic carbon content and other characteristics of the solid phase.

Remarks: Concentration in liquid phase between 1 and 80 mg/L.

Reference: Painter, H.A. and Zabel, T.F. 1988. Review of the environmental safety of LAS. Wrc Medmenham, UK. Report No. CO 1659-M/1/EV 8658.

Reliability: 4 Not assignable

(e)

Type: Adsorption ; Desorption ; Volatility ; Other

Media: water - activated sludge and water - river sediment

Method: Comparison of K_d for LAS C₁₀ to C₁₄ - Year 1982

Results: Adsorption coefficients (L/kg) in activated sludge and river sediment, respectively, for LAS homologues:

C₁₀: 220 and 41

C₁₁: 1000 and 100

C₁₂: 3070 and 330
 C₁₃: 9330 and 990
 C₁₄: 2950 (for river sediment – not determined for activated sludge)
 Remarks: K_d is highly dependent on the alkyl chain length of LAS with approximately a factor of 3 increase per carbon and on the position of the phenyl group on the alkyl chain. The organic carbon content is also an important factor, varying by a factor of 10 between activated sludge and river sediment.
 Reference: 1) Games, L.M. 1982. Field validation of exposure analysis modelling systems (EXAMS) in a flowing stream. Ch. 18. In: Dickson, K.L., Maki, A.W. and Cairns, J. (ed.). 1981. Modelling and Fate of Chemicals in the Aquatic Environment. 4th Meeting. Sci. Ann Arbor Michigan Pp. 325-346.
 2) Painter, H.A. and Zabel, T.F. 1988. Review of the environmental safety of LAS. Wrc Medmendham, UK. Report No. CO 1659-M/1/EV 8658.
 Reliability: 4 Not assignable because no details of LAS concentration were provided.

(f)

Type: Adsorption [X]; Desorption []; Volatility []; Other []
 Media: water - river sediments
 Method: estimation of K_d with Freundlich equation
 Results: K_d between 6 and 300 L/kg (5 citations), dependent on organic carbon content and other characteristics of the solid phase.
 Remarks: Concentration in liquid phase between 0.06 and 15 mg/L
 Reference: Painter, H.A. and Zabel, T.F. 1988. Review of the environmental safety of LAS. Wrc Medmendham, UK. Report No. CO 1659-M/1/EV 8658.
 Reliability: 4 Not assignable

(g)

Type: Adsorption [X]; Desorption []; Volatility []; Other []
 Media: water - soil
 Method: estimation of K_d with Freundlich equation
 Results: K_d between 2 and 20 L/kg (3 citations), dependent on organic carbon content and other characteristics of the solid phase.
 Remarks: Concentration in liquid phase between 0.06 and 15 mg/L
 Reference: Painter, H.A. and Zabel, T.F. 1988. Review of the environmental safety of LAS. Wrc Medmendham, UK. Report No. CO 1659-M/1/EV 8658.
 Reliability: 4 Not assignable

(h)

Type: Adsorption [X]; Desorption []; Volatility []; Other []
 Media: water - soil
 Method: compilation of K_F data from other sources. The Freundlich isotherm is a general sorption isotherm which describes sorption behaviour and often is used in studies of surfactant sorption. K_F is the Freundlich isotherm coefficient which expresses the affinity of a surfactant for a given solid sorbent. As shown in the equation and table below, the exponent n is a measure of isotherm non-linearity. For n approaching 1, the Freundlich model of sorption becomes equivalent to a linear sorption model.

$$C_s = K_F \times C_w^n$$

Results: Log K_F values for a selection of C₁₂-LAS types is shown in the table below:

Log K_F	n	Sorbent	Reference
1.7	1	EPA-B1	Hand and Williams (1987)
2.0	1	EPA-5	Hand and Williams (1987)
2.7	1	RC4	Hand and Williams (1987)
3.2	1	RC3	Hand and Williams (1987)
~0.1	1	different soils	Ou et al. (1996)
2.8	1.15	marine sediment	Rubio et al. (1996)
0.6	0.77	soil, clay loam(A)	Abe and Seno (1985)
1.4	1.20	soil, clay loam(B)	Abe and Seno (1985)
1.2	1.19	soil, sandy loam	Abe and Seno (1985)

Sorbent refers to the standard or natural soil or sediment used, as the affinity for sorption depends on both the chemical substance and the characteristics of the sorbent. All data were drawn from the original sources referenced in the table.

Remarks: The nonlinearity parameter implies that sorption affinity decreases with increasing LAS concentrations, which suggests that concentration dependency should be taken into account when assessing sorption of surfactants such as LAS.

Reference: Tolls, J. and Sijm, D.T.H.M. 2000. Estimating the properties of surface-active chemicals. In: Boethling, R.S. and Mackay, D. Handbook of Property Estimation Methods for Chemicals. Lewis Publishers.

Reliability: 4 Not assignable because the original articles were not directly reviewed.

3.3.2 THEORETICAL DISTRIBUTION (FUGACITY CALCULATION)

(a)

Media: Air-biota []; Air-biota-sediment-soil-water [X]; Soil-biota []; Water-air []; Water-biota []; Water-soil []

Method: Fugacity level I []; Fugacity level II []; Fugacity level III [X]; Fugacity level IV []; Other (calculation) []; Other (measurement) []

Five stage Mackay-type modelling including evaluative, regional and local-scale models. The first two stages involve classifying the chemical and quantifying the emissions into each environmental compartment. In the third stage, the characteristics of the chemical are determined using a quantitative equilibrium criterion model (EQC), which is conducted in three steps using levels I, II, and III versions of the model that introduce increasing complexity and more realistic representations of the environment. The EQC uses a generic, evaluative environment, which is 100,000 km² in area. In the fourth stage, ChemCAN, which is a level III model for specific regions of Canada, was used to predict the chemical's fate in southern Ontario. The final stage was to apply local environmental models to predict environmental exposure concentrations. For LAS, the WW-TREAT, GRiDS, and ROUT models were used to predict the fate of LAS in a sewage treatment plant and riverine receiving waters. Estimated properties used as input parameters to the models are shown below for LAS (from various sources; average values; based on the best default

environmental and physicochemical values available at the time of modeling):

Molecular mass	348
Air-water partition coefficient	0
Aerosol-water partition coefficient	100
Soil-water partition coefficient (L/kg)	20
Sediment-water partition coefficient (L/kg)	570
Fish-water partition coefficient (L/kg)	250
Half-life in air (h)	--
Half-life in water (h)	24
Half-life in soil (h)	480
Half-life in sediment (h)	96

The level I calculation assumes a steady-state equilibrium partitioning of a fixed quantity of LAS (100,000 kg) with no reaction or advection processes. The level II calculation assumes a fixed input of 1000 kg/h, which is balanced by reaction and advection losses. Relative partitioning is identical to level I. For level III, the ChemCAN model assumes the following estimated input quantities for LAS:

Total discharge to the environment (kg/yr)	1,444,000
Discharge to the air	--
Discharge to water	144,000
Discharge to soil	1,296,000
Total input in the region (kg/year)	1,440,000
Total input in the region (kg/hour)	164.4

The authors base these input quantities for the ChemCAN model on a recent estimate of LAS annual production in North America, western Europe and Japan as approximately 1.4 million tons and an annual per capita consumption of LAS in the United States of 1.3 kg/year. LAS is disposed "down-the-drain" and approximately 98% is removed in sewage treatment. About 30% of the LAS is removed in treatment by adsorption onto primary and secondary sewage solids. Over 60% of the sludge was assumed to be disposed of in landfills or applied to agricultural soils, thus there is the potential for LAS to reach the soil environment. Therefore, the level III model assumes a substantial discharge (90%) of LAS to soil following sewage treatment. Inputs considered the specific nature of nonvolatile surfactants such as LAS. For example, the use of Kow as a descriptor for organic phase-water partitioning is inappropriate for LAS and there is no need for a vapor pressure or air-water partition coefficient. Because LAS is a mixture, average properties were used as inputs to the models. In the EQC model, LAS is treated using the equivalence approach as the equilibrium criterion.

Results:

The level I and II models each resulted in LAS partitioning to air, water, soil, and sediment at percentages of 0%, 25.97%, 56.09%, and 17.76%, respectively. The overall residence time of LAS is 100 hours and removal is primarily by biodegradation in water (76%) and partitioning in sediment (13%). Thus, the impacts of LAS will be restricted to local receiving waters and their sediments and biota. In level III, when

discharges are directly to water, the residence time is 33 hours and more than 99% remains in the water, though in shallower receiving waters more partitioning to sediments might be expected. When the discharge is to soil, as was assumed in the ChemCAN model, the residence time is 28 days because of the slower biodegradation rate and little transfers to other media. Based on these findings, the dominant fate processes are degradation rates in water and soil, and water-sediment transfer.

Using the ChemCAN 4 model, of the total amount of LAS released to the environment assuming the discharge rates above, the distribution and concentrations were predicted to be:

to air:	0%	(0 mg/m ³)
dissolved in water:	0.64%	(0.44 µg/m ³)
in soil:	99.35%	(7.06 µg/kg)
in sediment:	0.0036%	(0.00347 µg/kg)

Remarks: Based on an estimated total discharge to the environment of 1.44 x 10⁶ kg/year (1.44 x 10⁵ kg to water and 1.296 x 10⁶ kg to soil). It should be noted that the discharge assumptions used by the authors are highly conservative and likely overpredict the amount of LAS entering various compartments, for example, the soil compartment. This study was conducted by the model developer and acknowledged expert on fugacity to demonstrate that the approach was appropriate for different types of chemicals.

Reference: Mackay, D., Di Guardo, A., Paterson, S., Kicsi, G., Cowan, C. and Kane, D. 1996. Assessment of chemical fate in the environment using evaluative, regional and local scale models: Illustrative application to chlorobenzene and linear alkylbenzene sulfonates. Environ. Toxicol. Chem. 15:1638-1648.

Reliability: 2 Valid with restrictions

(b)

Media: Air-biota []; Air-biota-sediment-soil-water [X]; Soil-biota []; Water-air []; Water-biota []; Water-soil []

Method: Fugacity level I []; Fugacity level II []; Fugacity level III [X]; Fugacity level IV []; Other (calculation) []; Other (measurement)[] Multi-media model HAZCHEM derived from MacKay type level III model, including a water purification module (comparable to SIMPLETREAT model used by the Netherlands Authorities). Regional model for Europe. Includes probabilistic evaluation of natural variability and inaccuracy in determination of input parameters using Monte Carlo simulation. Input parameters held constant in the simulations for the LAS used in the model were based on the best available data at the time of modeling and are:

Molecular weight	347
Melting point (°C)	10
Vapor pressure (Pa)	1 x 10 ⁻¹⁰
Solubility (mg/L)	350
Log Kow	2.5
Half-life life in air (h)	8
Half-life in water (h)	35
Half-life in soil (h)	339

Half-life in sediment (h)	17
Soil-water partition coefficient (L/kg)	1000
Sediment-water partition coefficient (L/kg)	1000
Suspended solids-water partition coefficient (L/kg)	1000

Parameters that were varied using lognormal distributions for the Monte Carlo simulations included water surface and arable land fractions, depths of various compartments, fraction organic carbon in various compartments, advective residence times, temperature, and others.

Results: Predicted concentrations in the various compartments as defined by the model as shown in the table below. Degradation in the soil was not taken into account and the model was not calibrated. Measured concentrations in arable soil were generally below 1 ppm. Results are the 5th and 95th percentile values from the Monte Carlo analysis and the normal average.

	Average	5 percentile	95 percentile
air (ug/m ³)	3.23 E-12	--	--
biota (ppm)	5.99 E-2	7.11 E-3	1.95 E-1
sediment (ppm)	4.90 E-3	3.23 E-3	1.38 E-2
arable soil (ppm)	44.2	7.02	1.11 E+2
suspended. matter (ppm)	3.79	0.449	12.3
dissolved in water (mg/L)	3.79 E-3	4.49 E-4	1.23 E-2
suspended in water (mg/L)	7.81 E-5	1.54 E-5	2.16 E-4

Remarks: Input data were the best estimates from ECETOC Task Force members, including detergent industries. Note the predicted average value for arable soil was reported in ECETOC 1993 as 4.42E-4, which is not possible given the 5th and 95th percentile values. Therefore, the recalculated value provided in the IUCLID HEDSET (Year 2000 data) was used above.

Reference: 1) ECETOC. 1993. Environmental hazard assessment of substances. Technical Report No. 51, European Centre for Ecotoxicology and Toxicology of Chemicals, Brussels.
2) BKH. 1993. The use of existing toxicity data for estimation of the Maximum Tolerable Environmental Concentration of Linear Alkyl Benzene Sulfonate, Part I: Main report; Part II: Data base. Study carried out for ECOSOL, BKH Consulting Engineers, Delft, NL.

Reliability: 4 Not assignable because of uncertainty related to the input parameters.

3.4 MODE OF DEGRADATION IN ACTUAL USE

Remarks: Refer to other sections.

3.5 BIODEGRADATION

(a)
Type: aerobic [X]; anaerobic []
Inoculum: adapted []; non-adapted [X] Marl, East treatment plant (Germany)
Concentration: 10 mg/L
Medium: Semi-continuous activated sludge (SCAS)

Method: The Marl purification plant consists of a mechanical prepurifying area, two parallel trickling filters of 800 m³ volume each, and a stimulation plant of 106 m³ volume, as well as mechanical final clarification. Samples were collected at the inlet of the trickling filters and at the exit of the final clarifier. Three-hour composite samples were taken for each analysis. The MBAS procedure was used to quantify LAS concentrations. The specific method for analyzing the homologues is reported in Wickbold 1964.

Results: Results of the percent removal by activated sludge for different alkyl chain length homologues and phenyl positions are shown in the following table:

	Phenyl Position	% Degradation
C ₁₀	5	52
	4	68
	3	88
	2	92
C ₁₁	6	58
	5	72
	4	89
	3	93
	2	94
C ₁₂	6	81
	5	92
	4	94
	3	95
	2	95
C ₁₃	7,6 ^a	92
	5	94
	4	95
	3	96
	2	96

^a Not analytically separable

GLP: Yes [] No [X] ? []

Test Substance: C₁₀ to C₁₃ homologues of LAS with varying phenyl positions

Remarks: The results show that longer chain homologues are removed faster than the shorter chain homologues, indicating that primary biodegradation increases as the chain length increases. In addition, the greater the distance between the sulfonic group and the more distant terminal methyl group on the alkyl chain the faster the degradation (i.e., phenyl position 2 degrades faster than phenyl position 5 for the same homologue). For example, percent removal was 52, 68, 88 and 92% for the C₁₀ LAS with phenyl positions 5, 4, 3, and 2, respectively.

Reference: 1) Bock, K.J. and Wickbold, R. 1966. Auswirkungen der Umstellung auf leicht abbaubare Waschrohstoffe in einer großtechnischen Kläranlage und im Vorfluter. Vom Wasser 33:242-253.
2) Wickbold, R. 1964. Zwischenprodukte beim Abbau eines geradkettigen Alkylbenzolsulfonates. Vorträge IV, Intern. Kongr. F. grenzflächenaktive Stoffe, Brüssel. (not available for review)

Reliability: 2 Valid with restrictions

(b)

Type: aerobic ; anaerobic
 Inoculum: adapted ; non-adapted ; activated sludge
 Concentration of the chemical: 10 mg/L related to COD ; DOC test substance
 Medium: water ; water-sediment ; soil ; sewage treatment
 Degradation: 69.6% after 28 days (acclimated inoculum)
 66.7% after 28 days (non-acclimated inoculum)
 Results: readily biodeg. ; inherently biodeg. ; under test condition no biodegradation observed , other
 Kinetics:

Days	% Biodegradation (Acclimated)	% Biodegradation (non-Acclimated)
5	14.3	9.2
8	32.4	24.5
12	49.5	43.6
19	63.9	60.8
28	69.6	66.7

Method: OECD 301 B, modified Sturm test, also reported as method C 5 in the Italian D.M. 109. The reference standard sodium benzoate was used at the same concentration of the tested surfactants (about 10 mg/L). The testing period was 28 days and during this time the testing solutions were kept in dark glass vessels. A supernatant solution from a sludge, which was aerated and left to settle, was used as an inoculum for each biodegradation test at an amount equal to 1% of the solution. Both the STP sludges and the laboratory assimilated sludges were analysed for the dry matter contents. Tests were conducted using acclimated or non-acclimated inocula to determine whether this affected ultimate biodegradation. Acclimation was achieved by running the laboratory activated sludge plant for seven days before testing with a synthetic influent containing LAS at a concentration of about 10 mg/L. Tests were conducted on LAS and three other surfactants.

GLP: Yes No ?
 Test substance: C₁₀₋₁₃ LAS; average alkyl chain length = C_{11.6}
 Remarks: LAS was biodegradable using both acclimated and non-acclimated inocula. Acclimated inocula did not significantly improve the total biodegradation, but did accelerate the attainment of the degradation plateau. The 10-day time-window criterion was missed slightly, so this study does not meet the criteria for ready biodegradability. However, this is expected given biodegradation kinetic curve dynamics related to increasing dissolved organic carbon content because the CO₂ generated during the biodegradation process is not totally evolved and removed from the test medium.
 Reference: Ruffo, C., Fedrigucci, M.G., Valtorta, L., and Cavalli, L. 1999. Biodegradation of anionic and non-ionic surfactants by CO₂ evolution. Acclimated and non-acclimated inoculum. Riv. It. Sostanze Grasse LXXVI: 277-283.
 Reliability: 1 Valid without restriction

(c)
 Type: aerobic ; anaerobic
 Inoculum: adapted ; non-adapted ; domestic activated sludge from Enid, OK

Concentration: 10 mg/L
 Medium: Semi-continuous activated sludge (SCAS)
 Degradation: >98% in 20 days (primary biodegradation)
 62% in 20 days (inherent biodegradation)
 Results: readily biodeg. ; inherently biodeg. ; under test condition no biodegradation observed , other
 Method: ¹⁴C ring-labeled LAS and ¹⁴C alkyl-labeled C₁₂ LAS were introduced to a simulated secondary waste treatment system (SCAS) following the ASTM and SDA standard methods.
 GLP: Yes No ?
 Test Substance: 1) LAS with the following homologue composition: C₁₁ 42%, C₁₂ 38%, C₁₃ 20%; average alkyl chain length C_{11.8}
 2) C₁₂ LAS
 Remarks: In a secondary waste treatment environment, the alkyl and ring portions of LAS both biodegrade extensively, with the fate of the LAS alkyl and ring carbon nearly identical. Within the 20 day test period, 62% of the alkyl and ring carbon converted to carbon dioxide.
 Reference: Nielsen, A.M. and Huddleston, R.L. 1981. Ultimate biodegradation of linear alkylbenzene sulfonate alkyl and ring carbon. *Developments in Industrial Microbiology*. 22:415-424.
 Reliability: 2 Valid with restrictions

(d)

Type: aerobic ; anaerobic
 Inoculum: Bacterial biomass obtained from the settled supernatant slurry solution of a fertile soil
 Concentration: 10 mg/L
 Medium: water ; water-sediment ; soil ; sewage treatment
 Degradation: See methods
 Results: See remarks
 Method: OECD 301E Ready Biodegradability test, with the following modifications: LAS was the sole source of carbon introduced (i.e., no activated sludge inoculum), along with an enriched level of bacterial biomass. Preliminary tests showed that more than 90% of the LAS disappeared within 4 days, so LAS was restored by adding about 10 mg/L of fresh substance every 4 days for 80 days. The test was stopped 4 days after the last LAS addition (i.e., at 84 days). Specific HPLC analysis was used to measure LAS and SPCs.
 GLP: Yes No ?
 Test Substance: Commercial HF-type LAS with a C₁₀₋₁₃ alkyl chain and a linearity of about 93% (DATS <0.5%; iso-branching 5-6%). (CAS #68411-30-3); average alkyl chain length = C_{11.6}
 Remarks: The final organic residue of this prolonged biodegradation test was characterized in detail and showed that no accumulation of iso-branching structures had occurred. This indicates that iso-branched material of LAS is amenable to biodegradation as well as the linear components.
 Reference: Cavalli, L., Cassani, G., Lazzarin, M., Maraschin, C., Nucci, G. and Valtorta, L. 1996b. Iso-branching of linear alkylbenzene sulphonate (LAS). *Tenside Surf. Det.* 33:393-398.
 Reliability: 2 Valid with restrictions

(e)

Type: aerobic ; anaerobic

Inoculum: adapted ; non-adapted ; activated sludge, domestic

Concentration: 10.8 mg/L related to COD ; DOC test substance

Medium: water ; water-sediment ; soil ; sewage treatment

Degradation: 93% after 28 days

Results: readily biodeg. ; inherently biodeg. ; under test condition no biodegradation observed , other

Kinetic: 7 day = 59%
14 day = 73%
21 day = 82%

Method: Directive 79/831/EEC, Appendix V, C.4-A 1990. DOC Die-Away Test. (OECD 301A Test). Samples were collected from an activated sludge basin with predominantly local municipal waste water. The final sludge concentration was 19.3 mg/L. Two replicates were used for the LAS test concentration (9.44 mg/L) with inoculum, one with inoculum without LAS, and two control replicates (sodium benzoate, 10.13 mg/L) with inoculum. A total of 900 mLs of the solutions were put into 2000 mL Erlenmeyer flasks at the beginning of the test. The loosely covered flasks were incubated at 21.5 to 22.6°C in the dark on a mechanical shaker for 28 days. Samples were collected on days 0, 7, 14, 21 and 28 for DOC analysis.

GLP: Yes No ?

Test substance: Marlon A 390 (CAS #68411-30-3) C₁₀₋₁₃ LAS; average alkyl chain length = C_{11.6}

Remarks: LAS is readily biodegradable. The 10-day window criterion was fulfilled. The control substance (sodium benzoate) showed 99% degradation after 28 days. This is a key study for ready biodegradability (see SIAR Table 4).

Reference: Schoeberl, P. 1993b. Bestimmung der biologischen Abbaubarkeit von Marlon A 390 im DOC-DIE AWAY Test. Huels Final Report No. DDA-21.

Reliability: 1 Valid without restriction

(f)

Type: aerobic ; anaerobic

Inoculum: adapted ; non-adapted ; activated sludge

Concentration: 9.7 mg/L related to COD ; DOC test substance

Medium: water ; water-sediment ; soil ; sewage treatment

Degradation: 94% after 28 days

Results: readily biodeg. ; inherently biodeg. ; under test condition no biodegradation observed , other

Kinetic: 3 day = 38%
7 day = 81%
14 day = 88%
21 day = 94%

Method: Directive 79/831/EEC, Appendix V, C.4-A - Year: 1990. DOC Die-Away Test. (OECD 301A Test). Samples were collected from an activated sludge basin with predominantly local municipal waste water. The final sludge concentration was 18.1 mg/L. Two replicates were used for the LAS test concentration (8.96 mg/L) with inoculum, one with inoculum without LAS, and two control replicates (sodium benzoate, 11.65 mg/L) with inoculum. A total of 900 mLs of the solutions were put into 2000 mL

Erlenmeyer flasks at the beginning of the test. The loosely covered flasks were incubated at 21.8 to 22.2°C in the dark on a mechanical shaker for 28 days. Samples were collected on days 0, 3, 7, 14, 21, 27 and 28 for DOC analysis.

GLP: Yes No ?
Test substance: Marlon A 390 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}
Remarks: LAS is readily biodegradable. The 10-day window criterion was fulfilled. The control substance (sodium benzoate) showed 96% degradation after 28 days. This is a key study for ready biodegradability (see SIAR Table 4).
Reference: Schoeberl, P. 1993c. Bestimmung der biologischen Abbaubarkeit von Marlon A 390 im DOC-DIE AWAY Test. Huels Report No. DDA-32.
Reliability: 1 Valid without restriction

(g)

Type: aerobic ; anaerobic
Inoculum: adapted ; non-adapted ; municipal sewage treatment plant effluent.
Concentration: 5 mg/L related to COD ; DOC test substance
Medium: water ; water-sediment ; soil ; sewage treatment
Degradation: 76% after 28 day
Results: readily biodeg. ; inherently biodeg. ; under test condition no biodegradation observed , other
Method: Directive 84/449/EEC, C.3 Modified OECD screening test. (OECD 301E Test).
GLP: Yes No ?
Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}
Reference: European Commission. 2000b. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).
Reliability: 4 Not assignable

(h)

Type: aerobic ; anaerobic
Inoculum: adapted ; non-adapted ; activated sludge
Concentration: 10 mg/L related to COD ; DOC ; test substance
Medium: water ; water-sediment ; soil ; sewage treatment
Degradation: 91.6% based on DOC reduction
Results: readily biodeg. ; inherently biodeg. ; under test condition no biodegradation observed , other
Method: OECD Guideline 303 A "Simulation Test - Aerobic Sewage Treatment: Coupled Unit Test" 1981. The studies were carried out in the OECD Confirmatory Test plant at different laboratories using synthetic wastewater as specified in the EC Guidelines 82/242 and 82/243. The amount of surfactant supplied was 10 mg/ of MBAS/L. The amount of MBAS in the wastewater feed corresponds approximately to that detected in the feed of municipal sewage plants, which corresponds to about 6 mg DOC/L. Test periods in the different laboratories ran from 33 to 139 days.
GLP: Yes No ?
Test substance: C₁₀₋₁₃ LAS, sodium salt (CAS #68411-30-3); average alkyl chain length = C_{11.6}

Remarks: The degradation rate of 91.6% is the mean of 10 studies conducted at 7 different laboratories, based on DOC reduction. Since numerous studies have shown that not only anionic surfactants are shown in the MBAS analysis, the degradation plant discharge was analyzed on days 22, 24, 29 and 30 using HPLC analysis. Results showed that the content of intact LAS was < 20 µg/L, which is about 8% of the 250 µg/L MBAS content in the discharge. This means that the real LAS primary degradation reaches 99.8%.

Reference: Schoeberl, P. 1991. Coupling the OECD confirmatory test with continuous ecotoxicity tests. Tenside Surf. Det. 28:6-14.

Reliability: 2 Valid with restrictions

(i)

Type: aerobic [X]; anaerobic []

Inoculum: adapted []; non-adapted [X]; municipal sewage treatment plant effluent

Concentration: 5 mg/L related to COD []; DOC []; test substance [X] MBAS

Medium: water [X]; water-sediment []; soil []; sewage treatment []

Degradation: 95% after 19 days

Results: readily biodeg. [X]; inherently biodeg. []; under test condition no biodegradation observed [], other []

Method: OECD Screening Test according to "Verordnung ueber die Abbaubarkeit anionischer und nichtionischer grenzflaechenaktiver Stoffe in Wasch- und Reinigungsmittel vom 30.1.1977". Bundesgesetzblatt Teil I, S. 244. 1977

GLP: Yes [] No [X] ? []

Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}

Reference: European Commission. 2000b. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).

Reliability: 4 Not assignable

(j)

Type: aerobic [X]; anaerobic []

Inoculum: adapted []; non-adapted [X]; synthetic sewage

Concentration: 19.2 mg/L related to COD []; DOC []; test substance [X]

Medium: water [X]; water-sediment []; soil []; sewage treatment []

Degradation: 92.3% based on DOC reduction

Results: readily biodeg. []; inherently biodeg. []; under test condition no biodegradation observed [], other [X]

Method: OECD-Guideline 303A coupled units test. The studies were carried out in the OECD Confirmatory Test plant at different laboratories using synthetic wastewater as specified in the EC Guidelines 82/242 and 82/243. In these studies, LAS was added to the test at 10 mg/L. Test periods in the different laboratories ran from 33 to 139 days. Test temperature ranged from 19.6-23.0°C.

GLP: Yes [X] No [] ? []

Test substance: Marlon A390 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.8}

Remarks: The degradation rate of 92.3% is the mean of 10 studies conducted at 7 different laboratories, based on DOC reduction, with LAS added to the confirmatory test plant.

Reference: 1) Schoeberl, P. 1991. Coupling the OECD confirmatory test with continuous ecotoxicity tests. Tenside Surf. Det. 28:6-14.
2) European Commission. 2000b. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).

Reliability: 2 Valid with restrictions

(k)

Type: aerobic [X]; anaerobic []
Inoculum: adapted []; non-adapted [X];
Concentration: 10 mg/L
Medium: related to COD []; DOC []; test substance [X] as MBAS water [X]; water-sediment []; soil []; sewage treatment []
Degradation: 99.8% after 3 days (for sewage dose of 1.0 mg/L)
89.4% after 7 days (for sewage dose of 0.5 mg/L)
74.5% after 7 days (no sewage dose, but aerated)
40.7% after 7 days (no sewage, non aerated)
Results: readily biodeg. []; inherently biodeg. []; under test condition no biodegradation observed [], other [X] primary biodegradation
Method: Samples from test flasks were taken at 12 hour intervals and the detergent concentration expressed as methylene blue active substance was determined. The initial concentration of detergent was 10 mg/L. The degradation of LAS was evaluated in raw canal water and in canal water seeded with either 0.5 ml/L or 1.0 ml/L sewage from the Ismailia sewage treatment plant.
GLP: Yes [] No [X] ? []
Test substance: Commercial LAS detergent (provided by Merck, Darmstadt, Germany); likely average alkyl chain length = C_{11,6}
Remarks: Medium was Ismailia Canal water (Cairo, Egypt). ABS and a 1:1 mixture of ABS:LAS were also examined. Degradation of LAS was rapid, whereas degradation of ABS and the 1:1 mixture of ABS/LAS was slower. In all cases, aeration and addition of sewage microflora enhanced degradation.
Reference: Abdel-Shafy, H.I., Azzam, A.M. and El-Gamal, I.M. 1988. Studies on the degradation of synthetic detergents by sewage. Bull. Environ. Contam. Toxicol. 41:310-316.
Reliability: 2 Valid with restrictions

(l)

Type: aerobic [X]; anaerobic []
Inoculum: adapted []; non-adapted [X];
Concentration: 15 mg/L related to COD []; DOC []; test substance [X] HPLC
Medium: water []; water-sediment []; soil []; sewage treatment [X]
Degradation: 95% after 28 days (LAS-A)
98% after 28 days (LAS-B)
Results: readily biodeg. []; inherently biodeg. []; under test condition no biodegradation observed [], other [X]
Method: OECD303A coupled units test modified by AIS/CESIO "ad hoc" working group.
GLP: Yes [X] No [] ? []
Test substance: Two C₁₀₋₁₃ LAS commercial products as described in the remarks.

Remarks: LAS-A, produced by the HF process, was 93% linear with 0.5% tetralins and 6.5% iso-branching and the following homologue distribution of the alkyl chain: C₁₀ 15%, C₁₁ 34%, C₁₂ 31%, C₁₃ 20% (average alkyl chain length = C_{11.56}). LAS-B, produced by the AlCl₃ process, was 98% linear with 0.5% tetralins and 1.5% iso-branching and the following homologue distribution of the alkyl chain: C₁₀ 15%, C₁₁ 29%, C₁₂ 32%, C₁₃ 24%, average alkyl chain length = C_{11.65}. HPLC methods specific to LAS were used to directly measure the test substances (LAS as well as the biodegradable intermediates). The ultimate biodegradation rates determined by HPLC are >10% higher than those obtained using DOC determination. The studies were conducted according to high standards and should be considered reliable.

Reference: Cavalli, L., Cassani, G. and Lazzarin, M. 1996a. Biodegradation of linear alkylbenzene sulphonate (LAS) and alcohol ethoxylate (AE). *Tenside Surf. Det.* 33:158-165.

Reliability: 2 Valid with restrictions

(m)

Type: aerobic ; anaerobic

Inoculum: adapted ; non-adapted ; None Stated

Concentration: 15 mg/L related to COD ; DOC ; test substance HPLC

Medium: water ; water-sediment ; soil ; sewage treatment

Degradation: 95% after 6 days

Results: readily biodeg. ; inherently biodeg. ; under test condition no biodegradation observed , other

Method: LAS and water collected from a briny pond (salinity: 9.5 g/L) were tested in an aerated cylindrical reactor at 21°C. Samples were removed periodically and analyzed for parent compound and metabolites by HPLC.

GLP: Yes No ?

Test substance: LAS (CAS #25155-30-0); activity: >99%; average alkyl chain length = C_{11.6}

Remarks: The half-life of LAS was 1.5 days. The HPLC method employed accurately defines the metabolites formed by primary biodegradation. All metabolites were not persistent and rapidly underwent further biodegradation.

Reference: Sarrazin, L., Arnoux, A., Rebouillon, P. and Monod, J.L. 1997. Biodegradation of linear alkylbenzenesulfonate (LAS) in briny water and identification of metabolites using HPLC analysis by direct injection of samples. *Toxicological and Environmental Chemistry.* 58:209-216.

Reliability: 2 Valid with restrictions

(n)

Type: aerobic ; anaerobic

Inoculum: adapted ; non-adapted ; activated sludge

Concentration: 1 mg/L related to COD ; DOC ; test substance

Medium: Activated sludge

Degradation: >96% after 6 hours

Results: Primary and complete biodegradation were described by a first-order model, with rate constants of 0.96-1.10/h (t_{1/2} = 0.63–0.72 h) for primary loss and 0.50-0.53/h (t_{1/2} = 1.30–1.38 h) for complete degradation.

Method: Radiolabeled LAS was dosed at an environmentally relevant concentration into biotic and abiotic activated sludge. Activated sludge mixed liquor

was used from two STP (Polk Run and Sycamore) near Cincinnati. Standard methods were used and the test flasks (reactors) were maintained at 20⁺/₋₂°C.

GLP: Yes [] No [] ? [X]

Test Substance: ¹⁴C [uniform ring] C₁₂ LAS

Reference: Federle, T.W. and Itrich, N.R. 1997. Comprehensive approach for assessing the kinetics of primary and ultimate biodegradation of chemicals in activated sludge: Application to linear alkylbenzene sulfonate. Environ. Sci. Technol. 31:1178-1184.

Reliability: 2 Valid with restrictions

(o)

Type: aerobic []; anaerobic [X]

Methods: Experiments were conducted in which enriched cultures of anaerobic bacteria were provided with 60 μmole/L LAS as the sole source of sulfur. Conditions were maintained anoxic in salts-medium containing several sources of carbon.

Results: Strain RZ LAS, an anaerobic bacteria, was isolated from wastewater treatment plants in Germany. RZLAS was shown to degrade LAS, indicating that microorganisms able to metabolize LAS in anaerobic conditions exist in nature.

GLP: Yes [] No [] ? [X]

Test Substance: Commercial C₁₀₋₁₃ LAS (CAS #68411-30-3; average alkyl chain length = C_{11.6}) and C₁₂ LAS (pure homologue)

Reference: Denger, K. and Cook, A.M. 1999. Linear alkylbenzene sulphonate (LAS) bioavailable to anaerobic bacteria as a source of sulphur. Journal of Applied Microbiology. 86:165-168.

Reliability: 2 Valid with restrictions

(p)

Type: aerobic []; anaerobic [X]

Inoculum: adapted []; non-adapted [X]; granular sludge

Concentration: 9 concentrations ranging from 12.5 to 350 mg/bottle

Medium: Liquid

Degradation: 5-44% after 14 days

Method: Upflow Anaerobic Sludge Blanket (UASB) type reactors with a working volume of 315 mL in serum type glass bottles were used to measure degradation. The bottles were incubated at 30⁺/₋₂°C for 14 days. The sludge concentration was 1.5 g/L. Methane production was measured daily using both the Head Space method and the Displacement method. LAS was specifically determined using HPLC.

GLP: Yes [] No [] ? [X]

Test Substance: LAS, sodium salt derived from commercial LAB with the homologue distribution: <C₁₀ 0.7%, C₁₀ 8.4%, C₁₁ 40.9%, C₁₂ 32.5%, C₁₃ 16.6%, and C₁₄ 0.9%. Final molecular weight of 341.5. Average alkyl chain length = C_{11.54}

Remarks: The study determined an IC₅₀ of 40 to 150 mg/L as inhibitory to anaerobic microbial populations. LAS concentrations are usually found in anaerobic digestors at 5-25 g/kg, which is about an order of magnitude lower than the observed IC₅₀ values. The study demonstrates that LAS anaerobic biodegradation does occur under conditions that are not sulphur-limited, using anaerobic digester sludge and specific HPLC methods to measure the loss of parent material.

Reference: Sanz, J.L., Rodriguez, M., Amils, R., Berna, J.L., de Ferrer, J. and Moreno, A. 1999. Anaerobic biodegradation of LAS (Linear Alkylbenzene Sulfonate): Inhibition of the methanogenic process. *La Rivista Holiana Delle Sostanze Grasse*. LXXVI:307-311.

Reliability: 2 Valid with restrictions

(q)

Type: aerobic []; anaerobic [X]

Inoculum: Activated sludge

Medium: Sewage sludge

Degradation: 30-93% after 250 days; 50% in a second 90 day experiment

Results: LAS biodegrades under strict anaerobic conditions.

Method: The standard ECETOC-28 method, which measures the pressure of the biogas as an indicator of anaerobic degradation, was extended to 250 days and supplemented with specific HPLC analysis of LAS concentrations. Testing bottles were prepared in triplicate and monitored for the biodegradation of LAS already present in the sludge without extra LAS being added to the system. A second experiment was also based on the ECETOC-28 method, with several modifications used to further control the anaerobic test conditions.

GLP: Yes [] No [] ? [X]

Test Substance: LAS, as exists in sewage sludge

Remarks: Specific HPLC analysis (loss of LAS) confirms that LAS undergoes primary biodegradation anaerobically in the standard ECETOC-28, although increased gas production (mineralization) was not observed. A redissolution of precipitated adsorbed LAS seemed to occur in the digestion process. The product in liquid solution is probably the fraction being degraded.

Reference: Prats, D., Rodriguez, M., Llamas, J.M., De La Muela, M.A., de Ferrer, J., Morena, A. and Berna, J.L. 2000. The use of specific analytical methods to assess the anaerobic biodegradation of LAS. 5th World CESIO Congress V2:1655-1658, Firenze, Italy.

Reliability: 2 Valid with restrictions

(r)

Type: aerobic [X]; anaerobic []

Inoculum: Trickling filters

Medium: Sewage sludge

Results: ROC supported sulfur-limited growth of *P. putida*. Extensive desulfonation of ROC was observed.

Method: Other studies have confirmed that LAS is completely biodegradable in trickling filters and by-products in commercial LAS (e.g., DATS, SPC) are subject to biotransformation to nondegraded compounds termed refractory organic carbon (ROC). The current study investigated the further desulfonation of ROC by a strain of *Pseudomonas putida*. ROC was generated from commercial LAS, which served as a carbon source, in a trickling filter and isolated by solid-phase extraction. The solution of ROC was then used as a potential sulfur source for the growth of *P. putida*. Experiments were conducted in triplicate at 30°C and cultures were aerated on an orbital shaker. Dissolved Organic Carbon was measured using a total organic carbon analyzer and HPLC.

GLP: Yes [] No [] ? [X]

Test Substance: Commercial LAS (Sirene 113)

Remarks: Earlier work shows that the biodegradation and biotransformation of commercial LAS as a carbon source for growth leads to a residue of sulfonated aromatic compounds, termed refractory organic carbon (ROC), from the synthetic by-products. This study demonstrates that this ROC, after separation from sulfate ion, is utilized extensively as a sulfur source for bacterial growth. The products of desulfonation are expected to be biodegradable.

Reference: Mampel, J., Hitzer, T., Ritter, A. and Cook, A.M. 1998. Desulfonation of biotransformation products from commercial linear alkylbenzene sulfonates. *Environ. Toxicol. Chem.* 17:1960-1963.

Reliability: 2 Valid with restrictions

(s)

Type: aerobic []; anaerobic [X]

Inoculum: adapted []; non-adapted []; other [X]; lake sediments

Concentration: 20, 100, and 200 mg/L

Medium: Water

Results: Degradation occurred under anaerobic conditions when exposed to inoculum obtained from lake sediments. In addition, inocula that were found in aerobic environments such as compost and activated sludge from a wastewater treatment plant also showed capability of anaerobic degradation of LAS.

Method: Tests were performed in batch serum vials under anaerobic conditions. The vials were filled with the appropriate pH 7 medium, autoclaved at 140°C for 30 minutes, and inoculated with 5 to 10% of the respective inocula. Inocula originated from several different natural environments and from anaerobic reactors. LAS was added at three different concentrations (20, 100, and 200 mg/L) plus three sets of controls, all in triplicate. Incubation time was 2 months.

GLP: Yes [] No [] ? [X]

Test Substance: Mixture of LAS with an alkyl chain length of 9 to 13 units; likely average alkyl chain length = C_{11.6}

Remarks: This paper indicates qualitatively that LAS undergoes anaerobic degradation, but no quantitative results are presented.

Reference: Angelidaki, I., Mogenen, A.S. and Ahring, B.K. 2000b. Degradation of organic contaminants found in organic waste. *Biodegradation.* 11:377-383.

Reliability: 2 Valid with restrictions

(t)

Type: aerobic []; anaerobic [X]

Inoculum: Activated sludge

Concentration: 100 mg/L

Medium: water []; water-sediment []; soil []; sewage treatment [X]

Degradation: Transformation of C₁₂ LAS occurred under anaerobic conditions. The degree of transformation varied between 14 to 25%.

Methods: Two lab-scale continuous stirred tank reactors (CSTR) were set up with automatic, semi-continuous feeding and were run under mesophilic conditions (37°C) with a hydraulic retention time of 15 days. The reactors were started with anaerobic stabilizer sewage sludge and operated for several months before the experiment started. The feed was diluted

sludge at a total solids concentration of 20 g TS/L. The sludge was spiked with C₁₂ LAS at a concentration of 100 mg/L and the two reactors were operated similarly for 36 days. After this period, the LAS concentration in reactor 1 was increased to 268 mg/L, while for reactor 2 the influent TS was decreased to 11.4 g TS/L, and both reactors continued to operate for a total of 90 days (including the original 36 days).

GLP: Yes No ?

Test Substance: C₁₂ LAS (pure homologue)

Remarks: A clear correlation was shown between degradation of organic matter contained in the sludge and anaerobic degradation of LAS, giving an increase in transformation with the higher the reduction of organic matter. Transformation was limited by bioavailability due to sorption of LAS (i.e., only the bioavailable fraction of LAS is transformed by anaerobic digestion). When the reduction degree of the organic matter increased from 22% to 28%, the transformation degree of C₁₂ LAS increased from 14% to 20%. Decreasing the total solids concentration of the influent sludge or increasing the spiked concentration of C₁₂ LAS did not significantly alter the degree of LAS transformation.

Reference: Angelidaki, I., Haagsen, F. and Ahring, B.K. 2000a. Anaerobic transformation of LAS in continuous stirred tank reactors treating sewage sludge. 5th World CESIO Congress. V.2:1551-1557, Firenze, Italy.

Reliability: 2 Valid with restrictions

(u)

Type: aerobic ; anaerobic

Medium: coastal sea water

Concentration: 5 mg/L related to test substance

Results: LAS primary degradation half-lives ranged from 3.4 to 13.8 days, with 4-9 days being the most frequent values.

Method: Coastal sea water from the Mediterranean Sea was collected from three areas in Spain (Barceloneta, Ebro delta, and Sant Feliu de Guixols, Girona). Samples of 1.5- L were placed in 3-L flasks and incubated in the dark at 20°C with orbital shaking (100 rpm) for 30 days. Viable bacteria were determined by plate counts on marine agar media, while total bacteria were determined by flow cytometry after SYTO-13 staining. LAS degradation was monitored by HPLC. A reference substance was not used. LAS quantification was based on an external standard.

GLP: Yes No ?

Test Substance: C₁₀₋₁₄ LAS, activity 66.62%; average alkyl chain length = C_{11.7}

Remarks: In most cases, sea water samples showed a similar evolution of bacterioplankton over time, characterized by three phases: (a) a progressive increase in bacterial density; (b) a later decrease; and (c) a fluctuating stationary phase. Bacterioplankton degraded the LAS by growing to populations with a high percentage of viable bacteria. The bacteria were readily grazed by protozoa, preventing anomalous high bacterial growth and ensuring the later channeling of LAS carbon to upper trophic levels.

Reference: Vives-Rego, J., Lopez-Amoros, R., Guindulain, T., Garcia, M.T., Comas, J., and Sanchez-Leal, J. 2000. Microbial aspects of linear alkylbenzene sulfonate degradation in coastal water. *Journal of Surfactants and Detergents*. 3:303-308.

Reliability: 2 Valid with restrictions

(v)

Type: Respirometer

Method: Degradation of LAS in Ohio River water collected below the discharge of a municipal wastewater treatment plant (Muddy Creek, OH) was measured in an electrolytic respirometer. Background LAS concentrations were less than 0.5 mg/L. Oxygen consumption over time was determined at five LAS concentrations (5, 10, 20, 40, and 80 mg/L) plus a control until plateau values were reached. The maximum initial rates of oxygen uptake were calculated based on methods described in Larson and Perry (1981).

Results: LAS degradation was not affected in river water until the LAS concentration exceeded 10 mg/L. The degradation was partially affected at 20 mg/L but was not completely inhibited until 40 mg/L.

Substance: LAS; average chain length $C_{11.6}$.

Remarks: The level at which inhibition of degradation was complete (40 mg/L) is significantly higher than the levels observed in model ecosystem studies conducted by these researchers [see 4.7 (h) and (i)].

Reference: 1) Larson, R.J. and Maki, A.W. 1982. Effect of LAS on the structure and function of microbial communities in model ecosystems. Aquatic Toxicology and Hazard Assessment: Fifth Conference, ASTM STP 766, Pearson, J.G., Foster, R.B., and Bishop, W.E., Eds., American Society for Testing and Materials, pp. 120-136.
2) Maki, A.W. 1981. A laboratory model ecosystem approach to environmental fate and effects studies. Unpublished Internal Report, Environmental Safety Department Procter & Gamble Company, Cincinnati, Ohio.
3) Larson, R.J. and Perry, R.L. 1981. Water Research 15:697-702.

Reliability: 2 Valid with restrictions

(w)

Type: aerobic [X]; anaerobic []

Substance: LAS

Remarks: The biodegradation of LAS has been thoroughly studied for its primary and total degradation and catabolism. It is mineralized biologically to form carbon dioxide, water and sulphate. Using UV spectroscopic analysis, Swisher (1987) and others showed the aromatic ring to be degradable up to 80%. Using the more reliable tracer technique with ^{14}C -ring labelled LAS, it was determined that degradation of the ring is predominantly between 50 and 80%. Further studies have shown that LAS degradation proceeds through oxidative conversion of the methyl groups of the alkyl chain into a carboxyl group (ω -oxidation), oxidative shortening of the alkyl chain by 2-carbon units (β -oxidation), oxidative ring splitting, then cleavage of the carbon-sulfur bond. This process forms sulfophenyl carboxylates (SPCs) as biodegradation intermediates. The first detectable degradation product of LAS is ω -carboxylate. For example, Huddleston and Allred (1963) detected sulfophenyl decanoic acid as a catabolite of 2-benzenedecylsulfonate. Oxidative degradation of the alkyl chain begins as soon as LAS has been converted into sulfophenyl carboxylic acid. The principal degradation pathway is β -oxidation. The rate of biodegradation is inversely related to the distance between the terminal alkyl-methyl group and the point of benzene ring attachment.

Simple branching does not impair the oxidation of alkylbenzene, though more complex branching does decrease the rate.

Reference: 1) Swisher, R.D. 1987. Surfactant Biodegradation, second edition. Surfactant Science Series, Volume 18. Marcel Dekker, Inc. New York.
2) Schoeberl, P. 1989. Basic principles of LAS biodegradation. Tenside Surf. Detergents 26:86-94.
3) Huddleston, R.L. and Allred, R.C. 1963. Microbial oxidation of sulfonated alkylbenzenes. Dev. Ind. Microbiol. 4:24-38.

Reliability: 2 Valid with restrictions

(x)
Type: aerobic [X]; anaerobic []
Method: Dialkyltetralin sulfonates (DATS) and LAS with single methyl branching on the alkyl chains (iso-LAS) are minor components in commercial LAS. In this study, DATS and iso-LAS were synthesized and exposed to simulated activated sludge, soil, and receiving water environments. In addition, the effluents coming from activated sludge treatment, which contained biodegradation intermediates, were exposed to simulated receiving water environments. Radiolabeled LAS, DATS and iso-LAS were used and all samples were analyzed using chemical-specific HPLC procedures. Surface soils were collected at three locations to represent "pristine" soil, sludge-amended soil, and gray water contaminated soil from the top of a percolation bed that receives surface applications of laundry water from a Laundromat. All samples were screened to remove vegetation, rocks and debris, and mixed with a mineral salts medium containing the test substance. Sediment samples were collected from the upper inch of a small stream that received effluent from a domestic wastewater treatment plant. Periphyton samples were collected as rocks coated with heavy growth from the same stream locations as the water and sediment samples. Each test system consisted of duplicate test flasks and a control flask. Tests lasted at least 30 days. For assessing biodegradation, the porous pot method was used in a simulated wastewater activated sludge modified from ASTM test method E1798-96. A 21-day acclimation phase was followed by a 15-day test phase in which radioactivities in CO₂, liquids and solids, and effluent total suspended solids and COD were determined each day. Radiochemical recoveries for the porous pot test were calculated. For the die-away tests with porous pot effluents, the combined effluents from individual units were tested for mineralization of radiolabeled parent and intermediate compounds. All tests were run at least 30 days and the radioactivities measured at the end of each test.

Results: Results indicate that radiolabeled DATS and iso-LAS is mineralized by indigenous microbial populations in laboratory simulations of aquatic and soil environments. Half-lives ranged from 2 to 20 days. In addition, upon exposure to laboratory activated sludge treatment, most iso-LAS compounds showed >98% parent compound removal, extensive mineralization (>50%), and 79-90% ultimate biodegradation. Activated sludge treatment of DATS resulted in >98% removal, 3-12% ultimate biodegradation, and apparent formation of carboxylated biodegradation intermediates that accounted for 88-97% of the original material. These intermediates continued to mineralize in simulated receiving water and soil

	environments at rates similar to that of sulfophenyl carboxylate (SPC) intermediates of a standard LAS.
Test Substances:	¹⁴ C-benzene ring labeled C ₁₂ LAS (97.5% radiochemical purity); ¹⁴ C-benzene ring labeled iso-LAS of the following types (IA, 97.8% purity; IB, 77.6% purity; IIA, 94.7% purity; IIB, 97.5% purity); ¹⁴ C-benzene ring labeled DATS (97.3% purity), plus the non-labeled versions of the same.
Reference:	Nielsen, A.M., Britton, L.N., Beall, C.E., McCormick, T.P. and Russell, G.L. 1997. Biodegradation of coproducts of commercial linear alkylbenzene sulfonate. Environ. Sci. Technol. 31:3397-3404.
Reliability:	2 Valid with restrictions
(y)	
Type:	aerobic [X]; anaerobic []
Method:	OECD 301E. The study was designed to investigate the biodegradation of a relatively high iso-branched form of commercial LAS. The test was a prolonged batch-biodegradation experiment in which the material is kept "alive" for 80 days and in which the test compound present in a mineral salts medium is the sole carbon source. An enriched level of bacterial biomass, three times the amount recommended, was added at the test start using an inoculum obtained from the settled supernatant slurry solution of a fertile soil, without any previous exposure to the test compound. LAS was maintained by adding about 0 mg/L of fresh substance every four days for 80 days. After 80 days the test solution was sampled, centrifuged, sterilized with HgCl ₂ solution and analyzed with a chemical specific HPLC method with fluorescence detection.
Results:	Results indicate a residual LAS amount of 1.5 mg/L and SPC intermediate amount of 28.7 mg/L at the end of the 80 day study. Four distinct SPCs originating from the linear components of LAS were formed from the biodegradation experiment, and made up most of the organic residue. No evidence of structures related to the iso-branched material was found in the residue, therefore no accumulation of these materials is indicated. The iso-branched component of LAS and the corresponding SPCs mineralized at rates as fast as the linear components.
Test Substances:	Commercial LAS (HF type) with a C ₁₀ -C ₁₃ alkyl chain and a linearity of about 93%, with a low DATS content (<0.5%) and a relatively high iso-LAS content (6.5%).
Reference:	Cavalli, L., Cassani, G., Lazzarin, M., Maraschin, C., Nucci, G., and Valtorta, L. 1996. Iso-branching of linear alkylbenzene sulphonate (LAS). Tenside Surf. Det. 33:393-398.
Reliability:	2 Valid with restrictions
(z)	
Type:	aerobic [X]; anaerobic []
Inoculum:	adapted []; non-adapted [X]; activated sludge
Concentration:	34.3 mg/L related to COD []; DOC [X] test substance []
Medium:	water [X]; water-sediment []; soil []; sewage treatment []
Degradation:	85% after 29 days
Results:	readily biodeg. [X]; inherently biodeg. []; under test condition no biodegradation observed [], other []
Kinetic:	0 day = -1% 2 day = -2% 5 day = 22%

9 day = 52%
 12 day = 70%
 14 day = 70%
 21 day = 78%
 28 day = 83%
 29 day = 85%

Method: OECD Test Guideline 301B and EC Directive 92/69/EEC C.4-C. Modified Sturm Test.. The test substance was added to a defined liquid mineral medium which was inoculated with an activated-sludge inoculum and aerated at 19.7-21.9°C (mean 21.1°C). The inoculum used was activated non-adapted sludge from the Marl-Ost municipal sewage treatment plant. The inoculum had a bacterial count of 81×10^4 CFU/mL as determined by the Koch pour-plate method. The CO₂ released was bound in the form of sodium carbonate in sodium hydroxide solution. Samples were collected and analyzed in duplicate for bound CO₂ by TIC analysis after 0, 2, 5, 9, 12, 14, 21, 28 and 29 days. Sodium benzoate was used as a suitable control substance to monitor the activity of the inoculum. On the 29th day, residual dissolved CO₂ was expelled by acidification.

GLP: Yes No ?

Test substance: Marlon A 365 WEL 6859 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}; Activity: 65%

Remarks: LAS is readily biodegradable. The 10-day window criterion was fulfilled. The control substance (sodium benzoate) showed 89% degradation after 29 days. This is a key study for ready biodegradability (see SIAR Table 4).

Reference: Enste-Diefenbach, R.. 2002. Marlon A 365 WEL 6859: Determination of biodegradability in the modified Sturm test.. Infracor GmbH Analytical Technical Services, Report ST-204/02.

Reliability: 1 Valid without restriction

3.6 BOD₅, COD OR RATIO BOD₅/COD

BOD₅

Method: DIN 38H03, Part 1. Bestimmung des biochemischen Sauerstoffbedarfs in 5 Tagen nach den Verduennungsprinzip.

Value: <10 mg O₂/g for both Marlon A 350 and Marlon A 375

GLP: Yes No ?

COD

Method: DIN 38H03, Part 1

Values: 1151 and 1760 mg O₂/g for Marlon A 350 and Marlon A 375, respectively.

GLP: Yes No ?

Ratio BOD₅/COD:

Remarks: < 0.005
 Marlon A 350 (C₁₀₋₁₃ LAS, average chain length = 11.6, 50% a.i.)
 Marlon A 375 (C₁₀₋₁₃ LAS, average chain length = 11.6, 75% a.i.)
 The extended term BOD determinations yield 60 to 70% of COD. The substance is degradable.

Reference: European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts (CAS #68411-30-3). Year 2000 CD-ROM edition, citing data by Huels AG Marl.

Reliability: 4 Not assignable

3.7 BIOACCUMULATION

(a)
 Species: *Pimephales promelas* (fish, fresh water)
 Exposure period: 48, 168, 192 hours
 Temperature: Per Protocol
 Concentration: 2.7 and 4.1 µM
 BCF: Values of Steady-State Bioconcentration Factor (BCF_{ss}) and Average Length of Alkyl Chain (n_{C,Av}) are shown in the following table.

expt	comp*	BCF _{ss} L/kg	n _{C,Av}
A	C ₁₀₋₂	1.7	10.8
	C ₁₁₋₂	5.8	
	C ₁₂₋₂	47.6	
	C ₁₃₋₂	353.8	
B	C ₁₁₋₅	6.1	11.7
	C ₁₂₋₂	99.1	
	C ₁₂₋₅	10.0	
	C ₁₃₋₅	34.0	
C	C ₁₁₋₅	9.8	11.4
	C ₁₂₋₂	168.4	
	C ₁₂₋₃	42.1	
	C ₁₂₋₆	31.9	
D	C ₁₀₋₂	6.0	10.6
	C ₁₁₋₂	31.9	
	C ₁₂₋₂	211.5	
	C ₁₃₋₂	987.2	
	C _{10-in}	3.0	
	C _{11-in}	9.1	
	C _{12-in}	29.9	
C _{13-in}	112.4		

*In the format C_{n-m}, n and m are the length of the alkyl chain and the position at which the sulfophenyl moiety is substituted to the alkyl chain, respectively.

Elimination: Yes No ?
 Method: OECD 305 E. The exposure phase in Experiment A was 48-hours. The exposure phase in Experiments B-D ranged from 168 to 192 hours. Due to the rapid equilibrium demonstrated in these studies, a longer exposure period was not needed. Fish were then transferred to untreated water for the depuration phase (duration not stated).
 Type of test: calculated ; measured
 static ; semi-static ; flow-through
 GLP: Yes No ?
 Test substance: LAS (C₁₀₋₁₃), tested individually and as mixtures, activity: >97.4%

Remarks: As shown in the table, BCF values ranged between 2-1000 L/kg. Experiments A, B and D showed that BCFs increase with increasing alkyl chain length for a given isomer. In addition, the results of Experiments B and C demonstrate that the closer the p-sulfophenyl moiety is positioned to the terminal carbon of the alkyl chain, the higher the BCF. However, alkyl chain length has a much bigger effect than does the phenyl position. To address differences in composition of mixtures, bioconcentration potential was calculated for a mixture typical of LAS in European detergent formulations (C₁₀ 12%, C₁₁ 29%, C₁₂ 34%, C₁₃ 24%; average alkyl chain length = C_{11.6}) and a mixture typical of LAS in filtered Mississippi river water (C₁₀ 45%, C₁₁ 23%, C₁₂ 23%, C₁₃ 2%; average chain length = C_{10.8}), using BCF values for the individual components. This calculation of BCF for the typical mixtures was done using the following equation developed in the above testing:

$$(\sum C_{f,i} / \sum C_{w,i})_{rel} = \sum (\theta_{i,w} \cdot BCF_{i,rel})$$

The BCFs were 87 L/kg for a standard mixture typical of LAS in European detergent formulations (average alkyl chain length = C_{11.6}) and 22 L/kg for a representative environmental sample (filtered Mississippi river water, average alkyl chain length = C_{10.8}), indicating that the bioconcentration potential of LAS is low and is decreased by environmental processes such as biodegradation and absorption, which reduce aquatic concentrations.

Reference: Tolls, J., Haller, M., DeGraaf, I., Thijssen, M.A.T.C. and Sijm, D.T.H.M. 1997. Bioconcentration of LAS: Experimental determination and extrapolation to environmental mixtures. Environ. Sci. Technol. 31:3426-3431.

Reliability: 2 Valid with restrictions

(b)

Species: *Lepomis macrochirus*

Exposure Period: 21 days

Temperature: 17⁺-1^oC

Concentration: 0.5 mg/L

BCF: 104 (whole body); 36 (muscle)

Elimination: Yes

Method: Bluegill sunfish (avg wt. 4.0 g; avg length 68 mm) were placed in a 60 liter aquarium (375 fish total) and maintained for 21 days. A second aquarium held 100 control fish. Fish were fed daily with a dry pelleted trout chow ration of approximately 2% of body weight. Water samples were removed periodically for radiometric analysis of ¹⁴C-labeled LAS. Four fish were removed on each of days 1, 2, 3, 5, 7, 9, 11 and 14 for radiometric analysis.

Type of test: calculated ; measured ; static ; semi-static ; flow-through

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS (CAS #68411-30-3) with the following alkyl chain length distribution: C₁₁ 45%, C₁₂ 36.5%, C₁₃ 18.5%. Average chain length was 11.7 and molecular weight was 344.

Remarks: The site of greatest concentration was the gall bladder with a BCF of 5000, based on total radiolabeled materials. The BCFs for liver, gills and viscera, remaining carcass, and blood ranged from 64 to 283. Clearance

of radiolabeled materials was rapid with half-lives of 2 to 5 days. However, no quantitative conclusions specific to LAS can be drawn from these data, as total radiolabeled materials were measured, and these likely include LAS metabolites.

Reference: Kimerle, R.A., Macek, K.J., Sleight, B.H. and Burrows, M.E. 1981. Bioconcentration of linear alkylbenzene sulfonate (LAS) in bluegill (*Lepomis macrochirus*). *Wat. Res.* 15:251-256.

Reliability: 2 Valid with restrictions

3.8 ADDITIONAL REMARKS

A. Sewage treatment

(a)

Results: LAS removal in sewers due to biodegradation can reach as high as 50% of the total LAS load when the sewer system is properly aerated.

Method: Integrated composite samples were collected in November and December 1988 from the sewer system of Estepona (Malaga), Spain. LAS was analyzed using a specific HPLC technique.

Remarks: This study demonstrates that significant biodegradation of LAS occurs prior to reaching wastewater treatment plants. Additional removal (up to >95%) occurs in the plants themselves.

Reference: Moreno, A., de Ferrer, J. and Berna, J.L. 1990. Biodegradability of LAS in a sewer system. *Tenside Surf. Det.* 27:312-315.

Reliability: 2 Valid with restrictions

(b)

Results: The average removal rate for LAS in activated sludge treatment was > 99%. A lower and more variable rate was observed in trickling filter treatment plants with an average removal of 82% for LAS.

Remarks: Samples were collected from six trickling filter and four activated sludge treatment plants located in the midwestern United States.

Reference: McAvoy, D.C., Dyer, S.D., Fendinger, N.J., Eckhoff, W.S., Lawrence, D.L. and Begley, W.M. 1998. Removal of alcohol ethoxylates, alkyl ethoxylate sulfates, and linear alkylbenzene sulfonates in wastewater treatment. *Environ. Toxicol. Chem.* 17:1705-1711.

Reliability: 2 Valid with restrictions

(c)

Results: The model predicted average removal of LAS from wastewater treatment plants is 99.2%. Predicted 90th-percentile concentrations at 1,000 m downstream from the sewage outfall, based on actual measured raw sewage concentrations and actual measured effluent calculations, ranged from 3.7 to 9.2 µg/L for different predicted instream removal rates.

Method: Modeling was conducted to predict the 90th-percentile environmental concentration (PEC) of LAS and other detergent substances in aquatic environments in the Netherlands. Inputs included emissions data, prediction of raw sewage concentration and initial material characterization. Model predictions included the removal of LAS in wastewater treatment plants, concentrations in surface waters, and prediction of the 90th-percentile concentrations.

Remarks: The authors emphasize that to provide a fate assessment adequate for regulatory purposes, a need clearly exists for a fundamental interplay between monitoring, laboratory data, and these predictive models. This study is part of an extensive monitoring program executed jointly by the Dutch Soap Association (NVZ) and the Dutch Ministry of Housing, Spatial Planning and the Environment (VROM). Monitoring data from this program can be found in Matthijs et al. 1999.

Reference: Feijtel, T.C.J., Struijs, J., and Matthijs, E. 1999b. Exposure modeling of detergent surfactants – Prediction of 90th-percentile concentrations in the Netherlands. *Environmental Toxicology and Chemistry* 18:2645-2652.

Reliability: 2 Valid with restrictions

(d)

Results: The average concentration of LAS in the treated sewage of the sum wastewater treatment plants was 39 µg/L. The average total removal of LAS was 99.2%.

Method: Twenty four hour flow proportional samples of raw, settled, and treated sewage were collected by automatic samples during three consecutive days at seven sewage treatment plants in the Netherlands. All samples were collected between April and July 1994 and analyzed for traditional sewage treatment plant water quality parameters. Samples for the analysis of LAS and other surfactants were taken every 15 minutes (hourly composites) using a time proportional automatic sampler. The LAS in these samples was analyzed using an HPLC method with fluorescence detection.

Remarks: This study is part of an extensive monitoring program executed jointly by the Dutch Soap Association (NVZ) and the Dutch Ministry of Housing, Spatial Planning and the Environment (VROM). The authors indicate that field studies suggest that in-sewer removal can play a significant role in reducing the concentrations of surfactants entering the sewage treatment plant.

Reference: Matthijs, E., Holt, M.S., Kiewiet, A., and Rijs, G.B.J. 1999. Environmental monitoring for linear alkylbenzene sulfonate, alcohol ethoxylate, alcohol ethoxy sulfate, alcohol sulfate, and soap. *Environmental Toxicology and Chemistry* 18:2634-2644.

Reliability: 2 Valid with restrictions

(e)

Methods: LAS was monitored seasonally for one year (winter, spring, summer) in a small river in Spain receiving untreated sewage from a non-industrial village (Caserras). Sampling was carried out during November 1994, May 1995 and July 1995. Grab samples were collected in the morning, afternoon and evening at five sampling sites representing the raw sewage discharge, a pre-discharge location on the river, and three downstream locations on the river (1.5, 3.0 and 4.8 km downstream of the discharge).

Results: Seasonal differences were observed in biodegradation, with total LAS removals (dissolved and adsorbed) at 4.8 km downstream of 31.8%, 95.5% and 98.3% for winter, spring and summer respectively.

Remarks: The seasonal differences in biodegradation are explained by hydraulic conditions. River flow rates are much greater in winter (75 m³/min) versus spring (4.5 m³/min) and summer (0.2 m³/min), which results in a much reduced hydraulic retention time (and thus less contact time for

biodegradation) in winter of 1.6 hrs compared to 26.6 hrs in spring and 25 days in summer. Overall, even in situations of direct discharge of untreated sewage, LAS biodegradation of >98% can be expected provided that the receiving water stream has adequate hydraulic conditions.

Reference: de Ferrer, J., Moreno, A., Vaquero, M.T. and Comellas, L. 1997. Monitoring of LAS in direct discharge situations. *Tens. Surfactants Det.* 34:278-283.

Reliability: 2 Valid with restrictions

(f)

Results: LAS removal of 98-99% and biodegradation of 80-84% was observed. Sulfophenyl carboxylates (SPC) were found only in water and not the absorbed phases (sludge).

Remarks: This study was conducted to specifically study LAS biodegradation in real WWTP conditions in Italy. LAS data was obtained by HPLC of influent, effluent, dissolved waters and sludges to reach a complete mass balance.

Reference: 1) Cavalli, L., Gellera, A., Lazzarin, A., Nucci, G.C., Romano, P., Ranzani, M. and Lorenzi, E. 1991. Linear alkylbenzene sulphonate removal and biodegradation in a metropolitan plant for water treatment. *Riv. Ital. Sostanze Grasse* 68:75-81.

2) Cavalli, L., Gellera, A. and Landone, A. 1993. LAS removal and biodegradation in a wastewater treatment plant. *Environmental Toxicology and Chemistry* 12:1777-1788.

Reliability: 2 Valid with restrictions

B. Other information

(a)

Remarks: A significant number of additional literature articles report data on the environmental fate of LAS. An additional bibliography of literature citations for LAS can be found in an Appendix to this dossier.

(b)

Type of Measurement: Background ; At contaminated site ; Other

Medium: soil

Remarks: Where surfactant and hydrophobic organic compounds (HOCs) co-exist in soil-water systems there are a number of possible interactions which can occur simultaneously: 1) distribution of surfactant between monomeric, hemimicellar and miscellar forms, 2) competition for hydrophobic adsorption sites between the surfactant and HOC and 3) partitioning of HOC among soil hydrophobic adsorption sites, surfactant micelles and hemimicelles. The interaction of HOCs with surfactant monomers is usually very weak and insignificant. At concentrations where micelles and hemimicelles are present interactions can take place. Sorbed HOCs can be solubilised by free micelles, resulting in mobilisation. HOCs in solution are in equilibria between sorption onto hydrophobic adsorptive sites on the soil, partitioning into hemimicelles – both resulting in immobilisation, and partitioning into free micelles. Whether the HOCs are previously sorbed onto soil or are in solution, partitioning into micelles, hence mobilisation, is favoured by increasing surfactant concentration. A model has been put forward describing the

effect of non-ionic surfactant on the distribution of HOC in a soil-water system. In simple terms the model illustrates that sorbed surfactant molecules tend to increase HOC sorption onto soil by increasing its fractional organic carbon content, and free surfactant tends to decrease sorption by increasing the apparent aqueous solubility of the HOC.

The biodegradation of HOCs in soil can be enhanced by surfactants due to enhanced solubility in the presence of micelles. In some cases however, biodegradation appears to be inhibited by micelles forming a barrier to the degrading organism. Any such inhibition is unlikely to be prolonged due to the biodegradable nature of most modern surfactants. In fact, it is very unlikely that micelles would be present in sludge-amended soils due to the low concentration of surfactants.

Although there is evidence that surfactants can effect the fate and behaviour of HOCs in soil, the potential for detergent ingredients to cause significant effects is limited due to the relatively low concentrations found compared with critical micelle concentrations (CMCs). In addition, the effective CMC in environments such as soil and sediments is generally much higher than in clean water systems. Typical soil concentrations of LAS, the most heavily used surfactant in domestic detergents, are significantly lower than those required to produce micelles in pore water. Therefore, it is unlikely that surfactants present in domestic detergents will contribute significantly to the mobilisation of HOCs in sludge-amended soil.

Reference:

Haigh, S.D. 1996. A review of the interaction of surfactants with organic contaminants in soil. *The Science of the Total Environment* 185:161-170.

4. ECOTOXICITY

4.1 ACUTE/PROLONGED TOXICITY TO FISH

(a)

Type of Test: static []; semi-static []; flow-through [X]
Open-system [X]; closed-system []

Species: *Lepomis macrochirus* (Fish, fresh water)

Exposure Period: 96 hours

Results: $LC_{50} = 1.67$ mg/L

Analytic Monitoring: Yes [X]; No []; ? []

Method: USEPA (1975) Stock solutions were made up in deionized water without the use of a solvent and metered into diluter chambers by peristaltic pump. Mortality was analyzed by four methods: (a) the toxic unit (TU) concept, (b) the additive index, (c) concentration addition, and (d) response addition.

GLP: Yes []; No [X]; ? []

Test Substance: $C_{11,8}$ LAS; Average molecular weight 345; 27.3% active; Alkyl chain Composition: C_{10} 9.5%, C_{11} 29.2%, C_{12} 37.7%, C_{13} 19.0%, C_{14} 4.9%

Remarks: Average size of individual fish 1.1 g, mean standard length 4.2 cm; water hardness 137 mg/L Ca CO_3 ; temperature $20 \pm 2^\circ C$; dissolved oxygen 8.4-9.6 mg/L; pH 7.3-8.1. Ten randomly selected fish were exposed to each of the five concentrations and the control. A 16:8 (light:dark) photoperiod was maintained. Fish were not fed during the study. Mortality was recorded at 1, 3, and 5 hours after test initiation and then daily until test termination. Samples were collected at the beginning and end of the study and analyzed for the test substance. Results reported are mean measured concentrations. The LC_{50} value reported represents the lowest value for this species. In addition to the studies conducted on LAS, the anionic LAS was also tested in binary and ternary equitoxic or equimolar mixtures with non-ionic C_{14-15} linear alkyl ethoxylate (AE) and cationic C_{12-14} MDAC. These mixture results are not reported in this robust summary. This is a key study for aquatic toxicity to fish (see SIAR Table 10).

Reference: Lewis, M.A. and Perry, R.L. 1981. Acute toxicities of equimolar and equitoxic surfactant mixtures to *Daphnia magna* and *Lepomis macrochirus*. Aquatic Toxicology and Hazard Assessment: Fourth Conference, ASTM STP 737, D.R. Branson and K.L. Dickson, Eds., American Society for Testing and Materials, pp. 402-418.

Reliability: 2 Valid with restrictions. The studies are very well documented in this peer-reviewed publication.

(b)

Species: *Lepomis macrochirus* and *Pimephales promelas*

Results: LC_{50} values ranged from 1.67 to 7.7 mg/L for *L. macrochirus* (10 records)
 LC_{50} value for *P. promelas* = 4.1 mg/L (1 record) See study (q) below for robust summary

Test Substance: C_{10-13} LAS (CAS #68411-30-3)

Remarks: A total of 18 fish studies for these two species were reviewed by HERA in 2004. Seven of these studies were rejected because the test material was not commercial LAS, the study deviated significantly from standard protocols, or non-standard endpoints were measured. The remaining

eleven studies were evaluated for reliability and the results reflect the range of acute LC₅₀ values obtained for the most commonly tested fish species. These studies are tabulated and discussed further in the SIAR and SIAP in a weight-of-evidence approach. A robust summary for the study with the lowest LC₅₀ value was prepared (see Lewis and Perry 1981 above).

Reference: HERA-LAS team, May 2004; see SIAR, Annex 2.
Reliability: 4 Not assignable. This study is given a reliability score of 4 because it is a summary evaluation of a series of studies conducted by other researchers.

(c)

Species: *Lepomis macrochirus*
Results: LC₅₀ = 3.0 mg/L (geometric mean of 88 records)
Test Substance: C₁₀₋₁₄ LAS (all LAS in range, including data for individual homologues)
Remarks: Mean LC₅₀ for bluegill sunfish was derived from a total of 88 records compiled from the BKH (1993) literature review.
Reference: van de Plassche, E.J., de Bruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J. and Belanger, S.E. 1999. Predicted no-effect concentrations and risk characterization of four surfactants: LAS, AE, AES, and soap, Environ. Toxicol. Chem. 18:2653-2663.
Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(d)

Species: *Pimephales promelas*
Results: LC₅₀ = 3.2 mg/L (geometric mean of 35 records)
Test Substance: C₁₀₋₁₄ LAS (all LAS in range, including data for individual homologues)
Remarks: Mean LC₅₀ for fathead minnow was derived from a total of 35 records compiled from the BKH (1993) literature review. The range of LC₅₀ values (0.40-100 mg/L) is very large due to the range of materials tested (including many materials not representative of commercial LAS) and the differences in test design (not necessarily following standard guidelines). It is unclear which values within the range refer to the commercial LAS products and the individual records are not available for validation.
Reference: van de Plassche, E.J., de Bruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J. and Belanger, S.E. 1999. Predicted no-effect concentrations and risk characterization of four surfactants: LAS, AE, AES, and soap, Environ. Toxicol. Chem. 18:2653-2663.
Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(e)

Species: *Leuciscus idus melanotus*
Results: LC₅₀ = 2.9 mg/L (geometric mean of 11 records)
Test Substance: C₁₀₋₁₄ LAS (all LAS in range, including data for individual homologues)
Remarks: Mean LC₅₀ for golden orfe was derived from a total of 11 records compiled from the BKH (1993) literature review.
Reference: van de Plassche, E.J., de Bruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J. and Belanger, S.E. 1999. Predicted no-effect

concentrations and risk characterization of four surfactants: LAS, AE, AES, and soap, Environ. Toxicol. Chem. 18:2653-2663.
Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(f)

Species: *Carassius auratus* (goldfish)
Oncorhynchus mykiss (rainbow trout)
Oryzias latipes (medaka)
Poecilia reticulata (guppy)
Results: LC₅₀ (*C. auratus*) = 9.5 mg/L (46 records)
LC₅₀ (*O. mykiss*) = 3.0 mg/L (10 records)
LC₅₀ (*O. latipes*) = 13 mg/L (5 records)
LC₅₀ (*P. reticulata*) = 3.8 mg/L (9 records)
Test Substance: C₁₀₋₁₄ LAS (all LAS in range, including data for individual homologues)
Remarks: Geometric mean LC₅₀ values for the number of records listed for each species. The interspecies variation decreases considerably when the geometric mean value per species is calculated.
Reference: van de Plassche, E.J., de Bruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J. and Belanger, S.E. 1999. Predicted no-effect concentrations and risk characterization of four surfactants: LAS, AE, AES, and soap. Environ. Toxicol. Chem. 18:2653-2663.
Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(g)

Species: Fish species (all marine)
Results: LC₅₀ = 1.58 mg/L (6 records; SD = 0.16)
Test Substance: LAS; average alkyl chain length C_{11.7-12.0}
Remarks: LC₅₀ is geometric mean of 6 records compiled from literature reviews. Geometric mean LC₅₀ for all taxa (36 records; SD = 0.79) was 4.36 mg/L.
Reference: Temara, A., Carr, G., Webb, S., Versteeg, D., and Feijtel, T.C.J. 2001. Marine risk assessment: Linear alkylbenzenesulphonates (LAS) in the North Sea. Mar. Poll. Bulletin 42:635-642.
Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(h) (Zebra fish)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system
Species: *Brachydanio rerio* (Fish, fresh water)
Exposure period: 96 hour
Results: LC₅₀ = 5.1 mg/L
Analytical monitoring: Yes No ?
Method: OECD Guide-line 203 "Fish, Acute Toxicity Test"
GLP: Yes No ?
Test substance: C₁₀₋₁₃ LAS (CAS #68411-30-3)

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. The submitter (Huels AG) judged the study quality to be good. Water hardness: 310 mg/L CaCO₃; tap water diluent; 25 °C; adult fish tested.

Reference: European Commission. 2000f. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, AT/FU/80/90.

Reliability: 4 Not assignable. The original study was not available for review.

(i)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system

Species: *Brachydanio rerio* (Fish, fresh water)

Exposure period: 96 hour

Results: LC₅₀ = 7.8 mg/L

Analytical monitoring: Yes No ?

Method: ISO 7346/1-3

GLP: Yes No ?

Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = 11.6

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Concentration of test substance related to MBAS. LC₀ and LC₁₀₀ = 5.6 and 11 mg/L, respectively

Reference: European Commission. 2000b. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).

Reliability: 4 Not assignable. The original study was not available for review.

(j)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system

Species: *Brachydanio rerio* (Fish, fresh water)

Exposure period: 14 day

Results: NOEC = 2 mg/L
LOEC = 8 mg/L

Analytical monitoring: Yes No ?

Method: OECD Guide-line 204 "Fish, Prolonged Toxicity Test: 14-day Study"

GLP: Yes No ?

Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = 11.6

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Concentration of test substance related to MBAS.

Reference: European Commission. 2000b. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).

Reliability: 4 Not assignable. The original study was not available for review.

(k) (Rainbow trout)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system

Species: *Salmo gairdneri* (Fish, estuary, fresh water)

Exposure period: 96 hour

Results: LC₅₀ = 5.8 mg/L

Analytical monitoring: Yes No ?
Method: OECD Guide-line 203 "Fish, Acute Toxicity Test"
GLP: Yes No ?
Test substance: C₁₀₋₁₃ LAS (CAS # 68411-30-3)
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. The submitter (Huels AG) judged the study quality to be good. Analysis showed 92% of nominal concentration. Tap water diluent; water hardness = 96-120 mg/L CaCO₃; pH 6.8-7.3; daily renewal; 14.5-16°C; 5 month old fish tested.
Reference: European Commission. 2000i. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, AT/17.
Reliability: 4 Not assignable. The original study was not available for review.

(l)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system
Species: *Salmo gairdneri* (Fish, estuary, fresh water)
Exposure period: 96 hour
Results: LC₅₀ = 3 mg/L
Analytical monitoring: Yes No ?
Method: See remarks.
GLP: Yes No ?
Test substance: DOBANIC ACID 102, C₁₀₋₁₃ LAS (CAS #68411-30-3)
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. Daily renewal of test solutions; 14-16 °C; pH 7.6-8.4; water hardness = 210-240 mg/L CaCO₃; DO=10.0-10.4 mg/L. Fingerlings were tested, mean weight 5.2 g
Reference: European Commission. 2000w. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Shell Research Ltd, SBGR.81.083, RR Stephenson.
Reliability: 4 Not assignable. The original study was not available for review.

(m) (Bluegill sunfish)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system
Species: *Lepomis macrochirus* (Fish, fresh water)
Exposure period: 96 hour
Results: LC₅₀ = 5.0 mg/L (mean of 8 tests)
Analytical monitoring: Yes No ?
Method: EPA-660/3-75-009
GLP: Yes No ?
Test substance: C₁₀₋₁₃ LAS, average chain length 11.8 (CAS #68411-30-3)
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. LC₅₀ values for the 8 tests conducted ranged from 3.7 to 7.7 mg/L. Nominal concentration, (expected deviation <20%), reconstituted water, water hardness = 30-48 mg/L CaCO₃; pH 7.3-7.8; 20-23°C; fish size: 0.35-0.89 g
Reference: European Commission. 2000q. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, 22852, 23613, 23612, 23617, 23722, 22824, 28661, 27917.

Reliability: 4 Not assignable. The original studies were reviewed by HERA (2004) for Annex 2 and judged to be reliable.

(n)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system

Species: *Lepomis macrochirus* (Fish, fresh water)

Exposure period: 96 hour

Results: $LC_{50} = 2.2$ mg/L

Analytical monitoring: Yes No ?

Method: EPA-660/3-75-009

GLP: Yes No ?

Test substance: C_{10-13} LAS, average chain length 11.8 (CAS #68411-30-3)

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. Nominal concentrations, (expected deviation <20%). Reconstituted water with hardness = 44 mg/L $CaCO_3$; pH 7.58; 22°C; fish size: 0.35 g, 40 mm; Note that this study was not included in the HERA LAS acute toxicity data summary tables in the SIAR because only a summary of the study, not the full report, is available.

Reference: European Commission. 2000p. Benzenesulfonic acid, C_{10-13} -alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, 22581, 28361.

Reliability: 4 Not assignable. The original study was not available for review.

(o) (Golden Orfe)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system

Species: *Leuciscus idus* (Fish, fresh water) (Golden orfe)

Exposure period: 48 hour

Results: $LC_{50} = 4.6$ mg/L

Analytical monitoring: Yes No ?

Method: Determination of the effect of substance in water on fish, DIN 38412 part 15

GLP: Yes No ?

Test substance: C_{10-13} LAS, sodium salt (CAS #68411-30-3)

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3.

Reference: European Commission. 2000a. Benzenesulfonic acid, C_{10-13} -alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Huels Study (unpublished).

Reliability: 4 Not assignable. The original study was not available for review.

(p) (Fathead minnow)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system not stated

Species: *Pimephales promelas* (Fish, fresh water)

Exposure period: 96 hour

Results: $LC_{50} = 4.6$ mg/L

Analytical monitoring: Yes No ?

Method: Ten fathead minnows were exposed for 96 hours to LAS under the following conditions: hardness 35 mg/L as $CaCO_3$; pH 7.1; temperature 21°C. Fish were not fed during the exposure.

GLP: Yes No ?

Test substance: Low molecular weight LAS, sodium salt (CAS #68411-30-3); C₁₀ 5%, C₁₁ 27%, C₁₂ 53%, C₁₃ 13%, 2-phenyl 23%; average alkyl chain length = C_{11.1}.

Remarks: The carboxylated intermediates formed in the biodegradation of LAS were also tested and found to be several orders of magnitude less toxic than LAS. These intermediates undergo further biodegradation, more rapidly in a natural river water than in a synthetic medium.

Reference: Swisher, R.D., Gledhill, W.E., Kimerle, R.A. and Taulli, T.A. 1978. Carboxylated intermediates in the biodegradation of linear alkylbenzene sulfonates (LAS). VII International Congress on Surface Active Substance, Proceedings, Moscow, 1976 4:218-230.

Reliability: 2 Valid with restrictions

(q)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system

Species: *Pimephales promelas* (Fish, fresh water)

Exposure period: 96 hour

Results: LC₅₀ = 4.1 mg/L

Analytical monitoring: Yes No ?

Method: USEPA methods for acute toxicity tests with fish, macroinvertebrates, and amphibians. Ecol. Res. Series. EPA-660/3-75-009.

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS, average chain length 11.7 (CAS #68411-30-3) (C₁₀ 7.3%; C₁₁ 26.5%; C₁₂ 56.7%; C₁₃ 9.0%; C₁₄ 0.5%); Mean phenyl position = 3.9; Mean molecular weight = 345.

Remarks: Acute tests were conducted at EG&G Bionomics from 1971 to 1976 with 2-3 month old fathead minnows in 20-L glass vessels with soft reconstituted water (hardness = 40 mg/L as CaCO₃). Tests with LASs of average alkyl chain length of 11.2 and 13.3 were conducted concurrently, with the resultant 96 hour LC₅₀ values of 12.3 and 0.86 mg/L, respectively.

Reference: Holman, W.F. and Macek, K.J. 1980. An aquatic safety assessment of linear alkylbenzene sulphonate (LAS); chronic effects on fathead minnows. Trans. Am. Fish. Soc. 109(1):122-131.

Reliability: 2 Valid with restrictions

(r)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system

Species: *Pimephales promelas* (Fish, fresh water)

Exposure period: 48 hour

Results: The following table shows the acute toxicity of the tested materials (LC₅₀, mg/L).

	Average chain length	Fathead minnow LC ₅₀ (mg/L)	
		24 hour	48 hour
High molecular weight LAS	13.3	1.9	1.7
Individual homologues			

LAS			
C ₁₀	10	48.0	43.0
C ₁₁	11	17.0	16.0
C ₁₂	12	4.7	4.7
C ₁₃	13	1.7	0.4
C ₁₄	14	0.6	0.4
Nonlinear LAS components (DTIS)			
C ₁₀	10	87.0 ± 7.5	86.1 ± 15.0
C ₁₂	12	24.8 ± 5.8	21.5 ± 5.5
C ₁₄	14	8.1 ± 5.1	5.3 ± 3.9
Model biodegradation intermediates			
C ₄ (SØ Butyrate)	4	~10,000	~10,000
C ₅ (SØ Valerate)	5	~10,000	~10,000
C ₁₁ (SØU)	11	85.9 ± 5.1*	76.6 ± 12.4*

*Subsequent repurification of this sample yielded a product with the same isomeric composition but with LC₅₀ values over 1000 mg/L for fatheads.

Analytical monitoring: Yes No ? MBAS

Method: EPA-660/3-75-009 1975. Method for acute toxicity tests with fish, macroinvertebrates and amphibians.

Acute toxicity tests were conducted on high molecular weight LAS, individual pure homologues, non-linear LAS components (dialkyl tetralin or indane sulfonates, DTIS), and model biodegradation intermediates (sulfophenylundecane, SOU) in order to determine whether biodegradation decreases toxicity. Toxicity tests were conducted in 5 L of 100 mg/L hardness water using 5 fathead minnows per concentration.

GLP: Yes No ?

Test substance: 1) High molecular weight LAS: Average chain length = 13.3; C₁₁ 1%, C₁₂ 8%, C₁₃ 52%, C₁₄ 39%
 2) Individual LAS homologues of C₁₀, C₁₁, C₁₂, C₁₃, and C₁₄
 3) Nonlinear LAS components (DTIS)
 4) Model biodegradation intermediates

Remarks: The length of the alkyl chain is the most important factor influencing acute toxicity, which increases as the alkyl chain increases. These longer alkyl chain homologues are the first constituents of the LAS mixture to degrade. The nonlinear components (DTIS) showed 1/2 to 1/10 the toxicity of LAS with the same carbon chain length. Toxicity of biodegradation intermediates is significantly less than the parent LAS. Because of the shorter than normal study duration and smaller than standard number of fish per concentration, the reliability of the absolute values cannot be assessed. However, the study is considered reliable for the trends in the data because the homologues were tested under identical conditions.

Reference: Kimerle, R.A. and Swisher, R.D. 1977. Reduction of aquatic toxicity of linear alkylbenzene sulfonate (LAS) by biodegradation. Water Research 11:31-37.

Reliability: 2 Valid with restrictions

4.2 ACUTE TOXICITY TO AQUATIC INVERTEBRATES

A. Daphnia

(a)

Type of Test: Static [X]; semi-static []; flow-through
Open-system [X]; closed-system []
Species: *Daphnia magna* (Crustacea)
Exposure Period: 48 hours
Effect Criteria: Immobility
Results: EC₅₀ = 1.62 mg/L; The number of immobile animals at each concentration is shown in the following table:

Concentration of test substance (mg/L)								
Time (h)	0	3.2	5.6	10	18	32	56	100
0	0	0	0	0	0	0	0	0
24	0	0	0	0	0	0	12	20
48	0	0	0	1 ^a	6 ^a	12 ^a	20	20

^a Condition of mobile animals was impacted as compared to the controls in a dose-responsive manner.

Analytic Monitoring: Yes [X]; No []; ? []
Method: OECD Guideline 202. Seven concentrations (3.2, 5.6, 10, 18, 32, 56, and 100 mg/L) plus controls were tested. The dilution water was DSWL water, prepared from ground water. Four beakers containing five *Daphnia* each were used for each test or control solution. *Daphnia* were less than 24 hours old at test initiation. A fifth beaker with 100 mg/L solution and five *Daphnia* were added 24 hours after initiation. Test and control solutions were not renewed and the *Daphnia* were not fed. Dissolved oxygen and pH were measured at 0 and 48 hours in all concentrations, as well as at 24 hours in the 100 mg/L chambers 24 hours after initiation because of total immobility observed at that level. Dissolved oxygen ranged from 8.7 to 9.6 mg/L and pH ranged from 7.9 to 8.1. Water hardness was 215 mg/L as CaCO₃. Test temperature was maintained at 20 ± 1°C under a 16:8 (light:dark) cycle. The chambers were not aerated. Immobile animals were counted at 24 and 48 hours. Samples for analysis of each concentration were taken at 0, 24, and 48 hours. This is a key study for aquatic toxicity to invertebrates (see SIAR Table 10).
GLP: Yes [X]; No []; ? []
Test Substance: LAS; 87.85% activity
Remarks: A 24 hour EC₅₀ of 3.58 mg/L and a 48 hour NOEC of 0.379 mg/L (both based on immobility) were also calculated, as well as a 48 hour NOEC of 5.6 mg/L based on condition. Only nominal concentrations are reported. No further information on the test substance is reported.
Reference: Hooftman, R.N. and van Drongelen-Sevenhuijsen, D. 1990. The acute Toxicity of E-3473.01 (ETS 311) to *Daphnia magna*. TNO Netherlands Organization for Applied Research. TNO Report No. R 89/403.
Reliability: 1 Valid without restrictions.

(b)

Species: *Daphnia magna*
Results: EC₅₀ values ranged from 1.62 to 9.3 mg/L
Test Substance: C₁₀₋₁₃ LAS (CAS #68411-30-3)

Remarks: A total of 20 daphnid studies were reviewed by HERA in 2004. Nine of these studies were rejected because the test material was not commercial LAS, the study deviated significantly from standard protocols, or non-standard endpoints were measured. The remaining 11 studies were evaluated for reliability and the results reflect the range of acute EC₅₀ values obtained for *Daphnia magna*. These studies are tabulated and discussed further in the SIAR and SIAP in a weight-of-evidence approach. A robust summary for the study with the lowest acute EC₅₀ value was prepared (see Hooftman and van Drongelen-Sevenhuijsen above).

Reference: HERA-LAS team, May 2004; see SIAR Annex 2.

Reliability: 4 Not assignable. This study has been given a reliability score of 4 because it is a summary evaluation of a series of studies conducted by other researchers.

(c)

Species: *Daphnia magna*

Results: EC₅₀ = 4.7 mg/L (139 records)

Test Substance: C10-14 LAS (all LAS in range, including data for individual homologues)

Remarks: EC₅₀ is geometric mean of 139 records compiled from a BKH (1993) literature review. Values range from 0.26 to 55 mg/L. This large range is caused by differences in the LAS tested with respect to alkyl chain and/or phenyl isomer distribution and differences in test design.

Reference: van de Plassche, E.J., de Bruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J. and Belanger, S.E. 1999. Predicted no-effect concentrations and risk characterization of four surfactants: LAS, AE, AES, and soap, Environ. Toxicol. Chem. 18:2653-2663.

Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(d)

Type of test: static ; semi-static ; flow-through ; other ;
open-system ; closed-system not stated

Species: *Daphnia magna* (Crustacea)

Exposure period: 48 hour

Results: EC₅₀ = 6.8 mg/L

Analytical monitoring: Yes No ?

Method: Directive 84/449/EEC, C.2 "Acute toxicity for *Daphnia*" 1984

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS, sodium salt (CAS #68411-30-3)

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3.

Reference: European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Huels Study (unpublished).

Reliability: 4 Not assignable. The original study was not available for review.

(e)

Type of test: static ; semi-static ; flow-through ; other ;
open-system ; closed-system

Species: *Daphnia magna* (Crustacea)

Exposure period: 48 hour

Results: LC₅₀ = 5.5 mg/L (mean of 3 valid tests)

Analytical monitoring: Yes [] No [X] ? []
 Method: EPA-660/3-75-009
 GLP: Yes [] No [] ? [X]
 Test substance: C₁₀₋₁₃ LAS, average chain length 11.8 (CAS #68411-30-3)
 Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Life-stage: <24 h. Effect: immobility. LC₅₀ values for 4 tests ranged from 4.4 to 10.4 mg/L. Huels AG judged study quality to be good. Nominal concentrations (expected deviation <20%). Reconstituted water with hardness = 162-220 mg/L CaCO₃. Note that all four of these studies are included in Appendix 2, the HERA acute toxicity data review. Three of the studies, Reports 23618, 22853 and 23611 with respective values of 4.4, 4.9 and 7.1 mg/L, are considered reliable and are included in the table of acute toxicity values. The fourth study, Report 23276 with a value of 10.4 mg/L, is listed among the rejected studies because it was conducted as part of a QA program to qualify various labs and the result is not considered reliable.
 pH 7.86-8.53. 21-22°C
 Reference: European Commission. 2000r. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, 23618, 22853, 23611, 23276.
 Reliability: 2 Valid with restrictions for the three valid studies. The original studies were reviewed by HERA (2004) for Annex 2.

(f)
 Type of test: static [X]; semi-static []; flow-through []; other [];
 open-system [X]; closed-system []
 Species: *Daphnia magna* (Crustacea)
 Exposure period: 48 hour
 Results: The following table shows the acute toxicity of original materials (LC₅₀, mg/L).

		<i>Daphnia magna</i>	
	Average chain length	24 hour	48 hour
High molecular weight LAS	13.3	2.6 ± 0.1	2.3 ± 0.1
Individual homologues LAS			
C ₁₀	10	53.1 ± 0.4	12.3 ± 2.6
C ₁₁	11	15.8 ± 3.0	5.7 ± 0.6
C ₁₂	12	10.7 ± 1.6	3.5 ± 1.0
C ₁₃	13	2.7 ± 0.4	2.0 ± 0.3
C ₁₄	14	1.2 ± 0.2	0.7 ± 0.2
Nonlinear LAS components (DTIS)			
C ₁₀	10	106.0 ± 27.0	98.0 ± 21.3
C ₁₂	12	55.1 ± 9.1	34.1 ± 5.1
C ₁₄	14	12.4 ± 1.4	10.0 ± 1.0
Model biodegradation intermediates			
C ₄ (SØ Butyrate)	4	~12,000	~6,000
C ₅ (SØ Valerate)	5	~12,000	~5,000

		<i>Daphnia magna</i>	
	Average chain length	24 hour	48 hour
C ₁₁ (SØU)	11	355 ± 150*	208 ± 85*

*Subsequent repurification of this sample yielded a product with the same isomeric composition but with LC₅₀ values over 1000 mg/L for daphnids (Swisher et al., 1976).

Analytical monitoring: Yes No ? MBAS
Method: EPA-660/3-75-009 1975. Method for acute toxicity tests with fish, macroinvertebrates and amphibians.
Acute toxicity tests were conducted on high molecular weight LAS, individual pure homologues, non-linear LAS components (dialkyl tetralin or indane sulfonates (DTIS), and model biodegradation intermediates (sulfophenyl undecane, SØU) in order to determine whether biodegradation decreases toxicity. In 250 mL beakers with 200 mL of well water of approximately 250 mg/L hardness, ten *Daphnia*, less than 18 hours old, were placed in each of the three beakers. No food was added for the duration of the test.
GLP: Yes No ?
Test substance: 1) High molecular weight LAS: Average chain length = 13.3; C₁₁ 1%, C₁₂ 8%, C₁₃ 52%, C₁₄ 39%
2) Individual LAS homologues of C₁₀, C₁₁, C₁₂, C₁₃, and C₁₄
3) Nonlinear LAS components (DTIS)
4) Model biodegradation intermediates
Remarks: The length of the alkyl chain is the most important factor influencing acute toxicity, which increases as the alkyl chain increases. These longer alkyl chain homologues are the first constituents of the LAS mixture to degrade. The nonlinear components (DTIS) showed 1/2 to 1/10 the toxicity of LAS with the same carbon chain length. Toxicity of biodegradation intermediates is significantly less than the parent LAS. *Daphnia* acute toxicity tests on partially degraded LAS demonstrated that toxicity is significantly lessened as LAS biodegraded. Because of the shorter than normal study duration and smaller than standard number of *Daphnia* per concentration, the reliability of the absolute values cannot be assessed. However, the study is considered reliable for the trends in the data because the homologues were tested under identical conditions.
Reference: Kimerle, R.A. and Swisher, R.D. 1977. Reduction of aquatic toxicity of linear alkylbenzene sulfonate (LAS) by biodegradation. *Water Research* 11:31-37.
Reliability: 2 Valid with restrictions

B. Other aquatic invertebrates

(a)
Species: *Gammarus pulex* (amphipod)
Mysidopsis bahia (mysid)
Panaeus duorarum (pink shrimp)
Results: EC₅₀ (*G. pulex*) = 6.2 mg/L (25 records)
EC₅₀ (*M. bahia*) = 1.7 mg/L (6 records)
EC₅₀ (*P. duorarum*) = 49 mg/L (5 records)
Test Substance: C₁₀₋₁₄ LAS (all LAS in range, including data for individual homologues)

Remarks: Geometric mean EC₅₀ values for number of records listed for each species. The interspecies variation decreases considerably when the geometric mean value per species is calculated.

Reference: van de Plassche, E.J., de Bruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J. and Belanger, S.E. 1999. Predicted no-effect concentrations and risk characterization of four surfactants: LAS, AE, AES, and soap, Environ. Toxicol. Chem. 18:2653-2663.

Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(b)

Species: Crustacean species

Results: LC₅₀ = 17.0 mg/L (14 records; SD = 0.68)

Test Substance: LAS; all in the alkyl chain length C₁₀₋₁₄

Remarks: LC₅₀ is geometric mean of 14 records compiled from literature reviews. Geometric mean LC₅₀ for all taxa (36 records) was 4.36 mg/L.

Reference: Temara, A., Carr, G., Webb, S., Versteeg, D., and Feijtel, T.C.J. 2001. Marine risk assessment: Linear alkylbenzenesulphonates (LAS) in the North Sea. Mar. Poll. Bulletin 42:635-642.

Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(c)

Type of test: static ; semi-static ; flow-through ; other ;
open-system ; closed-system

Species: *Chironomus riparius* (chironomid)

Exposure period: 96 hour

Results: LC₅₀ = 6.5 mg/L

Analytical monitoring: Yes No ?

Method: EPA

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS, average chain length 12.3 (CAS #68411-30-3)

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. Nominal concentrations (expected deviation <20%). Well water with hardness = 24-30 mg/L CaCO₃; pH 7.1; 21-22°C

Reference: European Commission. 2000v. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, 34845.

Reliability: 4 Not assignable. The original study was not available for review.

(d)

Type of test: static ; semi-static ; flow-through ; other ;
open-system ; closed-system

Species: *Limnodrilus hoffmeisteri* (aquatic worm)

Exposure period: 96 hour

Results: LC₅₀ = 1.8 mg/L

Analytical monitoring: Yes No ?

Method: EPA 660/3-75-009

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS, average chain length 12.3 (CAS #68411-30-3)
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Additional LC₅₀ values for extended exposure times: 144 h: 1.1 mg/L, 196 h: 0.96 mg/L. Huels AG judged study quality to be good. Nominal concentrations (expected deviation <20%). Well water with hardness = 24-30 mg/L CaCO₃; pH 7.1; 21-22°C.
Reference: European Commission. 2000v. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, 34845.
Reliability: 4 Not assignable. The original study was not available for review.

(e)

Type of test: static ; semi-static ; flow-through ; other ;
open-system ; closed-system
Species: *Planaria* sp. (aquatic worm)
Exposure period: 48 hour
Results: LC₅₀ = 1.8 mg/L
Analytical monitoring: Yes No ?
Method: EPA 660/3-75-009
GLP: Yes No ?
Test substance: C₁₀₋₁₃ LAS, average chain length 12 (CAS #68411-30-3)
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. Nominal concentrations (expected deviation <20%). Reconstituted water with hardness 165 mg/L CaCO₃. pH 8.1-8.4; 21-22°C; size 3.4 cm.
Reference: European Commission. 2000u. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, 31340.
Reliability: 4 Not assignable. The original study was not available for review.

(f)

Type of test: static ; semi-static ; flow-through ; other ;
open-system ; closed-system
Species: *Rhabditis* sp. (nematode)
Exposure period: 48 hour
Results: LC₅₀ = 16 mg/L
Analytical monitoring: Yes No ?
Method: EPA 660/3-75-009
GLP: Yes No ?
Test substance: C₁₂ LAS (CAS #25155-30-0) (average chain length 11.8)
Remarks: Nominal concentrations, (expected <20%), 3 replicates of 5 test concentrations plus a control. Reconstituted water with hardness = 65 mg/L CaCO₃; DO 5.3 mg/L; pH 8.1-8.4; 21-23°C; size: 0.3 mm; 12 hr. photoperiod at 40-70 L/ft². Five other species were also tested and had the following 48-hr LC₅₀ values (all mg/L): Midge 23; *Gammarus* (amphipod) 3.3; *Asellus* (isopod) 270; *Dugesia* (flatworm) 1.8; and *Dero* (oligochaete) 1.7.
Reference: Lewis, M.A. and Surprenant, D. 1983. Comparative acute toxicities of surfactants to aquatic invertebrates. *Ecotoxicol. Env. Saf.* 7(3):313-322.
Reliability: 2 Valid with restrictions

4.3 TOXICITY TO AQUATIC PLANTS, e.g. algae

(a)

Type of Test: Static [X]; semi-static []; flow-through []
Open-system [X]; closed-system []; not stated []

Species: *Selenastrum capricornutum* (algae)

Endpoint: Biomass []; Growth rate [X]; Other []

Exposure Period: 96 hours

Results: EC₅₀ = 29.0 mg/L
NOEC = 0.5 mg/L
LOEC = 1.0 mg/L

Analytic Monitoring: Yes []; No []; ? [X]

Method: ASTM. 1984. Standard Practice for Conducting Toxicity Tests with Microalgae, Draft #7, Philadelphia, PA. Algae were exposed to the test material for four days, after which cell counts were made. The EC₅₀ value was calculated using the method of Larson and Schaefer. Assumed 1 x 10⁴ cells/mL because ASTM protocol, but not reported.

GLP: Yes []; No [X]; ? []

Test Substance: C_{11.8} LAS; MW = 345; technical grade LAS from P&G

Remarks: The first significant effect concentrations were between 0.5 and 1.0 mg/L. Mean test temperature was 23.6 (21.2-25.6 °C). Total mean water hardness was 137 mg/L as CaCO₃. The pH range was 6.8 to 7.2. Mean dissolved oxygen was 9.1 mg/L. Results of the laboratory studies were compared with enclosure studies conducted with natural phytoplankton assemblages. Concentrations in the enclosures that first altered community structure were found to be between 27 and 108 mg/L. The EC₅₀ value reported represents the lowest acute value for algal species for which a report or publication was available. This is a key study for aquatic toxicity to algae (see SIAR Tables 10 and 12).

Reference: 1) Lewis, M.A. 1986. Comparison of effects of surfactants on freshwater phytoplankton communities in experimental enclosures and on algal population growth in the laboratory. Environ. Toxicol. Chem. 5:319-332.
2) Lewis, M.A. and Hamm, B.G. 1986. Environmental modification of the photosynthetic response of lake plankton to surfactants and significance to a laboratory-field comparison. Wat. Res. 20:1575-1582.

Reliability: 2 Valid with restrictions

(b)

Species: *Selenastrum capricornutum* and *Scenedesmus subspicatus*

Endpoint: Biomass []; Growth rate [X]; Other []

Results: E_rC₅₀ values ranged from 29 to 35.5 for *S. capricornutum* (two records)
E_rC₅₀ values ranged from 82 to 163 mg/L for *S. subspicatus* (three records)

Test Substance: C₁₀₋₁₃ LAS (CAS #68411-30-3)

Remarks: A total of 13 algae studies originally were reviewed by HERA in 2004. Eight of these studies were rejected because the test material was not commercial LAS, the study deviated significantly from standard protocols, or non-standard endpoints were measured. The remaining five studies were evaluated for reliability and the results reflect the range of acute E_rC₅₀ values obtained for the two most commonly tested algal species. These studies are tabulated and discussed further in the SIAR and SIAP in a weight-of-evidence approach. A robust summary for the study with the lowest acute E_rC₅₀

value was prepared (see Lewis 1986 above).
Reference: HERA-LAS team, May 2004; see SIAR Annex 2.
Reliability: 4 Not assignable. This study is given a reliability score of 4 because it is a summary evaluation of a series of studies conducted by other researchers.

(c)

Species: *Chlamydomonas reinhardi*, *Chlorella kessleri*, *Microcystis sp.*, *Plectonema boryanum*, *Scenedesmus subspicatus*, *Selenastrum sp.* (algae)

Results: NOEC (*C. reinhardi*) = 12 mg/L (1 record)
NOEC (*C. kessleri*) = 3.5 mg/L (1 record)
NOEC (*Microcystis sp.*) = 0.80 mg/L (4 records)
NOEC (*P. boryanum*) = 15 mg/L (1 record)
NOEC (*S. subspicatus*) = 7.7 mg/L (4 records)
NOEC (*Selenastrum sp.*) = 3.8 mg/L (9 records)

Test Substance: LAS normalized to C_{11.6}

Remarks: Geometric mean NOEC values for number of records listed for each species.

Reference: van de Plassche, E.J., de Bruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J. and Belanger, S.E. 1999. Predicted no-effect concentrations and risk characterization of four surfactants: LAS, AE, AES, and soap. Environ. Toxicol. Chem. 18:2653-2663.

Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(d) (***Scenedesmus***)

Species: *Scenedesmus subspicatus* (Algae)

Endpoint: Biomass []; Growth rate [X]; Other []

Exposure period: 72 hour

Results: The 72-hr EC₅₀ values were 240, 163, and 54.3 mg/L for the C₁₁, C_{11.6}, and C₁₃ LAS, respectively. Other endpoints not determined.

Analytical monitoring: Yes [] No [X] ? []

Method: OECD 201 Algal growth inhibition test. 1984. The test media recommended by AFNOR was used. All water quality parameters were maintained within acceptable ranges in compliance with the test protocol. Initial cell concentrations were 1 x 10⁴ cells/mL. Final cell counts not reported.
open-system [X]; closed-system []

GLP: Yes [X] No [] ? []

Test substance: Three different alkyl chain lengths of LAS (C₁₁, C_{11.6}, C₁₃), with the following homologue distributions:

	Alkyl Chain Length Distributions (%)		
	LAS C ₁₁	LAS C _{11.6}	LAS C ₁₃
<C ₁₀	1.5	0.4	--
C ₁₀	29.0	8.9	1.0
C ₁₁	39.0	33.7	3.5
C ₁₂	28.5	31.0	17.8
C ₁₃	1.8	24.0	37.0
C ₁₄	0.2	2.0	40.4
>C ₁₄	--	--	0.3

LAS MW (as Na-LAS)	334	343	363
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Remarks: German strain of *S. subspicatus* from the University of Gottingen. Other endpoints were not reported.

Reference: Verge, C. and Moreno, A. 1996a. Toxicity of anionic surfactants to green microalgae "*Scenedesmus subspicatus*" and "*Selenastrum capricornutum*." *Tenside Surf. Det.* 33:166-168.

Reliability: 2 Valid with restrictions

(e)

Species: *Scenedesmus subspicatus* (Algae)

Endpoint: Biomass []; Growth rate [X]; Other []

Exposure period: 72 hour

Results: The 72-hr EC₅₀ values were 270, 111, 48, 30, and 18 mg/L for pure homologues C₁₀, C₁₁, C₁₂, C₁₃, and C₁₄, respectively. The corresponding NOEC values were 80, 40, 18, 12, and 7, respectively.

Analytical monitoring: Yes [] No [X] ? []

Method: OECD 201 Algal growth inhibition test. 1984. The test media recommended by AFNOR was used. All water quality parameters were maintained within acceptable ranges in compliance with the test protocol. Initial cell concentrations were 1 x 10⁴ cells/mL. Final cell counts not reported.

GLP: open-system [X]; closed-system []

Test substance: Yes [X] No [] ? []

Test substance: Five pure homologue cuts were tested, with the following distribution.

	Alkyl Chain Length Distributions (%)				
	LAS C ₁₀	LAS C ₁₁	LAS C ₁₂	LAS C ₁₃	LAS C ₁₄
<C ₁₀	0.5	--	0.4	--	--
C ₁₀	96.8	5.5	13.9	0.7	--
C ₁₁	2.7	93.7	84.5	9.8	0.6
C ₁₂	--	0.8	1.2	78.3	1.0
C ₁₃	--	--	--	11.2	15.4
C ₁₄	--	--	--	--	82.1
>C ₁₄	--	--	--	--	0.9
LAS MW (as Na-LAS)	320.7	333.7	346.4	362.3	373.7

Remarks: French strain of *S. subspicatus* from the University of Metz. As NOEC (no observed effect concentration), the authors used the EC₅ because of the lack of noticeable variation in toxicity for the interval 0-5%. This is a key study for aquatic toxicity to algae (see SIAR Table 12).

Reference: Verge, C. and Moreno, A. 1996a. Toxicity of anionic surfactants to green microalgae "*Scenedesmus subspicatus*" and "*Selenastrum capricornutum*." *Tenside Surf. Det.* 33:166-168.

Reliability: 2 Valid with restrictions

(f)

Species: *Scenedesmus subspicatus* (Algae)

Endpoint: Biomass [X]; Growth rate [X]; Other []

Exposure period: 72 hour

Results: E_rC_{50} (growth rate) = 127.9 mg/L; E_bC_{50} (biomass) = 43.2 mg/L
 NOEC (growth rate) = 2.4 mg/L; NOEC (biomass) = 2.2 mg/L
 LOEC (growth rate) = 10 mg/L

Inhibition of cell growth by concentration and duration is shown in the table below:

Cell number (x 10 ⁴ cells/mL)				
Test concentration (mg/L)	Time Period (hours)			
	0	24	48	72
Control	2	8	29	95
0.6	2	8	30	89
2.4	2	9	28	85
10	2	9	27	75
40	2	8	20	48
160	2	7	9	8

Analytical monitoring: Yes [] No [X] ? []

Method: open-system [X]; closed-system []
 Algal growth inhibition test (88/302/EWG) 1988.

Nominal test concentrations were control, 0.6, 2.4, 10, 40, and 160 mg/L. Algae were exposed to LAS in Erlenmeyer flasks in an environmental chamber on a light table at 8000 lux. Cell numbers were photometrically determined (8 subsets were taken for each concentration).

GLP: Yes [X] No [] ? []

Test substance: Marlon A 390 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = 11.6; 91.3% activity

Remarks: Initial cell concentrations were 20,000 cells/mL. Cell concentrations at 72 h were 95, 89, 85, 75, 48 and 8 (all x 10⁴/mL) for the control, 0.6, 2.4, 10, 40 and 160 mg/L LAS concentrations, respectively. The pH ranged from 7.7 to 7.9 at the beginning of the study and 7.9 to 9.0 at the end of the study. Test temperature was maintained at 24 ± 2 °C. This is a key study for aquatic toxicity to algae (see SIAR Table 12).

Reference: Scholz, N. 1992. Bestimmung der Auswirkungen von Marlon A 390 auf das Wachstum von *Scenedesmus subspicatus* 86.81. SAG (Algenwachstumshemmtest nach Richtlinie 88/302/EWG) Huels Final Report No. AW-291.

Reliability: 1 Valid without restriction

(g)

Species: *Scenedesmus subspicatus* (Algae)
 Endpoint: Biomass [X]; Growth rate [X]; Other []

Exposure period: 72 hour

Results: E_rC_{50} (growth rate) = 82 mg/L; E_bC_{50} (biomass) = 20 mg/L
 NOEC (growth rate) = 0.4 mg/L; NOEC (biomass) = 0.1 mg/L

Inhibition of cell growth by concentration and duration is shown in the table below:

Cell number (x 10 ⁴ cells/mL)	
	Time Period (hours)

Test concentration (mg/L)	0	24	48	72
Control	2	9	32	94
0.1	2	9	32	91
0.4	2	9	29	88
1.6	2	9	28	76
6.4	2	9	24	60
25	2	9	21	54
160	2	6	7	7

Analytical monitoring: Yes No ?

Method: open-system ; closed-system
 Algal growth inhibition test (92/69/EWG)
 Nominal test concentrations were control, 0.1, 0.4, 1.6, 6.4, 25 and 160 mg/L. Test concentrations were measured at 0 and 72 h and found to confirm the nominal concentrations. Algae were exposed to LAS in Erlenmeyer flasks in an environmental chamber on a light table at 8000 lux. Cell numbers were photometrically determined (8 subsets were taken for each concentration).

GLP: Yes No ?

Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = 11.6; 52.1% activity

Remarks: Initial cell concentrations were 20,000 cells/mL. Cell concentrations at 72 h 94, 91, 88, 76, 60, 54 and 7 (all x 10⁴/mL) for the control, 0.1, 0.4, 1.6, 6.4, 25 and 160 mg/L LAS concentrations, respectively. The pH ranged from 8.2 to 8.3 at the beginning of the study and 8.0 to 9.4 at the end of the study. Test temperature was maintained at 24 ± 2 °C. This is a key study for aquatic toxicity to algae (see SIAR Table 12).

Reference: Scholz, N. 1994. Bestimmung der Auswirkungen von Marlon A 350 auf das Wachstum von *Scenedesmus subspicatus* 86.81. SAG (Algenwachstumshemmtest nach Richtlinie 92/69/EWG) Huels Final Report No. AW-372.

Reliability: 1 Valid without restriction

(h)

Species: *Scenedesmus subspicatus* (Algae)

Endpoint: Biomass ; Growth rate ; Other

Exposure period: 96 hour

Results: EC₅₀ = 5 mg/L

Analytical monitoring: Yes No ?

Method: DIN 38412 Part 9

GLP: Yes No ?

Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = 11.6

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Data refer to 100% active ingredient. Test method conforms with OECD-Guideline 201.

Reference: European Commission. 2000b. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).

Reliability: 4 Not assignable. The original study was not available for review.

(i)
Species: *Scenedesmus subspicatus* (Algae)
Endpoint: Biomass []; Growth rate [X]; Other []
Exposure period: 96 hour
Results: EC₅₀ = 9 mg/L
Analytical monitoring: Yes [] No [] ? [X]
Method: Directive 87/302/EEC, part C, p. 89 "Algal inhibition test"
GLP: Yes [] No [X] ? []
Test substance: C₁₀₋₁₃ LAS, sodium salt; average chain length 11.6 (CAS #68411-30-3)
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good.
Reference: European Commission. 2000c. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Huels AG, 1/90, N. Scholz, unpublished.
Reliability: 4 Not assignable. The original study was not available for review.

(j)
Species: *Scenedesmus subspicatus* (Algae)
Endpoint: Biomass []; Growth rate [X]; Other []
Exposure period: 96 hour
Results: EC₅₀ = 30 mg/L
Analytical monitoring: Yes [X] No [] ? []
Method: ISO 8692 "Water quality - Fresh water algal growth inhibition test with *Scenedesmus subspicatus* and *Selenastrum capricornutum*"
GLP: Yes [] No [] ? [X]
Test substance: C₁₁₋₁₃ LAS
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. Nominal concentrations (deviation <3%); BBM medium; pH 6.4-6.7; 20-22°C. Note that although this was cited in IUCLID as a Procter & Gamble report, Procter & Gamble indicates that it is unlikely that it is one of their reports.
Reference: European Commission. 2000h. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, AL/12.
Reliability: 4 Not assignable. The original study was not available for review.

(k) (***Selenastrum***)

Species: *Selenastrum capricornutum* (Algae)
Endpoint: Biomass []; Growth rate [X]; Other []
Exposure period: 96 hour
Results: EC₅₀ = 4.29-12.5 mg/L
Analytical monitoring: Yes [X] No [] ? []
Method: OECD Guideline 201 "Algae, Growth Inhibition Test", 1984
GLP: Yes [] No [] ? [X]
Test substance: LAS, average chain length 11.8 (CAS #68411-30-3)
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. Water hardness = 150 mg/L as NaHCO₃. Reported results are EC₅₀ values for 2 tests. Static mean EC₅₀ = 7.3 mg/L.
Reference: European Commission. 2000t. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, 29101.

Reliability: 3 Invalid. Based on correspondence with Procter & Gamble, it appears that this study was mis-cited. The P&G report 29101 indicates 96-h EC₅₀ values of 29 mg/L and greater than 10 mg/l, not the values reported above. In addition, the P&G study was not conducted under the OECD Guideline cited. Given these discrepancies, the values are uncertain and are considered invalid.

(l)

Species: *Selenastrum capricornutum* (Algae)
Endpoint: Biomass []; Growth rate [X]; Other []
Exposure period: 72 hour
Results: EC₅₀ = 11 mg/L
Analytical monitoring: Yes [X] No [] ? []
Method: ISO 8692 "Water quality - Fresh water algal growth inhibition test with *Scenedesmus subspicatus* and *Selenastrum capricornutum*"
GLP: Yes [] No [] ? [X]
Test substance: C₁₁₋₁₃ LAS (CAS #68411-30-3)
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. Nominal concentrations (deviation <13%); BBM medium; pH 6.5-6.6; static. Note that although this was cited in IUCLID as a Procter & Gamble report, Procter & Gamble indicates that it is unlikely that it is one of their reports.
Reference: European Commission. 2000g. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, AL/10.
Reliability: 4 Not assignable. The original study was not available for review.

(m)

Species: *Selenastrum capricornutum* (Algae)
Endpoint: Biomass []; Growth rate [X]; Other []
Exposure period: 96 hour
Results: EC₅₀ = 12.2 mg/L
Analytical monitoring: Yes [] No [] ? [X]
Method: EPA, 1987.
GLP: Yes [] No [] ? [X]
Test substance: C₁₀₋₁₃ LAS, average chain length 12.3 (CAS #68411-30-3)
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. AAP medium; 24⁺/-2°C.
Reference: European Commission. 2000n. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, P2636.01.
Reliability: 4 Not assignable. The original study was not available for review.

(n)

Species: *Selenastrum capricornutum* (Algae)
Endpoint: Biomass []; Growth rate [X]; Other []
Exposure period: 48 hour
Results: EC₅₀ ≈ 80 mg/L (Observation of graphical plot in paper indicates that the EC₅₀ is between 50-100 mg/L)
Analytical monitoring: Yes [] No [X] ? []
Method: Algal Assay Procedure Bottle Test, USEPA (1971).
Individual test flasks were spiked with a 1 mL algal suspension at 0.7

mg/L dry weight for each concentration. Test solutions were run in triplicate. Algae were maintained at $24 \pm 2^\circ\text{C}$ at 4000 lux for 48 hours. Cell counts were accomplished with a Coulter Counter and the growth Rate calculated.

GLP: Yes No ?
Test substance: C_{11.6} LAS; 23.4% activity
Remarks: Though generally valid, this study was not included in the HERA assessment because the exposure period was only 48 hours.
Reference: Yamane, A., Okada, M. and Sudo, R. 1984. The growth inhibition of planktonic algae due to surfactants used in washing agents. Water Res. 9:1101-1105.
Reliability: 2 Valid with restrictions. Duration of test considered too short for a chronic study, therefore not included in SIAR Table 12A.

(o) (*Elodea*)

Species: *Elodea canadensis* (aquatic plant)
Endpoint: Biomass ; Growth rate ; Other inhibition of growth, productivity
Exposure period: 28 day
Results: NOEC > 4 mg/L
Analytical monitoring: Yes No ?
Method: The effect of LAS on the structure and function of microbial communities was studied in a flow-through model ecosystem containing several trophic levels. Duplicate 19-L glass aquaria containing model ecosystems at four nominal concentrations (0.5, 1.0, 2.0, 4.0 mg/L) contained water and 2.5 cm lake sediment (Winton Lake, Cincinnati, OH) and several trophic levels (bacteria, algae, macrophytes [*Elodea canadensis*, *Lemna minor*], macroinvertebrates [*Daphnia magna*, *Paratanytarsus parthenogenica*], and fish [*Lepomis macrochirus*]). Flow rate in the proportional diluter delivered approximately 8 replacement volumes per day. Additionally, at each cycle of the diluter, 1.5 mL of a *Daphnia* food suspension diluted with a culture of *Selenastrum* was added to each chamber. Following an initial 3 day acclimation period to analytically confirm test concentrations, 4 glass periphyton slides (5 x 5 cm), 8 vegetative shoots of *Elodea*, 10 early instar *Daphnia*, 25 midge eggs and 5 pre-weighed juvenile bluegills (2.5-5.0 cm length) were added to each aquarium. Fish were screened from access to the macroinvertebrates by a 60 mesh stainless steel screen and were fed a daily supplement of frozen brine shrimp. Test duration was 28 days. Effects monitored included population and community effects. Nominal concentrations were confirmed with MBAS analysis. After 28 days the vegetative shoots of *Elodea* were removed, the total length measured, and the shoots were placed into aluminium dishes for determination of ash-free dry weights.

GLP: Yes No ?
Test substance: LAS; C¹⁴-LAS chain length C₁₂ (91% purity) plus unlabeled LAS with average chain length C_{11.6} (C₁₀ 9.7%, C₁₁ 27.9%, C₁₂ 54.4%, C₁₃ 8.0%; 95% purity) (tested together)
Remarks: Dissolved oxygen concentrations ranged between 7.0 and 9.0 mg/L. Temperature was maintained at 2°C and the mean pH was 8.1 ± 0.2 . The growth and productivity of *Elodea* was not significantly inhibited at the highest concentration tested (4 mg/L) in Phase I. Growth throughout the 28-day exposure approximately doubled the initial biomass of the

vegetative shoots from a mean of about 37 mg ash-free dry weight to a mean of about 72 mg for the treatment concentrations. Similarly, Phase II experiments resulted in an approximate doubling of the initial *Elodea* biomass in the controls and the 3.75% effluent concentration, though the heavy growth of attached bacteria and fungi (periphyton) that developed in the higher concentrations effectively covered the growing surface of the *Elodea* shoots resulting in progressively lower productivity in 7.5, 15 and 30% dilutions.

Reference: Maki, A.W. 1981. A Laboratory model ecosystem approach to environmental fate and effects studies. Unpublished Internal Report, Environmental Safety Department Procter and Gamble Company, Cincinnati, Ohio.

Reliability: 2 Valid with restrictions

(p) (*Lemna*)

Species: *Lemna minor* (aquatic plant)

Endpoint: Biomass []; Growth rate []; Other [X] frond count

Exposure period: 7 days

Results: EC₅₀ = 2.7 mg/L

NOEC = 0.9 mg/L (calculated as EC₅₀/3); normalized to C_{11.6} = 1.1 mg/L

Analytical monitoring: Yes [X] No [] ? []

Method: A 7-day flow-through growth inhibition test was developed using duckweed (*Lemna minor*). Control water (carbon and reverse osmosis filtered well water) and five concentrations of test material were delivered from diluter mixing chambers to Pyrex glass exposure chambers at a rate of 1.5 L every 16 minutes (approximately 14 replacement volumes per day). At time 0, each chamber received seven 2-frond, root excised duckweed colonies (four replicates per treatment level). Plants were given continuous illumination at 3875 lux (360 foot candles) and temperature was maintained between 21 and 23°C. Total hardness was 120-130 mg/L CaCO₃ and pH was 7.2-7.6. The number of fronds was recorded once every 24 hours for 7 days. LAS concentrations were measured using the MBAS method.

GLP: Yes [] No [] ? [X]

Test substance: C₁₀₋₁₄ LAS, average alkyl chain length 11.8, MW = 345, 27.3% active

Remarks: Results based on frond count were found to provide the most useful information per unit of laboratory time. Other endpoints resulted in 7-day EC₅₀ values of 3.6 mg/L (dry weight), 4.9 mg/L (root length), and 4.8 mg/L (growth rate/doubling time). Concurrent tests with bluegill sunfish (96-hr LC₅₀ = 1.7 mg/L) and *D. magna* (48-hr LC₅₀ = 4.4 mg/L) indicate that protection criteria developed for these species should be adequate to protect the aquatic macrophyte community without the need for additional testing.

Reference: Bishop, W.E. and Perry, R.L. 1981. Development and Evaluation of a Flow-Through Growth Inhibition Test with Duckweed (*Lemna minor*). Aquatic Toxicology and Hazard Assessment. ASTM STP 737. Branson, D.R. and Dickson, K.L. (ed.).

Reliability: 2 Valid with restrictions

(q)

Type of Test: Static [X]; semi-static []; flow-through []
Open-system []; closed-system []; not stated [X]

Species: *Chlamydomonas reinhardi* (algae)
 Exposure Period: not stated
 Effect Criteria: Growth inhibition
 Results: NOEC = 15 mg/L
 LOEC = 20 mg/L
 Analytic Monitoring: Yes []; No []; ? [X]
 Method: LAS concentrations of 1, 5, 10, 15, 20 and 30 mg/L were prepared in Deionized, sterilized double-distilled glass water. Algae were grown in Bold's medium. Cell counts were made using a spectrophotometer and dry weight observations.
 GLP: Yes []; No [X]; ? []
 Test Substance: C_{11.2} LAS
 Remarks: No morphological changes were observed. The growth rate was reduced at LAS concentrations of 20 mg/L. Protein analysis indicated that higher concentrations did affect protein synthesis. The NOEC normalized by van de Plassche et al. (1999) to C_{11.6} LAS was 12 mg/L.
 Reference: Dhaliwal, A.A., Campione, A., and Smaga, S. 1977. Effect of linear alkylbenzene sulfonate (C_{11.2} LAS) on the morphology and physiology of *Plectonema boryanum* and *Chlamydomonas reinhardi*. J. Phycol. 13:18.
 Reliability: 4 Not assignable. This study was given a reliability score of 4 because the document reviewed was an abstract.

(r)

Type of Test: Static [X]; semi-static []; flow-through []
 Open-system []; closed-system []; not stated [X]
 Species: *Chlorella kessleri* (algae)
 Exposure Period: 15 days
 Effect Criteria: Growth rate
 Results: NOEC = 3.1 mg/L
 LOEC = 10 mg/L
 Analytic Monitoring: Yes []; No [X]; ? []
 Method: EPA-600/9-78-018. Algal Assay Bottle Test. Determination of the Inhibitory effect of water constituents on green algae, by William E. Joseph C. Greene, and Tamotsu Shiroyama, Cornwallis Environmental Research Laboratory, Corvallis, Oregon.
 GLP: Yes []; No [X]; ? []
 Test Substance: Marlon A 350, Benzenesulfonic acid, C10-13-alkyl derives., sodium salts (CAS #68411-30-3); 25.7% activity.
 Remarks: No morphological changes were observed. The growth rate was reduced at LAS concentrations of 20 mg/L. Protein analysis indicated that higher concentrations did affect protein synthesis. The NOEC normalized by van de Plassche et al. (1999) to C_{11.6} LAS was 3.5 mg/L.
 Reference: Henkel KGaA, Biological Research and Product Safety/Ecology, unpublished results of study conducted in 1984; test substance Fi 5829.
 Reliability: 2 Valid with restrictions. Non-standard length of study, therefore not included in SIAR Table 12A.

(s)

Type of Test: Static [X]; semi-static []; flow-through []
 Open-system [X]; closed-system []; not stated []
 Species: *Microcystis aeruginosa* (algae)
 Exposure Period: 96 hours

Effect Criteria: Growth rate
 Results: $EC_{50} = 0.9$ mg/L
 Analytic Monitoring: Yes []; No [X]; ? []
 Method: The test species was cultured following the procedures of USEPA 1971. Temperature was maintained at $24 \pm 2^{\circ}C$. Water chemistry was determined at least once during each test according to Standard Methods (APHA 1985). EC_{50} values were calculated using the method of Larson and Schaeffer (1982) or graphical interpolation.

GLP: Yes []; No [X]; ? []
 Test Substance: C_{12} LAS; average molecular weight = 345
 Remarks: Mean hardness was 137 mg/L as $CaCO_3$, pH range was 6.8-7.2, and the mean dissolved oxygen was 9.1 mg/L. Comparison was also made to *in situ* studies conducted in which lake water was bottled and suspended in Lake Acton (Ohio) for 3 hour periods. The mean 3-h EC_{50} (photosynthesis) for the *in situ* studies was 3.4 mg/L (0.5-8.0 mg/L). The NOEC normalized by van de Plassche et al. (1999) to $C_{11.6}$ LAS was 0.35 mg/L. Using the acute to chronic ratio calculation (documented in Annex 3 of the LAS SIAR), the $EC_{50}/3$ for *Microcystis* is 0.3 mg/L. This is a critical study for this SIDS endpoint.

Reference: Lewis, M.A. and Hamm, B.G. 1986. Environmental modifications of the photosynthetic response of Lake Plankton to surfactants and significance to a laboratory-field comparison. *Wat. Res.* 20:1575-1582.

Reliability: 2 Valid with restrictions

(t)

Type of Test: Static []; semi-static []; flow-through []
 Open-system []; closed-system []; not stated [X]
 Species: *Plectonema boryanum* (algae); strain 597
 Exposure Period: not stated
 Effect Criteria: Growth rate
 Results: NOEC = 20 mg/L
 LOEC = 30 mg/L
 Analytic Monitoring: Yes []; No []; ? [X]
 Method: LAS concentrations of 1, 5, 10, 15, 20 and 30 mg/L were prepared in Deionized, sterilized double-distilled glass water. Algae were grown in Bold's medium.

GLP: Yes []; No [X]; ? []
 Test Substance: $C_{11.2}$ LAS
 Remarks: Growth rate was reduced at 30 mg/L concentrations of LAS as indicated by spectrophotometer readings and dry weight. The NOEC normalized by van de Plassche et al. (1999) to $C_{11.6}$ LAS was 15 mg/L.

Reference: Dhaliwal, A.S., Campione, A., and Smaga, S. 1977. Effect of linear alkylbenzene sulfonate ($C_{11.2}$ LAS) on the morphology and physiology of *Plectonema boryanum* and *Chlamydomonas reinhardtii*. *J. Phycol.* 12:18.

Reliability: 4 Not assignable This study was given a reliability score of 4 because the document reviewed was an abstract. However, these data were considered reliable as part of a weight-of-evidence approach in the analysis conducted by van de Plaasche et al. (1999).

4.4 TOXICITY TO BACTERIA

(a)
 Type: Aquatic ; Field ; Soil ; Other
 Species: activated sludge
 Exposure Period: 3 hour
 Results: EC₅₀ (Na LAS-C₁₁) = 760 mg/L
 EC₅₀ (Na LAS-C_{11.6}) = 550 mg/L
 EC₅₀ (Na LAS-C₁₃) = 650 mg/L
 Analytical monitoring: Yes No ?
 Method: OECD 209 Activated Sludge Respiration Inhibition Test. 1984.
 GLP: Yes No ?
 Test substance: Three different alkyl chain lengths of LAS (C₁₁, C_{11.6}, C₁₃ sodium salts), with the following homologue distributions.

	Alkyl Chain Length Distributions (%)		
	LAS C ₁₁	LAS C _{11.6}	LAS C ₁₃
<C ₁₀	1.5	0.4	--
C ₁₀	29.0	8.9	1.0
C ₁₁	39.0	33.7	3.5
C ₁₂	28.5	31.0	17.8
C ₁₃	1.8	24.0	37.0
C ₁₄	0.2	2.0	40.4
>C ₁₄	--	--	0.3
LAS MW (as Na-LAS)	334	343	363

Remarks: The purpose of the study was to determine the toxicity of three commercial LAS products to the activated sludge of a treatment plant basically operating on domestic sewage. A contact time of 3 hours instead of 15 minutes was chosen to better simulate the real residence time used in wastewater treatment plants (4-6 hours). The EC₅₀ values are far above environmental concentrations and therefore provide a high margin of safety. The 3-hour EC₅₀ range for the reference substance (3,5-dichlorophenol) ranged from 20-30 mg/L, within the valid range of 5-30 mg/L.

Reference: Verge C. and Moreno, A. 1996b. Toxicity of anionic surfactants to the bacterial population of a waste water treatment plant. Tenside Surf. Det. 33:323-327.

Reliability: 2 Valid with restrictions

(b)
 Type: Aquatic ; Field ; Soil ; Other
 Species: activated sludge
 Exposure Period: 3 hour
 Results: EC₅₀ (Na LAS-C₁₀) = 1042-1200 mg/L
 EC₅₀ (Na LAS-C₁₁) = 740-782 mg/L
 EC₅₀ (Na LAS-C₁₂) = 500-723 mg/L
 EC₅₀ (Na LAS-C₁₃) = 700-795 mg/L
 EC₅₀ (Na LAS-C₁₄) = 900-1045 mg/L
 Analytical monitoring: Yes No ?
 Method: OECD 209 Activated Sludge Respiration Inhibition Test. 1984.
 GLP: Yes No ?

Test substance: Five different pure homologues of LAS (C₁₀, C₁₁, C₁₂, C₁₃, C₁₄ sodium salts), with the following homologue distributions.

	Pure Homologues (%)				
	LAS C ₁₀	LAS C ₁₁	LAS C ₁₂	LAS C ₁₃	LAS C ₁₄
<C ₁₀	0.5	--	0.4	--	--
C ₁₀	96.8	5.5	13.9	0.7	--
C ₁₁	2.7	93.7	84.5	9.8	0.6
C ₁₂	--	0.8	1.2	78.3	1.0
C ₁₃	--	--	--	11.2	15.4
C ₁₄	--	--	--	--	82.1
>C ₁₄	--	--	--	--	0.9
LAS MW (as Na-LAS)	320.7	333.7	346.4	362.3	373.7

Remarks: The purpose of the study was to determine the toxicity of five pure homologues of LAS to the activated sludge of a treatment plant basically operating on domestic sewage. A contact time of 3 hours instead of 15 minutes was chosen to better simulate the real residence time used in wastewater treatment plants (4-6 hours). The EC₅₀ values are far above environmental concentrations and therefore provide a high margin of safety. The 3-hour EC₅₀ range for the reference substance (3,5-dichlorophenol) ranged from 20-30 mg/L, within the valid range of 5-30 mg/L.

Reference: Verge C. and Moreno, A. 1996b. Toxicity of anionic surfactants to the bacterial population of a waste water treatment plant. *Tenside Surf. Det.* 33:323-327.

Reliability: 2 Valid with restrictions

(c)

Type: Aquatic ; Field ; Soil ; Other

Species: activated sludge

Exposure Period: 15 minute

Results: EC₅₀ = 107-152 mg/L

Analytical monitoring: Yes No ?

Method: ESD-VIII-D-1, Issue II (9/8/80)

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS (CAS #68411-30-3)

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. Effect: inhibition of respiration. Nominal concentrations (expected deviation <20%). Mixed Liquor Suspended Solids 53.6-76.1 mg/g VSS/Sewage. Static, 25°C. Activated sludge (2600 mg SS/L)

Reference: European Commission. 2000s. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, 27896, 27897, 27915.

Reliability: 4 Not assignable. The original study was not available for review. Summary included for completeness.

(d)

Type: Aquatic ; Field ; Soil ; Other

Species: *Pseudomonas putida* (Bacteria)

Exposure Period: 18 hour
Results: $EC_{50} = 60.9-63.5$ mg/L
 $EC_{10} = 52.7-56.6$ mg/L
Analytical monitoring: Yes No ?
Method: Bacterial toxicity test according to DIN 38412 part 8. A total of 6 concentrations were tested (40-80 mg/L) under GLP conditions.
GLP: Yes No ?
Test substance: Marlon A 390 (CAS #68411-30-3) C_{10-13} LAS, average alkyl chain length = 11.6; activity 91.3%,
Remarks: Results show EC_{50} and EC_{10} values for two tests.
Reference: Scholz, N. 1993. Bestimmung der bacterientoxizitat von Marlon A 390 in Pseudomonas-zellvermehrungs-Hemmtest. Huels-Final Report No. PZ-93/10.
Reliability: 2 Valid with restrictions

(e)

Type: Aquatic ; Field ; Soil ; Other
Species: *Pseudomonas putida* (Bacteria)
Exposure Period: 30 minute
Results: $EC_{50} = 350$ mg/L
 $EC_0 = 250$ mg/L
Analytical monitoring: Yes No ?
Method: DIN 38412 Teil 27 (respiration inhibition test)
GLP: Yes No ?
Test substance: Marlon A 350 (CAS #68411-30-3) C_{10-13} LAS, average alkyl chain length = 11.6
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Data refer to 100% active ingredient.
Reference: European Commission. 2000b. Benzenesulfonic acid, C_{10-13} -alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).
Reliability: 4 Not assignable

(f)

Type: Aquatic ; Field ; Soil ; Other
Species: *Pseudomonas putida* (Bacteria)
Exposure Period: 16 hour
Results: $EC_{50} = 150$ mg/L
 $EC_0 = 50$ mg/L
Analytical monitoring: Yes No ?
Method: DIN 38412 Teil 8 (cell multiplication inhibition test)
GLP: Yes No ?
Test substance: Marlon A 350 (CAS #68411-30-3) C_{10-13} LAS, average alkyl chain length = 11.6
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Data refer to 100% active ingredient
Reference: European Commission. 2000b. Benzenesulfonic acid, C_{10-13} -alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).
Reliability: 4 Not assignable

(g)

Type: Aquatic ; Field ; Soil ; Other
Species: *Pseudomonas putida* (Bacteria)
Exposure Period: 30 minute
Results: NOEC = 64 mg/L
Analytical monitoring: Yes No ?
Method: DIN 38412, Teil 27
GLP: Yes No ?
Test substance: C₁₀₋₁₃ LAS, average chain length 11.8 (CAS #68411-30-3)
Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Synthetic water; static; pH 7.2⁺/-0.2; 20°C. Effect: Inhibition of oxygen consumption.
Reference: European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KgaA.
Reliability: 4 Not assignable

(h)

Type: Aquatic ; Field ; Soil ; Other
Species: *Pseudomonas putida* (Bacteria)
Exposure Period: 16 hour
Results: NOEC = 30 mg/L
Analytical monitoring: Yes No ?
Method: DIN 38412, Teil 8
GLP: Yes No ?
Test substance: C₁₀₋₁₃ LAS, average chain length 11.8 (CAS #68411-30-3)
Remarks: Synthetic medium; static; pH 7.4⁺/-0.3; 21°C. Effect: growth inhibition. Feitjel et al. reviews this study and also reports an NOEC = 35 mg/L for C_{11.6} LAS.
Reference: Feitjel, T.C.J., Matthijs, E., Rottiers, A., Rijs, G.B.J., Kiewiet, A., and de Nijs, A. 1995. AIS/CESIO environmental surfactant monitoring programme. Part 1: LAS monitoring study in “de Meer” STP and receiving river “Leidsche Rijn” Chemosphere 30:1053-1066.
Reliability: 4 Not assignable

(i)

Results: Based on digester performance, even a high and atypical concentration of LAS (30 g/kg) did not inhibit microbial populations present in STP activated sludge digesters.
Remarks: The study was designed to monitor LAS during the different steps of 9 different standard sewage treatment plants operating mainly with domestic sewage. LAS-specific analytical methods were used. More details on the study can be found in the summary at 3.2(f).
Reference: Berna, J.L., de Ferrer, J., Moreno, A., Prats, D. and Ruiz Bevia, F. 1989. The fate of LAS in the environment. Tenside Surf. Det. 26:101-107.
Reliability: 4 Not assignable. Treatment plant operational records are not available for review.

4.5 CHRONIC TOXICITY TO AQUATIC ORGANISMS

4.5.1 CHRONIC TOXICITY TO FISH

(a)

Type of Test: Static ; semi-static ; flow-through
 Open-system ; closed-system ; not stated

Species: *Brachydanio rerio* (Zebra Fish, fresh water)

Exposure Period: 14 days

Effect Criteria: Mortality, behavior

Results: NOEC = 2.0 mg/L
 LOEC = 4.0 mg/L

Analytic Monitoring: Yes ; No ; ?

Method: Based on UBA-Verfahrensvorschlag: Verlaengerter Toxizitaetstest beim Zebrabaerbling *Brachydanio rerio*, Bestimmung der Schwellenkonzentration der letalen und anderer Wirkungen, NOEC, mindestnes 14 Tage. This method conforms with OECD Guideline 204. Ten fish were exposed to each of seven concentrations (0.2, 0.4, 0.8, 1.6, 2.0, 4.0 and 8.0 mg/L) and the controls. Test chambers were 10-L basins containing 5-L of copper- and chlorine-free drinking water, maintained in a 16:8 light:dark illumination cycle. The NOEC normalized by van de Plassche et al. (1999) to C_{11,6} LAS was 2.3 mg/L.

GLP: Yes ; No ; ?

Test Substance: Marlon A 350 LAS (CAS #68411-30-3; Benzenesulfonic acid, C₁₀₋₁₃- alkyl derives., sodium salts, 25.7% activity)

Reference: Henkel KGaA, Biological Research and Product Safety/Ecology: unpublished results (Test substance number Fi 5959).

Reliability: 2 Valid with restrictions. Duration of test considered too short for a chronic study, therefore not included in SIAR Table 12A.

(b)

Type of test: static ; semi-static ; flow-through ; other
 open-system ; closed-system

Species: *Pimephales promelas* (Fish, fresh water)

Exposure period: 30 day

Effect criteria: Fry survival

Results: NOEC = 1 mg/L
 LOEC = 2 mg/L

Analytical monitoring: Yes No ? HPLC

Methods: Two replicates of 100 egg-fry stage fathead minnows were exposed for 30 days to LAS under the following conditions: Hardness 41 mg/L as CaCO₃; pH 7.2; temperature 24°C. The exchange rate was 1 to 3 volume changes/day. Test chambers were 3500 mL volume. The studies were conducted at EG&G Bionomics (now Springborn Smithers Laboratory).

GLP: Yes No ?

Test substance: Commercial C₁₀₋₁₃ LAS, sodium salt (CAS #68411-30-3); C₁₀ 5%, C₁₁ 27%, C₁₂ 53%, C₁₃ 13%; 2-phenyl 23%.

Remarks: Carboxylated intermediates formed in the biodegradation of LAS exhibit toxicity several orders of magnitude less than LAS; LC₅₀ values were >144 mg/L and >52 mg/L for sulfophenyl butarate and sulfophenyl undecanoate, respectively. NOEC based on fry survival. Egg hatchability and fry growth were less sensitive. This is a key study for chronic aquatic toxicity to fish (see SIAR Table 12).

Reference: Swisher, R.D., Gledhill, W.E., Kimerle R.A. and Taulli, T.A. 1978. Carboxylated intermediates in the biodegradation of linear alkylbenzene sulfonates (LAS). VII International Congress on Surface Active Substance, Proceedings, Moscow, 1976 4:218-230.

Reliability: 2 Valid with restrictions

(c)

Type of Test: Static []; semi-static [X]; flow-through []
Open-system [X]; closed-system []; not stated []

Species: *Poecilia reticulata* (Fish, Guppy, fresh water)

Exposure Period: 28 days

Effect Criteria: Mortality, behavior, and growth

Results: NOEC = 3.2 mg/L
LOEC = 10 mg/L

Analytic Monitoring: Yes []; No [X]; ? []

Method: Fish were 3-4 weeks old at test initiation. Fifty fish were used per group. Temperature was maintained at 23 ± 2°C. The test volume (10-L per chamber) was renewed three times per week. Circadian lighting (16:8 light:dark) was used. Fish were fed a Tetramin/Tetraphyll mixture. Dissolved oxygen, water hardness and pH were measured during the study.

GLP: Yes []; No []; ? [X]

Test Substance: LAS, 42.4% activity (C₈ <1%, C₉ 16.5%, C₁₀ 23%, C₁₁ 20%, C₁₂ 18%, C₁₃ 16%, C₁₄ 6.5%); average = C_{11.1}

Remarks: The only effect (98% mortality at 10 mg/L) occurred within 2 days of study initiation. The NOEC normalized by van de Plassche et al. (1999) to C_{11.6} LAS was 3.2 mg/L.

Reference: Canton, J.H. and Slooff, W. 1982. Substitutes for phosphate containing washing products: Their toxicity and biodegradability in the aquatic environment.

Reliability: 2 Valid with restrictions. Duration of test considered too short for a chronic study, therefore not included in SIAR Table 12A.

(d)

Type of Test: Static []; semi-static []; flow-through [X]
Open-system [X]; closed-system []; not stated []

Species: *Oncorhynchus mykiss* (Fish, Rainbow Trout, fresh water)

Exposure Period: 14 days

Effect Criteria: Mortality

Results: 14 day LC₅₀ = 0.12 mg/L

Analytic Monitoring: Yes []; No []; ? [X]

Method: Rainbow trout at different developmental stages (egg [160-200 eggs per concentration], alevin with partially absorbed yolk sac [2-3 days old], alevin [60 days old], and adult [270-300 days old]) were exposed to LAS for 14 days. Temperature was maintained at about 15°C, dissolved oxygen at greater than 80% of saturation, pH at 7.3-7.4, and water hardness of 290-310 mg/L as CaCO₃.

GLP: Yes []; No []; ? [X]

Test Substance: C₁₂ LAS, 45% activity

Remarks: The egg was the most sensitive life stage in the 14 day tests, in contrast to the 24 hour tests, which showed the egg to be the least sensitive (alevins with partially absorbed yolk sac were the most sensitive in the 24 hour tests). Malformations were seen only in embryos treated with lethal concentrations. A NOEC value was not determined in the study and the data provided are inadequate to calculate an EC₂₀ value.

Reference: Vailati, G., Calamari, D. and Marchetti, R. 1975. Effetti dell'alchilbenzene sofonato (LAS) sugli stadi di sviluppo del *Salmo gairdneri* Rich. Istituto di Ricerca sulle Acque (CNR) Sezione di Idrobiologia Applicata (Milano).
Reliability: 2 Valid with restrictions. Duration of test considered too short for a chronic study, therefore not included in SIAR Table 12A.

(e)

Type of Test: Static ; semi-static ; flow-through
Open-system ; closed-system ; not stated

Species: *Tilapia mossambica* (Fish, Tilapia, fresh water)

Exposure Period: 90 days

Effect Criteria: Feeding, growth rate, fecundity, yield

Results: NOEC = 0.25 mg/L
LOEC = 0.51 mg/L

Analytic Monitoring: Yes ; No ; ?

Method: Tests generally followed the standard methods of APHA 1975, with the following specifics. Tests were conducted in outdoor earthen vats (62 cm diameter, 30 cm mean depth) containing 60-L of borehole water and 5 kg of uncontaminated soil. Borehole water is unchlorinated water with the following parameters: pH 7.1 ± 0.1 , dissolved oxygen 10 mg/L, hardness 290 mg/L as CaCO₃, and temperature 27.9 ± 0.14 °C. Fifteen fish purchased from local farms (35 mm, 0.786 g) and acclimated to the test conditions for 168 hours were added per vat. Test concentrations were 0.25, 0.38, 0.51, and 1.10 mg/L. Fish were exposed six times at 15 day intervals with the water renewals and were fed daily with a 1:1 mixture of rice bran and mustard oil cake. Standard acute toxicity tests were also conducted in the laboratory. Statistical analysis was done using F and t tests and the significance of any change was measured at a 5% level of probability.

GLP: Yes ; No ; ?

Test Substance: LAS (Parnol J Liquid), 20% activity; clear yellow liquid; pH in solution was 8 ± 1

Remarks: The feeding rates decreased significantly at 0.25, 0.38 and 1.10 mg/L. Fish showed erratic behaviour, irregular opercular movement, and at higher concentrations, blood exuded from the base of the pectoral and pelvic fins and head. No apparent difference in condition factor (K) was observed at any concentration. The maturity index (MI) of both male and female fish appeared to decrease at all concentrations, but the biological significance of this is questionable because historic control values for this parameter were not provided and the magnitude of the response did not increase with dose. Fecundity decreased at 0.51 mg/L but not at 1.10 mg/L. The gastrosomatic index (GSI) was significantly different at 0.51 and 1.10 mg/L. Based on the most reliable endpoints (GSI and fecundity), the NOEC would be 0.38 mg/L and the LOEC would be 0.51 mg/L. However, the study is incompletely documented, so details of the test substance composition and testing procedure are uncertain. True replicates were not used so statistics can not be validly conducted, though they are reported by the authors. In view of these limitations, and previous evaluations of the study which have reported a NOEC of 0.25 mg/L (van de Plassche et al., 1999), a conservative (protective) NOEC for this study is 0.25 mg/L. This is a critical study for this SIDS endpoint.

Reference: Chattopahyay, D.N. and Konar, S.K. 1985. Acute and chronic effects of linear alkyl benzene sulfonate on fish, plankton, and worm. *Environment & Ecology*, 3:258-262.

Reliability: 2 Valid with restrictions.

(f)

Type of test: Various types and durations of tests.

Results: The article compiles the no observed effect concentration (NOEC) values for many tests conducted on an assortment of species. The following table shows the geometric mean NOEC values for each fish species (n = number of studies included for each species).

Species	Geometric mean NOEC (mg/L)	N
<i>Brachydanio rerio</i>	2.3	1
<i>Pimephales promelas</i>	0.87	14
<i>Poecilia reticulata</i>	3.2	1
<i>Oncorhynchus mykiss</i>	0.34	7
<i>Tilapia mossambica</i>	0.25	1

Remarks: All data were from tests conducted on commercial LAS with C₁₀₋₁₃ alkyl chains and average carbon lengths close to C_{11.6} and C_{11.8}. The NOEC values have been normalized using QSARs to the average structure of C_{11.6} LAS.

Reference: van de Plassche, E.J., DeBruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J., and Belanger, S.E. 1999. Predicted no-effect concentrations and risk characterization of four surfactants: Linear alkyl benzene sulfonate, alcohol ethoxylates, alcohol ethoxylated sulfates, and soap. *Environ. Toxicol. Chem.* 18: 2653-2663.

Reliability: 4 Not assignable This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(g) **(Rainbow trout)**

Type of test: static []; semi-static []; flow-through [X]; other []; open-system [X]; closed-system []

Species: *Salmo gairdneri* (*Oncorhynchus mykiss*, fish, estuary, fresh water)

Endpoint: Length of fish []; Weight of fish [];
Reproduction rate []; Other [X] Growth

Exposure period: 28 day

Results: NOEC = 0.43-0.89 mg/L

Analytical monitoring: Yes [] No [X] ? []

Method: Crossland, N O.

GLP: Yes [] No [] ? [X]

Test substance: C₁₀₋₁₃ LAS (CAS #68411-30-3)

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Mean NOEC: 0.62 mg/l (2 tests). Huels AG judged study quality to be good. Tap water with hardness 84-153 mg/L CaCO₃; pH 7.1-8.7; flow-through; 14-16°C; age of fish at start of study: 6 months.

Reference: European Commission. 2000k. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, CT/R153/01, CT/R153/06.

Reliability: 4 Not assignable. The original studies were not available for review.

(h)

Type of test: static ; semi-static ; flow-through ; other ; open-system ; closed-system

Species: *Salmo gairdneri* (Fish, estuary, fresh water)

Endpoint: Length of fish ; Weight of fish ; Reproduction rate ; Other Growth, Hatching, Survival

Exposure period: 70 day

Results: NOEC = 0.23 mg/L

Analytical monitoring: Yes No ?

Method: Unilever Research Protocol, Early Life Stage (ELS) test.

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS (CAS #68411-30-3)

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Huels AG judged study quality to be good. Tap water with hardness 70-133 mg/L CaCO₃, pH 7.3-7.8; flow-through; 8.5- 11.5°C; life-stage: ELS.

Reference: European Commission. 2000j. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, CT/R118/03.

Reliability: 4 Not assignable. The original study was not available for review.

(i)

Type of test: static ; semi-static ; flow-through ; other ; open-system ; closed-system

Species: *Salmo gairdneri* (Fish, estuary, fresh water)

Endpoint: Length of fish ; Weight of fish ; Reproduction rate ; Other Growth, Hatching, Survival

Exposure period: 70 day.

Results: NOEC = 0.3-0.35 mg/L

Analytical monitoring: Yes No ?

Method: Unilever Research Protocol.

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS (CAS #68411-30-3)

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Mean NOEC: 0.32 mg/l (2 tests). Huels AG judged study quality to be good. Nominal concentrations (expected <20%). Tap water with hardness 64-159 mg/L CaCO₃; pH 6.6-8.0; flow-through; 7.5-15 °C; life-stage: ELS

Reference: European Commission. 2000l. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Procter & Gamble, 1991, CT/R89/01, CT/R89/02.

Reliability: 4 Not assignable. The original studies were not available for review.

(j) **(Fathead Minnow)**

Type of test: static ; semi-static ; flow-through ; other ; open-system ; closed-system

Species: *Pimephales promelas* (Fish, fresh water)

Endpoint: Length of fish ; Weight of fish ; Reproduction rate ; Other Survival

Exposure period: 196 day

Results: NOEC = 0.63 mg/L; LOEC = 1.2 mg/L (based on effects on fry survival) Hatchability and growth were not significantly affected.

Analytical monitoring: Yes No ?

Method: Either a serial or proportional dilution unit was used to provide continuous exposures to fathead minnows. Each of the four test concentrations plus control received 12 randomly assigned fish obtained from ponds at the Newtown Fish Farm, Ohio Division of Wildlife. Pieces of half-tile were placed in each 10-gal aquarium for spawning sites. After spawning had been completed, the cluster of eggs was removed and counted. Four replicates of 100 eggs from each concentration were reared for 14 days and mortality of eggs and fry recorded daily. Mean dissolved oxygen, water hardness, and pH ranged from 5.84-6.42 mg/L, 194-214 mg/L CaCO₃, and 7.50-7.95, respectively. Test concentrations were 0.34, 0.63, 1.2 and 2.7 mg/L.

GLP: Yes No ?

Test substance: LAS, activity: 60.8%; equivalent MW = 348

Reference: Pickering, Q.M. and Thatcher, T.O. 1970. The chronic toxicity of linear alkylate sulphonate (LAS) to *Pimephales promelas* Rafinesque. Water Pollut. Control Fed. 42:243-254.

Reliability: 2 Valid with restrictions

(k)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system

Species: *Pimephales promelas* (Fish, fresh water)

Exposure period: 28 day

Results: NOEC (C_{11.8}) = 0.9 mg/L

NOEC (C₁₃) = 0.15 mg/L

Analytical monitoring: Yes No ?

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS (CAS #68411-30-3), average chain lengths 11.8 and 13

Remarks: Observations were made of the number of spawnings, total eggs produced, and number of eggs per female. Data were obtained from the literature.

Reference: Maki, A.W. 1979. Correlations between *Daphnia magna* and fathead minnow (*Pimephales promelas*), chronic toxicity values for several classes of test substances. J. Fish. Res. Bd Can. 36, 411-421.

Reliability: 4 Not assignable. This study was given a reliability score of 4 because the publication is a summary of previous tests and the original study reports were not available for review.

(l)

Type of Test: Static ; semi-static ; flow-through
Open-system ; closed-system ; not stated

Species: *Pimephales promelas* (Fish, Fathead Minnow, fresh water)

Exposure Period: Up to 30 days post-hatch

Effect Criteria: Embryolarval/fry survival

Results: NOEC = 1.02 mg/L

Analytic Monitoring: Yes ; No ; ?

Method: The publication summarizes the results of a series of critical life stage (embryolarval) tests, which are defined as exposure during the embryogenic period (incubation of the eggs), followed by exposure of fry for a period of 30-days after hatching for warm water fish with embryogenic periods ranging from 1 to 14 days.

GLP: Yes ; No ; ?

Test Substance: C_{11.7} LAS (commercial blend of C₁₀ 8%, C₁₁ 29%, C₁₂ 34%, C₁₃ 29%)

Remarks: All of the original studies summarized in this publication were conducted at EG&G Bionomics (now Springborn Smithers Labs) using standard protocols. The minimum threshold concentration, defined as the lowest concentration causing significant effect on any parameter, was reported as >1.02 mg/L <2.05 mg/L. The NOEC normalized by van de Plassche et al. (1999) to C_{11.6} LAS was 1.1 mg/L.

Reference: Macek, K.J. and Sleight, B.H. III. 1977. Utility of toxicity tests with embryos and fry of fish in evaluating hazards associated with the chronic toxicity of chemicals to fishes. Aquatic Toxicology and Hazard Evaluation, ASTM STP 634. Mayer, F.L. and Hamelink, J.L, Eds. American Society for Testing and Materials, pp. 137-146.

Reliability: 4 Not assignable. This study is given a reliability of 4 because the publication is a summary of previous tests and the original study reports were not available for review.

(m)

Type of test: static ; semi-static ; flow-through ; other
open-system ; closed-system

Species: *Lepomis macrochirus* (Fish, fresh water)

Exposure period: 28 day

Results: NOEC = 1.0 mg/L for bluegill
LOEC = 2.0 mg/L

Analytical monitoring: Yes No ?

Method: The effect of LAS on the structure and function of microbial communities was studied in a flow-through model ecosystem containing several trophic levels. Duplicate 19-L glass aquaria containing model ecosystems at four nominal concentrations (0.5, 1.0, 2.0, 4.0 mg/L) contained water and 2.5 cm lake sediment (Winton Lake, Cincinnati, OH) and several trophic levels (bacteria, algae, macrophytes [*Elodea canadensis*, *Lemna minor*], macroinvertebrates [*Daphnia magna*, *Paratanytarsus parthenogenica*], and fish [*Lepomis macrochirus*]). Flow rate in the proportional diluter delivered approximately 8 replacement volumes per day. Additionally, at each cycle of the diluter, 1.5 mL of a *Daphnia* food suspension diluted with a culture of *Selenastrum* was added to each chamber. Following an initial 3 day acclimation period to analytically confirm test concentrations, 4 glass periphyton slides (5 x 5 cm), 8 vegetative shoots of *Elodea*, 10 early instar *Daphnia*, 25 midge eggs and 5 pre-weighed juvenile bluegills (2.5-5.0 cm length) were added to each aquarium. Fish were screened from access to the macroinvertebrates by a 60 mesh stainless steel screen and were fed a daily supplement of frozen brine shrimp. Test duration was 28 days. Effects monitored included population and community effects. Nominal concentrations were confirmed with MBAS analysis.

GLP: Yes No ?

Test substance: LAS; C¹⁴-LAS chain length C₁₂ (91% purity) plus unlabeled LAS with average chain length C_{11.6} (C₁₀ 9.7%, C₁₁ 27.9%, C₁₂ 54.4%, C₁₃ 8.0%; 95% purity) (tested together)

Remarks: Dissolved oxygen concentrations ranged between 7.0 and 9.0 mg/L. Temperature was maintained at 2°C and the mean pH was 8.1 ± 0.2. Bluegill fish growth was reduced at the 2.0 and 4.0 mg/L concentrations but not at 0.5 or 1.0 mg/L. Results for the other species and community parameters tested are summarized in Section 4.7 (i). Juvenile growth was the most sensitive fish endpoint in this model ecosystem study and thus is

appropriate to use for chronic toxicity. This is a key study for chronic aquatic toxicity to fish (see SIAR Table 12).

Reference: Maki, A.W. 1981. A Laboratory model ecosystem approach to environmental fate and effects studies. Unpublished Internal Report, Environmental Safety Department Procter and Gamble Company, Cincinnati, Ohio.

Reliability: 2 Valid with restrictions

4.5.2 CHRONIC TOXICITY TO AQUATIC INVERTEBRATES

Water-only Exposures

(a)

Type of test: static []; semi-static []; flow-through []; open-system []; closed-system []; not stated [X]

Species: *Ceriodaphnia sp.* (Crustacea)

Endpoint: Mortality []; Reproduction [X]

Exposure period: not stated

Results: NOEC = 3 mg/L

Analytical monitoring: Yes [] No [] ? [X]

Method: Standard laboratory methods.

GLP: Yes [] No [] ? [X]

Test substance: C_{11.7} LAS

Remarks: Further information regarding study conditions was not provided in this peer-reviewed publication.

Reference: Kimerle, R.A. 1989. Aquatic and terrestrial ecotoxicology of linear alkylbenzene sulfonate. *Tenside Surfactants Detergents*, 26:169-176.

Reliability: 4 Not assignable. Data table cites an unpublished report by Procter & Gamble.

(b)

Type of test: static []; semi-static [X]; flow-through []; open-system []; closed-system [X]; not stated []

Species: *Ceriodaphnia sp.* (Crustacea)

Endpoint: Mortality [X]; Reproduction [X]

Exposure period: 7 day

Results: NOEC = 0.7 mg/L (geometric mean of two tests); 0.84 mg/L (normalized for C_{11.8} LAS)
NOEC = 0.5 mg/L (trout chow/algae diet)
EC₁₀ = 0.99 mg/L (yeast diet); normalized EC₁₀ = 1.18 mg/L

Analytical monitoring: Yes [] No [X] ? []

Method: ASTM (Comotto 1982; Mount and Norberg 1983). *Ceriodaphnia* were cultured individually in 50 mL beakers containing 30 mL culture water. Two tests were conducted, representing animals fed with two different diets (troutchow and algae – *Selenastrum capricornutum* – or baker's yeast). They were acclimated to the test conditions and diet for two generations before use in toxicity testing. Ten 50 mL beakers containing 30 mL of test solution were used for each test concentration. Nominal concentrations were 0.5, 1.0, 2.0, 3.5, 5.0 and 7.0 mg/L plus controls. Each beaker contained one *Ceriodaphnia* (for a total of 10 daphnids per concentration). Tests were begun with neonate animals (<24 hours old)

and lasted seven days. The young were counted and removed from each beaker daily. All test chambers were cleaned and renewed with fresh test solution three times (on the second, fourth, and sixth day). Total water hardness (Ohio river water) was 110 ± 9 mg/L as CaCO₃, pH was 7.4 ± 0.2 , dissolved oxygen was 9.7 ± 0.8 mg/L, and total suspended solids was 87 ± 106 .

GLP:

Yes [] No [X] ? []

Test substance:

C_{11.8} LAS; activity 30.8%

Results:

Ceriodaphnia fed trout chow/algae showed no dose-dependent response for any endpoint, so no EC₂₀ could be calculated. The NOEC is considered to be 5.0 mg/L based on 100% mortality at 7.0 mg/L. Results of the test run with the trout chow/algae diet are shown in the following table:

LAS Concentration	Percent Mortality	Total Reproduction	1 st Day of Reproduction	Reproduction per Individual	Broods	Brood Size
0	10	87	4.7	9.7	2.4	4.0
0.5	0	69	5.0	6.9	2.1	3.4
1.0	10	32	5.6*	3.6*	1.4*	2.4*
2.0	0	42	5.0	4.2*	1.8	2.2*
3.5	0	82	5.6*	8.2	2.0	4.2
5.0	0	105	4.7	11.6	2.3	4.9
7.0	100*	15	4.0	5.0*	--*	5.0

* Significantly different from the control (p<0.05)

Brood size and reproduction gave good dose-dependent responses for organisms fed yeast. Brood size was the most sensitive endpoint and resulted in an EC₂₀ of 1.44 mg/L. Reproduction resulted in an EC₂₀ of 2.70 mg/L. Results of the test run with the yeast diet are shown in the following table:

LAS Concentration	Percent Mortality	Total Reproduction	1 st Day of Reproduction	Reproduction per Individual	Broods	Brood Size
0	0	229	4.0	22.9	2.9	8.0
0.5	0	208	3.9	20.8	2.9	7.3
1.0	0	178	4.0	17.8	2.9	6.1*
2.0	10	145	4.0	16.1*	3.0	5.4*
3.5	10	78	4.4	8.7*	2.4	3.4*
5.0	10	8	6.6*	0.9*	0.6*	1.8*
7.0	100*	0	--*	--*	--*	--*

* Significantly different from the control (p<0.05)

Remarks:

Later experience with *Ceriodaphnia* has shown that a yeast diet is not optimum for this species. A geometric mean of 2.68 mg/L can be calculated for the two tests. This is a critical study for this SIDS endpoint.

Reference:

1) Procter & Gamble. 2004. *Ceriodaphnia* sp. Chronic toxicity test. Unpublished report, September 9, 2004.
 2) Comotto, R.M. 1982. Proposed standard practice for conducting renewal life cycle toxicity tests with *Daphnia magna*. Draft 1, August 1982, ASTM committee E-47. American Society for Testing and Materials, Philadelphia, PA.

- 3) Mount, D.I. and Norberg, T.J. 1983. A seven-day life cycle cladoceran toxicity test. Pre-publication. USEPA (Duluth).
- 4) Snedecor, G.W. and Cochran, W.G. 1967. Statistical Methods. 6th Edition. Iowa State University Press, Ames, IA.
- 5) Versteeg, D.J., Belanger, S.E., and Carr, G.J. 1999. Understanding single-species and model ecosystem sensitivity: Data-based comparison. Environ. Toxicol. Chem. 18:1329-1346.

Reliability: 2 Valid with restrictions (actual exposure concentrations might have been less than nominal values; not GLP)

(c)

Type of test: static []; semi-static []; flow-through [X]; open-system []; closed-system [X]; not stated [] diluter system based on Mount and Brungs 1967

Species: *Daphnia magna*. (Crustacea)

Endpoint: Mortality []; Reproduction [X]

Exposure period: 21 day

Results: NOEC = 1.18 mg/L
LOEC = Not reported

Analytical monitoring: Yes [X] No [] ? [] MBAS method

Method: A modified 0.5-L proportional diluter was used to deliver four replicates to each of five test concentrations plus a control. No solvent was used. Five *Daphnia* (<12 hours old) were randomly assigned to each replicate. *Daphnia* were fed a suspension of ground trout chow and alfalfa daily. The dilution water was carbon and reverse osmosis filtered well water. Water quality parameters were measured at test initiation and at intervals of 3-5 days for the remainder of the test. Tests were run at 21 ± 1 °C, dilution water hardness 120 mg/L as CaCO₃, pH 7.4 ± 0.2 , and dissolved oxygen 8.5 ± 0.5 mg/L. F₀ mortality was recorded at 24-h, 96-h, 7-d and daily thereafter. Total number of F₁ produced, mean brood size, and the percentage of days young were produced within each replicate was measured for all five concentrations and the controls. Mortality was evaluated using a computerized Probit procedure. The no effect concentration was determined as the highest measured concentration with no perceivable effects. This study was conducted in 1977.

GLP: Yes [] No [X] ? []

Test substance: C_{11.8} LAS; mean phenyl position 3.76; mean molecular weight = 345

Remarks: The most sensitive indicator of reproductive inhibition was the total number of young produced. This is a key study for chronic aquatic toxicity to invertebrates (see SIAR Table 12).

Reference: Maki, A.W. 1979. Correlations between *Daphnia magna* and fathead minnow (*Pimephales promelas*) chronic toxicity values for several classes of test substances. J. Fish. Res. Board Can. 36:411-421.

Reliability: 2 Valid with restrictions.

(d)

Species: *Daphnia magna*

Results: NOEC = 1.4 mg/L (12 records)

Test Substance: LAS normalized to C_{11.6}

Remarks: NOEC is geometric mean of 12 records compiled from literature reviews and normalized to C_{11.6}.

Reference: van de Plassche, E.J., de Bruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J. and Belanger, S.E. 1999. Predicted no-effect

concentrations and risk characterization of four surfactants: LAS, AE, AES, and soap. Environ. Toxicol. Chem. 18:2653-2663.

Reliability: 4 Not assignable. This study was given a reliability score of 4 because all the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(e)

Type of test: static ; semi-static ; flow-through ; open-system ; closed-system

Species: *Daphnia magna* (Crustacea)

Endpoint: Mortality ; Reproduction rate

Exposure period: 21 days

Results: NOEC = 1.25-3.25 mg/L; LOEC = 2.25-3.75 mg/L
Geometric Mean NOEC = 1.99 mg/L (mean of studies using 5 different diets)

Analytical monitoring: Yes No ?

Method: ASTM proposed standard practice for conducting renewal life cycle toxicity tests with *Daphnia magna*. Draft No. 1, August 1982. Ten 250 mL beakers were used for each test concentration. Seven beakers contained one daphnid each and three beakers contained five daphnids each, for a total of 22 daphnids per concentration. All conditions were maintained as per protocol.

GLP: Yes No ?

Test substance: Commercial C₁₀₋₁₃ LAS, average chain length C_{11.8} (CAS #68411-30-3).

Remarks: NOEC and LOEC values represent the range of results from five tests using different diets. Diet had at most a three-fold effect on the results, which is within the variation expected within the tests themselves. Therefore, results of different diets can be considered roughly equivalent to five replications of the same diet. This is a key study for chronic aquatic toxicity to invertebrates (see SIAR Table 12).

Reference: Taylor, M.J. 1985. Effect of diet on the sensitivity of *Daphnia magna* to linear alkylbenzene sulfonate. In: Cardwell, R.D., Purdy, R. and Bahner, R.C. Aquatic Toxicology and Hazard Assessment. Seventh Symposium pp. 53-72. ASTM STP 854, America Society for Testing and Materials, Philadelphia.

Reliability: 2 Valid with restrictions

(f)

Type of test: static ; semi-static ; flow-through ; open-system ; closed-system

Species: *Daphnia magna* (Crustacea)

Endpoint: Mortality ; Reproduction rate ; Other

Exposure period: 21 day

Results: NOEC = 0.3 mg/L

Analytical monitoring: Yes No ?

Method: OECD Guide-line 202, part 2 "*Daphnia* sp., Reproduction Test

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS, with average chain length of C_{11.8} (CAS #68411-30-3)

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. Natural water 3x weekly renewal pH 6.0-8.5; 20⁺/-2 °C; life-stage: 6-24 h.

Reference: European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA.

Reliability: 4 Not assignable. The original study was not available for review.

(g)

Type of test: static []; semi-static [X]; flow-through []; other []; open-system []; closed-system []

Species: *Daphnia magna* (Crustacea)

Endpoint: Mortality []; Reproduction rate [X]; Other []

Exposure period: 21 day

Results: NOEC = 0.3 mg/L

Analytical monitoring: Yes [] No [] ? [X]

Method: UBA – draft protocol

GLP: Yes [] No [X] ? []

Test substance: C₁₀₋₁₃, avg.: C_{11.6}

Remarks: Huels AG judged study quality to be good. Semistatic; pH 8.0; life-stage: adult.

Reference: European Commission. 2000d. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Huels AG, 2/87, N. Scholz, unpublished.

Reliability: 4 Not assignable. The original study was not available for review.

(h)

Type of test: static []; semi-static []; flow-through [X]; open-system []; closed-system [X]; not stated []

Species: *Paratanytarsus parthenogenica* (Insecta, Midge)

Endpoint: Reproduction and survival

Exposure period: 28 days

Results: NOEC = 3.4 mg/L (from previous study)

NOEC = >2.0-<4.0 mg/L (from model ecosystem community study)

LOEC = 4.0 mg/L

Analytical monitoring: Yes [X] No [] ? []

Method: Initial experiments (Phase I) were designed to determine the effects of LAS on the structure and function of model ecosystem communities. Each test concentration consisted of duplicate 19-L glass aquaria containing 2.5 cm of natural lake sediment and several trophic levels (bacteria, algae, macrophytes, macroinvertebrates, and bluegill sunfish). Dilution water was carbon and reverse-osmosis filtered well water of 120 mg/L as CaCO₃ hardness. Four nominal concentrations of 0.5, 1.0, 2.0, and 4.0 mg/L were delivered to duplicate chambers by a modified Mount and Brungs proportional diluter. Fish were screened from access to invertebrates with a stainless steel screen. In Phase II, conditions were similar except that the model ecosystem aquaria were treated with LAS in sewage effluent (supplied from a CAS unit) to more closely simulate actual receiving water conditions.

GLP: Yes [] No [X] ? []

Test substance: C₁₂ LAS

Remarks: Dissolved oxygen concentrations ranged between 7.0 to 9.0 mg/L with a mean of 7.8 mg/L during the Phase I studies. In Phase II, dissolved oxygen ranged between 3.1 and 7.3 mg/L (mean 5.4 mg/L), with the lowest values occurring in the chambers receiving the highest sewage effluent concentrations. Temperatures were maintained at 21 ± 2°C in both phases. The pH values were 8.1 ± 0.2 and 7.5 ± 0.3 for Phases I and II, respectively. No significant differences in the development and growth of

midge populations was observed in Phase I. Apparent inhibition of total population size was observed at the highest concentration (4.0 mg/L), where total individuals were 4100 as compared to 6300 in the controls. The results indicate that the effect concentrations after 28 days were found to be between 2.0 and 4.0 mg/L. This agrees with a previous 28-day study (Maki 1978) that resulted in a NOEC of 3.4 mg/L.

Reference: 1) Maki, A.W. 1981. A laboratory model ecosystem approach to environmental fate and effects studies. Procter & Gamble Company.
2) Maki, A.W. 1978. Development of a chronic toxicity test with the dipteran midge, *Paratanytarsus parthenogenica*. Presented at the Annual Meeting of the Entomological Society of America, Washington, DC, December.

Reliability: 2 Valid with restrictions.

(i)

Type of test: static ; semi-static ; flow-through ; open-system ; closed-system ; not stated

Species: *Brachionus calyciflorus* (rotifer)

Endpoint: Reproduction and survival

Exposure period: 2 days

Results: EC₁₀ = 1.18 mg/L

EC₂₀ = 1.4 mg/L (95% confidence intervals 0.882-2.27 mg/L)

EC₅₀ = 2.0 mg/L (95% confidence intervals 1.70-2.33 mg/L)

Analytical monitoring: Yes No ?

Method: Chronic toxicity tests were performed by placing six newly hatched (less than 3 h old), swimming rotifers in 10 mL of test water containing an equal mixture of green algae (a mixture of *Selenastrum capricornutum* and *Chlorella vulgaris*) at 1.0 x 10⁶ cells/mL as food. Three replicates were used for each concentration and control, with additional replicates used for analytical verification of the test compound as needed. Tests consisted of four to six concentrations and appropriate controls. Concentrations up to the limit of solubility were tested. All test vessels were placed on a rotator (1/5 rpm) in a 16/8 h light:dark cycle under low light conditions at 25±2 °C. Dilution water was a 50/50 blend of locally obtained well water and deionized water and had mean water quality properties of pH 8.6, dissolved oxygen 8.5 mg/L, hardness 152 mg/L as CaCO₃, and conductivity 450 µmhos. Rotifers were counted after 48 h in all control and test concentration replicates. Since rotifers produce multiple broods in 48 hours, the endpoint for this study is effects on reproduction. The 48 h EC₂₀ and EC₅₀ values with associated 95% confidence intervals were estimated by an iterative nonlinear regression technique using SAS, version 6.0.

GLP: Yes No ?

Test substance: C_{12,1} LAS, sodium salt; 92.3% purity

Remarks: LAS was one of about 20 surfactants tested in separate tests as part of this study. While test concentrations were measured, they were not reported in the publication. The authors do note that concentrations decreased by 20 to 90% over the two-day test (depending on which surfactant was tested) and time-weighted averages exposure concentrations were used. NOEC or LOEC values were not reported.

Reference: Versteeg, D.J., Stanton, D.T., Pence, M.A., and Cowan, C. 1997. Effects of surfactants on the rotifer, *Brachionus calyciflorus*, in a chronic toxicity

test and the development of QSARs. Environ. Toxicol. Chem. 16:1051-1058.

Reliability: 2 Valid with restrictions. Duration of test may be too short for a chronic study, therefore not included in SIAR Table 12A.

(j)

Type of test: static []; semi-static []; flow-through [X]; open-system [X]; closed-system []

Species: *Campeloma decisum* (freshwater mollusc; operculate snail)

Endpoint: Mortality [X]; Reproduction rate []; Other [X] mobility and feeding responses

Exposure period: 6 weeks

Results: NOEC = 0.4 mg/L
LOEC = 1.0 mg/L

Analytical monitoring: Yes [X] No [] ? []

Method: Two 6-week chronic studies were performed in which an amphipod, a pulmonate snail, and an operculate snail were tested together. A serial diluter was used to provide continuous flow conditions into duplicate 4-gallon glass aquaria for each of five concentrations plus controls. Ten snails were placed in each of two replicate chambers per concentration. The frequency of snails crawling on the test chamber walls was recorded throughout the tests. Mean measured LAS concentrations were 0.2, 0.4, 1.0, 1.9, and 4.4 mg/L.

GLP: Yes [] No [X] ? []

Test substance: Commercial LAS formulation containing 14% LAS, 2.3% alcohol ethoxylate oxide condensate, 2.5% sodium soap, 48% sodium tripolyphosphate, 9.7% sodium silicate, 15.4% sodium sulphate, and 8.1% moisture and miscellaneous.

Remarks: Survival of *C. decisum* was affected only in the highest test concentration (4.4 mg/L). Mobility and feeding responses were altered in LAS concentrations of 1.9 and 1.0 mg/L, respectively.

Reference: Arthur, J.W. 1970. Chronic effects on linear alkylbenzene sulfonate detergent on *Gammarus pseudolimnaeus*, *Campeloma decisum* and *Physa integra*. Water Res. 4:251-257.

Reliability: 4 Not assignable. Test substance is a formulation containing a mixture of materials, of which only 14% is LAS. The contribution of each component cannot be separately assigned.

(k)

Type of test: static []; semi-static []; flow-through [X]; open-system [X]; closed-system []

Species: *Physa integra* (freshwater mollusc; pulmonate snail)

Endpoint: Mortality [X]; Reproduction rate []; Other [X] mobility and feeding response

Exposure period: 6 weeks

Results: NOEC = 4.4 mg/L
LOEC = >4.4 mg/L

Analytical monitoring: Yes [] No [X] ? []

Method: Two 6-week chronic studies were performed in which an amphipod, a pulmonate snail, and an operculate snail were tested together. A serial diluter was used to provide continuous flow conditions into duplicate 4-gallon glass aquaria for each of five concentrations plus controls. The

frequency of snails crawling on the test chamber walls was recorded throughout the tests. Mean measured LAS concentrations were 0.2, 0.4, 1.0, 1.9, and 4.4 mg/L.

GLP: Yes No ?

Test substance: Commercial LAS formulation containing 14% LAS, 2.3% alcohol ethoxylate oxide condensate, 2.5% sodium soap, 48% sodium tripolyphosphate, 9.7% sodium silicate, 15.4% sodium sulphate, and 8.1% moisture and miscellaneous.

Remarks: No significant effects on survival, mobility, or feeding responses was observed in *P. integra* at any LAS concentration tested.

Reference: Arthur, J.W. 1970. Chronic effects on linear alkylbenzene sulfonate detergent on *Gammarus pseudolimnaeus*, *Campeloma decisum* and *Physa integra*. Water Res. 4:251-257.

Reliability: 4 Not assignable. Test substance is a formulation containing a mixture of materials, of which only 14% is LAS. The contribution of each component cannot be separately assigned.

(1)

Type of test: static ; semi-static ; flow-through ; open-system ; closed-system

Species: *Gammarus pseudolimnaeus* (freshwater amphipod)

Endpoint: Mortality ; Reproduction rate ; Other mobility and feeding responses

Exposure period: 6 weeks

Results: For adult survival: NOEC = 0.2 mg/L, LOEC = 0.4 mg/L
For reproduction: NOEC < 0.2 mg/L, LOEC = 0.2 mg/L

Analytical monitoring: Yes No ?

Method: Two 6-week chronic studies were performed in which an amphipod, a pulmonate snail, and an operculate snail were tested together. A serial diluter was used to provide continuous flow conditions into duplicate 4-gallon glass aquaria for each of five concentrations plus controls. Ten snails were placed in each of two replicate chambers per concentration. Complete immobilization was taken as a sign of death for the amphipods. Mean measured LAS concentrations were 0.2, 0.4, 1.0, 1.9, and 4.4 mg/L. Only adult amphipods were started in each 6-week test. Following the termination of the second 6-week test, however, the newly hatched *Gammarus* were counted and allowed to remain in each respective test chamber for 15 weeks of additional exposure.

GLP: Yes No ?

Test substance: Commercial LAS formulation containing 14% LAS, 2.3% alcohol ethoxylate oxide condensate, 2.5% sodium soap, 48% sodium tripolyphosphate, 9.7% sodium silicate, 15.4% sodium sulphate, and 8.1% moisture and miscellaneous.

Remarks: Adult survival was affected at all concentrations after 6-weeks in a somewhat dose-responsive manner, as shown in the following table. Based on adult survival, the NOEC is 0.2 mg/L and the LOEC is 0.4 mg/L.

Mean LAS Concentration (mg/L)	Duplicate Chambers	Trial 1	Trial 2
4.4	A	0	0

	B	0	0
1.9	A	10	0
	B	10	0
1.0	A	40	20
	B	30	30
0.4	A	30	20
	B	40	40
0.2	A	70	50
	B	40	50
Control	A	80	60
	B	70	40

Newly hatched amphipods were not produced in the highest concentration (4.4 mg/L). The results on survival of F₁ *Gammarus* from the second 6-week study, and the final numbers of gravid F₁ females and F₂ young produced after 15 weeks of exposure are shown below. Control F₁ females were the first to release F₂ young, and this occurred after 9 weeks. Females began liberation of F₂ young at 13 and 13.5 weeks for the 0.2 and 0.4 mg/L chambers, respectively. Based on variability and apparent reproductive effects at the lowest concentration, no NOEC value could be determined. The LOEC is 0.2 mg/L.

Mean LAS concentration (mg/L)	Duplicate Chambers	F ₁ Initial young numbers	Final F ₁ numbers		F ₁ Females		Number of births	Number F ₂ produced
			Males	Females	% Survival	Final number gravid		
4.4	A	0	0	0	0	0	0	0
	B	0	0	0	0	0	0	0
1.9	A	4	3	0	75	0	0	0
	B	9	3	3	66	0	0	0
1.0	A	32	3	4	22	1	0	0
	B	37	10	16	70	3	0	0
0.4	A	29	8	7	52	2	0	0
	B	59	25	24	83	4	1	32
0.2	A	52	11	9	38	8	2	45
	B	58	11	16	47	11	3	48
Control	A	77	26	22	62	5	6	109
	B	91	19	31	55	17	5	74

Reference: Arthur, J.W. 1970. Chronic effects on linear alkylbenzene sulfonate detergent on *Gammarus pseudolimnaeus*, *Campeloma decisum* and *Physa integra*. Water Res. 4:251-257.

Reliability: 4 Not assignable. Test substance is a formulation containing a mixture of materials, of which only 14% is LAS. The contribution of each component cannot be separately assigned.

(m)

Species: *Mysidopsis bahia* (marine mysid)

Results: NOEC = 0.12 mg/L (2 records)

Test Substance: LAS normalized to C_{11.6}

Remarks: NOEC is geometric mean of 2 records

Reference: van de Plassche, E.J., de Bruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J. and Belanger, S.E. 1999. Predicted no-effect concentrations and risk characterization of four surfactants: LAS, AE, AES, and soap, Environ. Toxicol. Chem. 18:2653-2663.

Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(n)

Type of test: static []; semi-static []; flow-through [X]; other []; open-system [X]; closed-system []

Species: *Gammarus pulex* (amphipod)

Endpoint: Mortality [X]; Reproduction rate [X]; Other []

Exposure period: Adults = 8 weeks; Juveniles = 15 weeks

Results: LC₅₀ (adults; 56 days) > 354 µg/L
 LC₅₀ (juveniles; 107 days) > 375 µg/L
 NOEC (number of offspring) = 97 µg/L

Analytical monitoring: Yes [X] No [] ? []

Method: Two separate groups of field-collected adults in the pre-copulatory amplexus and early juveniles were exposed to five concentration of LAS (nominally 30, 60, 120, 240, 480 µg/L) and control under flow-through conditions. The exposure vessel consisted of a plastic trough that was 40 cm by 10 cm. The water depth was approximately 3 cm with standpipe to drain and a 2 µm stainless steel piece of mesh was placed approximately 5 cm from the end of the vessel to prevent the loss of *Gammarus*. The solution was made up of a 1:1 mixture of pond water and carbon-filtered tap water. Mean measured concentrations for adult exposure were <1, 19, 35, 83, 176, 354 µg/L and for juvenile exposure were <1, 22, 36, 97, 141 and 375 µg/L. Survival and reproduction were recorded over an 8 week exposure for adults and a 15 weeks of exposure for juveniles. Offspring produced during the exposure were counted weekly but not removed from the test areas.

GLP: Yes [X] No [] ? []

Test substance: Commercial LAS, C₁₀₋₁₄ Sodium alkyl benzene sulfonate (CAS# 85117-50-6); Alkyl chain distribution: C₁₀ 10.3%, C₁₁ 34.6%, C₁₂ 32.7%, C₁₃ 21.6%, C₁₄ 0.9%; Average chain length 11.7.

Remarks: This study is considered invalid due to high control mortality, which was 23% and 37% for adult and juvenile exposures, respectively. Mortality of adults and juveniles exposed to test materials was in the same range, indicating that survival was not affected by exposure to LAS. Although the reproductive output at the highest two concentrations was lower than that in the lowest concentrations, it was not possible to conclude that this was due to LAS because the control reproduction was also lower. There was no significant difference in cumulative number of juvenile produced between the control and the highest concentration tested (354 µg/L). For the juveniles, the time of formation of pre-copular pairs was slower at the highest concentration tested, 375 µg/L. By day 86, the number of offspring produced at 141 and 375 µg/L was significantly lower than produced at the other LAS concentrations and control. The NOEC remained at 97 µg/L until the end of the study (day 107).

Reference: ERASM. 2000. Long-term toxicity of LAS on *Gammarus pulex*. Internal Report AISE/CESIO, Brussels.

Reliability: 3 Not valid due to excessive control mortality.

(o)

Type of test: static []; semi-static []; flow-through [X]; open-system []; closed-system [X]; not stated []

Species: *Chironomus riparius* (Insecta, Midge)

Endpoint: Survival and emergence

Exposure period: Approximately 24 day

Results: NOEC = 2.4 mg/L in water w/o sediment
LOEC = 3.72 mg/L in water w/o sediment

Analytical monitoring: Yes [X] No [] ? []

Method: Tests were conducted as an aqueous fraction in the absence of sediment. A flow-through diluter system delivered test material in water to glass containers with 120-140 cm² bottom surface area each. Intact egg masses were incubated in Petri dishes containing 20-30 mL of dilution water at 22 °C until hatching commenced. Newly hatched larvae were allowed to mature 72 hours before testing. Twenty larvae were randomly distributed to each duplicate test chamber for each of five test concentrations plus the controls. Larvae were fed daily until emergence of the first adult in each chamber. Tests were continued until each midge emerged as an adult or larvae were determined to be dead. The number of winged adults was recorded daily. The average test duration was 24 days. Total hardness, pH, dissolved oxygen, and temperature were monitored frequently during the test.

GLP: Yes [] No [] ? [X]

Test substance: C_{11.8} LAS; 30.4% activity; mean molecular weight = 346

Remarks: Adults typically emerged 12-14 days after hatching. Control values for adult emergence were similar to or exceeded the historical average observed in their laboratory (>90%). For comparison, additional flow-through studies were conducted with sediment from a naturally unspiked stream and using spiked sediments (see 4.5.2 (v)). The study with spiked sediments resulted in a NOEC of 319.0 ppm (LOEC = 993 ppm). This indicates that sorption onto sediment significantly mitigates LAS bioavailability. Thus, the water-only values above should be considered conservative. This is a critical study for this SIDS endpoint.

Reference: Pittinger, C.A., Woltering, D.M., and Masters, J.A. 1989. Bioavailability of sediment-sorbed and aqueous surfactants to *Chironomus riparius* (midge). Environ. Toxicol. Chem. 8:1023-1033.

Reliability: 2 Valid with restrictions.

Water and Sediment Exposures

(p)

Type of Test: Semi-static

Species: *Anodonta cygnea* (fresh water bivalve mollusc)

Endpoint: Mortality [X]; Reproduction rate []; Other [] growth, reproduction

Exposure period: 80 days

Results: NOEC ≥ 200 mg/kg dw

Analytical monitoring: Yes [X] No [] ? []

Method: Sediments were collected from a pond and LAS was sorbed to sediment by repeated additions for 80 days. Glass tanks containing a 4 cm thick

layer of spiked sediment and eight liters of dechlorinated tap water were used. A continuous water exchange allowed for a whole water mass change every 24 hours. Ten bivalves (10⁺-1 cm or larger diameter) were introduced into each tank. The LAS sorbed to sediment was 750 mg/kg dw at the beginning of the experiment and 200 mg/kg dw at the end of the experiment.

GLP: Yes No ?
Test Substance: Commercial LAS (unspecified)
Remarks: All animals survived the 80 day exposures and were actively filter-feeding without differences from the controls.
Reference: Bressan, M., Brunetti, R., Casellato, S., Fava, G.C., Giro, P., Marin, M., Negrisola, P., Tallandini, L., Thomann, S., Tosoni, L., Turchetto, M. and Campesan, G.C. 1989. Effects of linear alkylbenzene sulfonate (LAS) on benthic organisms. *Tenside Surf. Det.* 26:148-158.
Reliability: 2 Valid with restrictions (no GLP, statistical methods not described)

(q)
Type : Artificial soil ; Filter paper ; Other **Natural sediment**
Species: *Branchiura sowerbyi* (tubificid worm)
Endpoint: Mortality ; Weight ; Other **Reproductive Cycle**
Exposure period: 220 days
Results: NOEC ≥ 7.18 mg/kg.
Mean measured LAS concentrations in the sediment were 26, 9.8, and 7.18 mg/kg at 0, 45, and 220 days of exposure. No significant differences were observed between treated and control sediments for any parameter (survival, timing and percentage of cocoons, percent of hatching worms, number of eggs per cocoon, and mean embryonic development time). Total number of cocoons was slightly higher in treated worms than cocoons.
Method: Twenty specimens in duplicate were used for both the control and treated sediments for testing the possible effects of a long exposure to LAS adsorbed on sediment to oligochaetes. Worms with a well known reproductive cycle were collected and maintained in the dark at 15°C in a glass container with natural sediment for at least two weeks before the experiment. The sediment for the experiments was gently washed with deionized water and dried at 70°C. It was organized by grain size, carbonate and organic carbon contents. It was then treated in order to obtain the irreversibly adsorbed concentration of LAS.
The test substance and sediment mixture were equilibrated for 6 hours on a rotary shaker and then allowed to settle for 48 hours. Twenty eight washes were made with deionized water to ensure the sediment did not release any methylene blue material to the overlaying water. The residual concentration was checked after every wash in the overlaying water of desorbed LAS using the MBAS method. When an ~0 was observed, it was considered that the irreversibly adsorbed quantity of LAS in sediment was attained. This sediment was used in the experiment. The concentration of LAS in sediment was measured by HPLC at 0, 45, and 220 days. Endpoints measured included the number of cocoons, number of oocytes per cocoon, total number of oocytes, period of embryonic development, percent of degenerated cocoons, and percent of hatching worms.
GLP: Yes No ?

Test substance: LAS solution (1000 mg/kg) mixed with 200 g dried sediment (from EniChem Augusta Industriale S.A.)

Remarks: The LAS concentration in sediment used did not produce any effect on *B. sowerbyi* during the 220 days of exposure. The authors conclude that LAS absorbed on sediment has a much lower influence on the examined worms than LAS dissolved in water. The initial concentration of LAS in treated sediments was 25.87 mg/kg (3.99 mg/kg in control). After 45 days, a reduction of 62-63% of the nominal concentration was measured. After 220 days, the reduction reached 72%.

Reference: Casellato, S., Aiello, R., Negrisolo, P.A., and Seno, M. 1992. Long-term experiment on *Branchiura sowerbyi* Beddard (Oligochaeta, Tubificidae) using sediment treated with LAS (Linear Alkylbenzene Sulphonate). *Hydrobiologia* 232:169-173.

Reliability: 4 Not assignable. Documentation is incomplete, including identification of the structure, description of methods, lack of statistics, etc.

(r)

Type : Artificial soil [] ; Filter paper [] ; Other [X] **Spiked Sediment**

Species: *Lumbriculus variegatus* (Oligochaete)

Endpoint: Mortality [] ; Weight [] ; Other [X] **Survival, Reproduction, and Growth**

Exposure period: 28 days

Results: LC₅₀ (28 d) ≥ 105 mg/kg soil dry weight (see table)
NOEC = 81 mg/kg soil dry weight

Method: A 28 day chronic study was conducted using sediment spiked with radio-labelled material. The test species, *Lumbriculus variegatus*, is a true sediment feeder (i.e., subsurface ingestion of sediment particles). The nominal concentrations were 50, 75, 100, 150, 300, 600 mg/kg/dry weight and controls. The test sediment contained 44% sand, 48% silt, and 8% clay. Twenty grams (wet weight) of the prepared sediment was added to clean 60 mL glass vessels followed by 30 mL of groundwater drawn from an aquifer. After 24 hours of equilibration, 10 mature *Lumbriculus* (ca. 15 mm in length, 8 mg dry weight) were added to each vessel. Vessels were aerated for 5 minutes every day and the overlying water replenished with distilled water every two days. Each test concentration was replicated 6 times. LAS concentrations were measured at 0 and 28 days. After 28 days the sediment was removed and all live worms counted, blotted dry, and wet weighed prior to air drying for 48 hours to a constant dry weight. Toxicity endpoints included survival, reproduction and biomass. The mode of reproduction (architomy) necessitates the treatment of survival and reproduction as a single endpoint, i.e., number of organisms at test termination. Sediment concentrations were monitored using LSC and verified with HPLC.

GLP: Yes [X] No [] ? []

Test substance: LAS (Procter & Gamble), average alkyl chain length C_{11.4}. The radio-labelled LAS was 3-dodecylbenzene sulfonate (DOBS; 95% purity)

Results: There was a loss of between 15 and 78% of the LAS radioactivity over the duration of the test, which was attributed to mineralization of LAS by the worms and microorganisms present in the sediment (biodegradation). Results are therefore based on the average of day 0 and day 28 measured sediment concentrations. All results are shown in the following table.

Survival Endpoint	Sediment Concentration (mg/kg dw)			
	NOEC	LOEC	EC ₂₀	EC ₅₀
Based on nominal values	100	150	90	136
Based on measured day 0 values	136	170	131	164
Based on mean of days 0 & 28 values	81	110	73	105
Biomass Endpoint				
Based on nominal values	100	150	108	144
Based on measured day 0 values	136	170	146	166
Based on mean of days 0 & 28 values	82	110	102	109

Remarks: LAS half-life in aerobic sediment was approximately 20 days. This is shorter than studies conducted in the same sediment without worms (half-life of 38 days), most likely due to increased bioturbation due to worm activity. No specific endpoint was particularly sensitive to LAS.

Reference: Comber, S.D.W., Conrad, A.U., Hurst, K., Hoss, S., Webb, S., and Marshall, S. 2004. Chronic toxicity of sediment-associated linear alkylbenzene sulphonates (LAS) to freshwater benthic organisms. Manuscript in preparation.

Reliability: 2 Valid with restrictions

(s)

Type : Artificial soil ; Filter paper ; Other **Spiked Sediment**

Species: *Caenorhabditis elegans* (Nematode)

Endpoint: Mortality ; Weight ; Other **Survival, Fertility, Egg Production**

Exposure period: 3 days

Results: LC₅₀ (3 d) > 100 mg/kg soil dry weight
NOEC = 100 mg/kg soil dry weight (egg production)

Method: A 3 day chronic study was conducted using sediment spiked with cold-material LAS. Nominal concentrations were in the range of 10 to 1,000 mg/kg/dw. The test species is an infaunal bacterial feeder with a short life cycle, so 72 hours (3 days) is considered a chronic test. The nominal concentrations were 50, 75, 100, 150, 300, 600 mg/kg/dry weight and controls. The test sediment contained 44% sand, 48% silt, and 8% clay, with 2% organic matter. At the start of the test, ten juvenile worms of the first stage (270 ± 16 µm body length) were transferred to each test vial containing 0.75 g wet weight of spiked sediment mixed with 0.25 mL of a bacterial suspension. Five replicates were set up for each treatment, and the samples were incubated on a shaker at 20°C. After 72 hours the test was stopped by heat-killing the worms at approximately 50°C. The samples were mixed with an aqueous solution of rose Bengal to stain the worms for easier recovery. Sublethal toxicity endpoints were determined for growth based on the body length of the organisms, and fecundity by counting the number of eggs in the body of the test organism (egg production). The test was regarded as valid as the fertility of the test organisms in the control was ≥ 80%.

GLP: Yes No ?

Test substance: LAS (Procter and Gamble), average alkyl chain length C_{11.4}.

Results: Results are shown in the following table.

Test Parameter	Nominal Sediment Concentration (mg/kg dw)			
	NOEC	LOEC	EC ₁₀	EC ₃₀
Growth	200	300	275	
Fertility	200	300		258
Egg Production	100	200		125

Remarks: For growth, with a low variability, an EC₁₀ was chosen, whereas it was more appropriate to use an EC₃₀ for the more variable parameters fertility and egg production. Egg production was the most sensitive endpoint. Toxicity values might be underestimated slightly, as threshold values were calculated using nominal concentrations (chemical analysis was only performed for selected test concentrations). The chemical analysis at the end of the test showed values 73 to 80% of the initial, nominal concentrations, which equates to mean exposure concentrations of 87 to 90% for the nematodes during the test.

Reference: Comber, S.D.W., Conrad, A.U., Hurst, K., Hoss, S., Webb, S., and Marshall, S. 2004. Chronic toxicity of sediment-associated linear alkylbenzene sulphonates (LAS) to freshwater benthic organisms. Manuscript in preparation.

Reliability: 2 Valid with restrictions

(t)

Type of test: Various types and durations of tests

Results: The two articles compile the no observed effect concentration (NOEC) values for many tests conducted on an assortment of marine species. The following table shows the geometric mean NOEC values for each species for marine invertebrates, as well as one fish and two algae species.

Genus (and species)	Geometric mean NOEC (mg/L)
<i>Limanda (yokohamae)</i>	0.05
<i>Arbacia</i>	0.45
<i>Chaetopterus</i>	0.45
<i>Asterias</i>	0.35
<i>Mysidopsis (bahia)</i>	0.12
<i>Mysidopsis</i>	0.20
<i>Crassostrea (virginica)</i>	0.025
<i>Crassostrea</i>	0.04
<i>Mytilus (edulis)</i>	0.025
<i>Mytilus</i>	0.04
<i>Arcatia</i>	0.30
<i>Botrylloides</i>	1.94
<i>Molgula</i>	0.90
<i>Spisula</i>	0.80
<i>Botryllus</i>	0.75
<i>Laminaria</i>	5.00
<i>Dunaliella</i>	0.11

Remarks: All data were from tests conducted on commercial LAS with C₁₀₋₁₃ alkyl chains and average carbon lengths of C_{11.6} and C_{11.8}. The NOEC values have been normalized using QSARs to the average structure of C_{11.6} LAS.

Reference: 1) Temara, A., Carr, G., Webb, S., Versteeg, D. and Feijtel, T. 2001. Marine risk assessment: Linear alkylbenzenesulphonates (LAS) in the North Sea. *Marine Pollution Bulletin* 42:635-642.
2) van de Plassche, E.J., DeBruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J., and Belanger, S.E. 1999. Predicted no-effect concentrations and risk characterization of four surfactants: Linear alkyl benzene sulfonate, alcohol ethoxylates, alcohol ethoxylated sulfates, and soap. *Environ. Toxicol. Chem.* 18: 2653-2663.

Reliability: 4 Not assignable. This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(u)

Type: static []; semi-static [X]; flow-through []; other []; open-system []; closed-system []

Species: *Mytilus galloprovincialis* (marine mussel)

Endpoints: Other. filtration rate, oxygen uptake, nitrogen excretion

Exposure period: 7 days

Results: NOEC = 32.19 mg/kg dry weight (geometric mean of initial [132 mg/L] and final [7.85 mg/L] LAS concentration)

Analytical Monitoring: Yes

Method: Thirty mussels (5.8 cm average main axis length) collected from a mussel cultivation area of the Lagoon of Venice, Italy were divided into groups of 10 and placed in net tubes in a 60-L tank for 7 days in contact with 50 mg/L continuously suspended LAS-spiked sediments. Mussels were fed an algal suspension and water, food, and sediment were renewed daily. Filtration rate was determined twice a day (immediately before and after water changes) as defined by the volume of water cleared of algal particles/animal/hour. Faeces were collected daily and pooled for analysis of LAS concentrations by HPLC. Oxygen consumption and ammonia excretion rates were determined at the end of each treatment.

GLP: Yes [] No [X] ? []

Test substance: Commercial LAS; likely average alkyl chain length = C_{11.6}

Remarks: No significant differences in survival or physiological responses between treatments and controls were observed. The LAS concentration in treated sediments decreased by about 90% over the duration of the study (mean 132 mg/kg at initiation to mean 7.85 mg/kg at completion).

Reference: Marin, M.G., Pivotti, L., Campesan, G., Turchetto, M. and Tallandini, L. 1994. Effects and fate of sediment-sorbed linear alkylbenzene sulphonate (LAS) on the bivalve mollusk *Mytilus galloprovincialis* Lmk. *Wat. Res.* 28:85-90.

Reliability: 2 Valid with restrictions

(v)

Type of test: static []; semi-static []; flow-through [X]; open-system []; closed-system [X]; not stated []

Species: *Chironomus riparius* (Insecta, Midge)

Endpoint: Emergence

Exposure period: Approximately 24 day

Results: NOEC = 319 ppm in sediment
 LOEC = 993 ppm in sediment

Analytical monitoring: Yes No ?

Method: Tests were conducted as an aqueous fraction in the presence of sediment. Natural stream sediments (71% clay, 19% fine silt, 4% medium sand, 6% fine sand) were collected from a pristine site in Rapid Creek, SD. Before testing, wet sediment was autoclaved for 40-60 minutes to reduce microbial populations and minimize initial rates of surfactant biodegradation. LAS was added to a sediment slurry at a nominal concentration and stirred overnight, then 350 g was poured into each test chamber and allowed to settle. The organic carbon content of the test sediment was 4.2% prior to testing. A flow-through diluter system delivered test material in water to glass containers with 120-140 cm² bottom surface area each. Test concentrations were control, 8, 42, 146, 319, and 993 ppm. Intact egg masses were incubated in Petri dishes containing 20-30 mL of dilution water at 22 °C until hatching commenced. Newly hatched larvae were allowed to mature 72 hours before testing. Twenty larvae were randomly distributed to each duplicate test chamber for each of five test concentrations plus the controls. Larvae were fed daily until emergence of the first adult in each chamber. Tests were continued until each midge emerged as an adult or larvae were determined to be dead. The number of winged adults was recorded daily. The average test duration was 24 days. Total hardness, pH, dissolved oxygen, and temperature were monitored frequently during the test.

GLP: Yes No ?

Test substance: C_{11.8} LAS; 30.4% activity; mean molecular weight = 346

Remarks: Adults typically emerged 12-14 days after hatching. Control values for adult emergence were similar to or exceeded the historical average observed in their laboratory (>90%). Percent emergence was 98, 95, 90, 90, 90, and 73 for the control, 8, 42, 146, 319, and 993 ppm concentrations, respectively. For comparison, additional flow-through studies were conducted without sediment (see 4.5.2 (o)). Results indicate that sorption onto sediment significantly mitigates LAS bioavailability.

Reference: Pittinger, C.A., Woltering, D.M., and Masters, J.A. 1989. Bioavailability of sediment-sorbed and aqueous surfactants to *Chironomus riparius* (midge). Environ. Toxicol. Chem. 8:1023-1033.

Reliability: 2 Valid with restrictions.

4.6 TOXICITY TO TERRESTRIAL ORGANISMS

4.6.1 TOXICITY TO SOIL DWELLING ORGANISMS

(a)

Species/Endpoints: See table

Results: The following table shows the available NOEC, EC₁₀ and EC₅₀ values for eleven soil dwelling invertebrate species (in mg/kg dry weight).

Species	Endpoint	NOEC	EC ₁₀	EC ₅₀
<i>Eisenia foetida</i>	Reproduction		383	558
<i>Lumbricus terrestris</i>	Weight	667		
<i>Aporrectodea caliginosa</i>	Reproduction		14	129

Species	Endpoint	NOEC	EC ₁₀	EC ₅₀
<i>Aporrectodea longa</i>	Reproduction		27	137
<i>Folsomia fimetaria</i>	Reproduction		96	442
<i>Folsomia candida</i>	Reproduction		18	91
<i>Hypoaspis aculeifer</i>	Reproduction		81.7	236
<i>Enchytraeus albidus</i>	Reproduction		6.2	40.5
<i>Platynothrus peltifer</i>	Reproduction	320		467
<i>Isotoma viridis</i>	Growth		41	
<i>Hypogastrura assimilis</i>	Reproduction		99.8	421

Test Substance: LAS (unspecified)
Remarks: Values were extracted from a variety of original references and compiled for this article.
Reference: Jensen, J., Lokke, H., Holmstrup, M., Krogh, P.H. and Elsgaard, L. 2001. Effects and risk assessment of linear alkylbenzene sulfonates in agriculture soil. 5. Probabilistic risk assessment of linear alkylbenzene sulfonates in sludge-amended soils. Environ. Toxicol. Chem. 20:1690-1697.
Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(b)
Type : Artificial soil []; Filter paper []; Other [X] **Sandy, agricultural soil**
Species: *Enchytraeus albidus*, *Aporrectodea caliginosa*, *Aporrectodea longa*, *Folsomia fimetaria*, *Hypogastrura assimilis*, and *Hypoaspis aculeifer*
Endpoint: Mortality [X]; Weight [X]; Other [X] **Reproduction**
Exposure period: 21 days (28 days for *A. caliginosa* and *A. longa* growth tests)
Results: The following table shows the results of all tests. All values are nominal LAS concentrations in mg/kg dry weight.

Species	Parameter	NOEC	LOEC	LC ₁₀ or EC ₁₀	LC ₅₀ or EC ₅₀	Reliability Rating	Rationale for Reliability Rating
<i>Enchytraeus albidus</i>	Survival, adults	198	397	194	430	1	Draft ISO/WD 16387 protocol
	Reproduction	20	40	6	41	1	
<i>Aporrectodea caliginosa</i>	Survival, adults	278	793	329	535	2	Comparable to ISO 11268-2 Deviations; limited cocoons Comparable to ISO 11268-2, but with only weight measurement
	Cocoon production	>793	>793	14	129	3	
	Survival, juveniles	>397	>397	>397	>397	2	
	Growth, juveniles	278	397	105	354	2	
<i>Aporrectodea longa</i>	Survival, adults	278	793	329	535	2	Comparable to ISO 11268-2 Deviations; limited cocoons Comparable to ISO 11268-2, but with only weight measurement
	Cocoon production	>793	>793	27	137	3	
	Survival, juveniles	397	793	296	517	2	
	Growth, juveniles	79	278	84	349	2	
<i>Folsomia fimetaria</i>	Survival, adults	>793	>793	>793	>793	1	Comparable to ISO 11267
	Reproduction	278	278	85	424	1	
<i>Hypogastrura assimilis</i>	Reproduction	79	278	99	421	1	Comparable to ISO 11267
<i>Hypoaspis aculeifer</i>	Survival, adults	>793	>793	>793	>793	2	No guideline available
	Reproduction	278	793	82	236	2	

Method:

The effect of LAS on six species of soil invertebrates was determined using the following methods.

Earthworm tests:

No internationally accepted guideline is available for *A. caliginosa* and *A. longa*. For the reproduction tests, 1 kg dry weight of soil was carefully mixed with 160 mL of LAS solution using an electric mixer and filled into plastic pots. The six treatments consisted of one control and five concentrations of LAS and these treatments were randomly assigned to the experimental units. After 24-hour equilibration of the test soil, 3 (rather than 10) earthworms were added to closed containers with perforated lids for ventilation. Approximately 5 g per worm were added after the test animals had been introduced. The containers were then incubated for 21 days in darkness and the contents were later wet sieved through a 1-mm mesh. Water content was adjusted after 14 days.

For the growth test with juvenile *A. caliginosa* (2-3 weeks old), 60 g dry weight of soil were mixed with 9.6 mL of LAS solution with a spatula and filled into 160-mL polyethylene beakers with perforated lids for ventilation. The six treatments consisted of one control and five concentrations of LAS and these treatments were randomly assigned to the experimental units. After 24-hour equilibration of the test soil, one earthworm was added to each container. The beakers were incubated for 28 days in darkness and then the earthworms were recovered and their guts were cleared. The surviving animals were dried for 24 hours and their dry weight was recorded to the nearest 0.1 mg. The examination of the effects on growth of *A. longa* used the same method except the test period was 42 days.

Enchytraeid test:

The enchytraeid reproduction test followed a previously described protocol (draft ISO/WD 16387) using the potworm (*Enchytraeus albidus*). Forty grams dry weight of soil were mixed with 6.4 mL of LAS solution and filled into 160-mL beakers with perforated lids for ventilation. After 24-hour equilibration of the test soil, 10 adult *E. albidus* were added to each container and incubated in darkness for 21 days. After incubation, the surviving adult animals were removed from the soil. Now only containing cocoons, the soil was incubated in the beakers for another 21 days to allow development and hatching of the juveniles. After this period, the soil containing juveniles was stained with Bengal red, and water was added to facilitate counting of the juveniles. The test concentrations were not provided but can be estimated from Figure 5 to be 0, 20, 40, 80, 200 and 400 mg/kg with the numbers of adults surviving per replicate to be approximately 10, 10, 10, 10, 9, and 6, and the numbers of juveniles per replicate (reproduction) to be approximately 77, 50, 37, 21, 0, and 0, respectively.

Springtail tests:

No internationally accepted guideline is available for springtail reproduction. Effects of reproduction of *F. fimetaria* were determined using a method described by Wiles and Krogh. Twenty-seven grams dry weight of soil were mixed with 3 mL of LAS solution and filled into cylindrical test containers with lids. The bottom of the cylinder consisting of a 1-mm mesh to allow later extraction of the test animals. The mesh was covered with a layer of plastic film to prevent escape of the test animals. Adult, rather than juvenile, springtails were used. Ten male and

ten female *F. fimetaria* (23-26 days old) were added to the test containers after 24-hour equilibration of the test soil. The containers were incubated for 21 days with 12:12 photoperiod (h). After incubation, the animals were extracted using MacFadyen high-gradient extraction and the number of offspring counted. The same procedure was used for the springtail species *H. assimilis* Krausbauer, using ten male and ten female adults (16-19 days old). The test concentrations were not listed but can be estimated from Figure 6 to be 0, 10, 25, 75, 275, and 800, with the numbers of adult surviving per replicate to be approximately 145, 155, 115, 110, 165, and 165 and the numbers of juveniles per replicate (reproduction) to be approximately 285, 275, 190, 145, 205, and 10.

Predacious mite test:

No internationally accepted guideline is available for mite reproduction. Effects on reproduction on the predacious mite (*H. aculeifer*) were examined according to a method described by Krogh and Axelsen. A total of 54 g dry weight of soil was mixed with 6 mL of LAS solution and filled into test containers (as described for springtails). Ten female and five male *H. aculeifer* (16-19 days old) were added to each test container together with 100 *F. fimetaria* (16-19 days old) serving as prey for the mites. Incubation and extraction of mite offspring followed the same procedure as described for springtails.

A natural sandy, agricultural soil was used for all tests, rather than synthetic test soil. Nominal concentrations of LAS for some tests were verified by chemical analysis using HPLC.

GLP:

Yes No ?

Test substance:

C₁₀₋₁₃ LAS was obtained as an aqueous sodium salt solution with an active matter concentration of 16.1% (w/w), average molecular weight = 342 g/mol, distribution of the linear alkyl chains: C₁₀ 14%, C₁₁ 34%, C₁₂ 31%, and C₁₃ 21%; average alkyl chain length = C_{11.6}

Remarks:

Toxic effects on reproduction and growth were revealed when the concentration in soil exceeded 40 to 60 mg/kg dry weight. Reproduction was approximately fourfold more sensitive in earthworms and enchytraeids than in springtails and mites. It is argued that this difference in sensitivity is related to the dependency of soil pore water, which is high in the annelids but comparatively low in the arthropods. It should be noted that these studies report worst case exposures due to the use of a sandy test soil and the fact that LAS was added as an aqueous solution to the soil. In addition, too few replicates were used for the ECx approach (e.g., <5 controls) and several key deviations from draft protocols limited the reliability of endpoints for some studies (e.g., *A. caliginosa* and *A. longa* cocoon production). Nominal concentrations were derived from tables and figures since actual values were not found in the text.

Reference:

Holmstrup, M. and Krogh, P.H. 2001. Effects and risk assessment of linear alkylbenzene sulfonates in agricultural soil. 3. Sublethal effects on soil invertebrates. *Environmental Toxicology and Chemistry* 20:1673-1679.

Reliability:

See table for reliability of individual endpoints by species.

(c)

Type :

Artificial soil ; Filter paper ; Other

Species:

Eisenia foetida (Worm (Annelida), soil dwelling).

Endpoint:

Mortality ; Weight ; Other

Exposure period: 14 day
 Results: LC₅₀ >1000 mg/kg
 NOEC = 250 mg/kg soil dw
 Method: OECD Guideline 207, 1984. Ten adult worms (mean wt. 0.66 g/animal) were placed into each of four glass jars per concentration with soil comprised of 70% 5010 grade silica sand, 20% kaolinite clay, and 10% finely ground sphagnum peat. Nominal test concentrations in the soil were 1000, 500, 250, 125, 63 and 0 mg/kg dry weight. Temperature was maintained at 20⁺/-2°C with 24-hour continuous lighting at 600 lux. Earthworms were assessed for mortality, general health, body weight, and behavior after 7 and 14 days.
 GLP: Yes No ?
 Test substance: LAS (commercial blend) with average alkyl chain length C_{11.6} (typical of LAS chain lengths found in the environment).
 Remarks: No significant mortality was observed at the highest nominal concentration of 1000 mg/kg. A 33% and 23% reduction in body weight was observed at 100 and 500 mg/kg vs. a 14% reduction for the controls. Based on statistical analysis of the weight data, the no effect concentration was the nominal 250 mg/kg dose, which was confirmed by HPLC to be 235 mg/kg.
 Reference: Mieure, J.P., Waters, J., Holt, M. and Matthijs, E. 1990. Terrestrial safety assessment of LAS. Chemosphere 21:251-262.
 Reliability: 2 Valid with restrictions

(d)

Type : Artificial soil ; Filter paper ; Other
 Species: *Lumbricus terrestris* (soil dwelling worm)
 Endpoint: Mortality ; Weight ; Other
 Exposure period: 14 day
 Results: LC₅₀ >1333 mg/kg soil dw
 NOEC = 667 mg/kg soil dw
 Method: U.S.FDA Environmental Assessment Technical Guide No. 412, 1987. Ten adult earthworms (mean wt. 3.2 g/animal) were placed in each of four replicate one-gallon glass jars for each test concentration. Nominal test concentrations in the soil were 1333, 667, 333, 167, 84, and 0 mg/kg dry weight. Soil was comprised of 70% silica sand, 20% kaolinite clay, and 10% sphagnum peat. Rabbit faeces was added as food at 50 g/kg. Temperature was maintained at 13⁺/-2°C with 24 hour continuous lighting at 700-750 lux. Worms were assessed for mortality, general health, body weight, and behavior after 7 and 14 days.
 GLP: Yes No ?
 Test substance: LAS (commercial blend) with average alkyl chain length C_{11.6} (typical of LAS chain lengths found in the environment).
 Remarks: No statistically significant mortality was observed at the highest nominal concentration of 1333 mg/kg. Based on weight and burrowing behavior, the no effect concentration was the nominal 667 mg/kg dose, which was confirmed by HPLC to be 613 mg/kg. It should be noted that the continuous lighting prevented normal feeding, which normally occurs at night on the surface, and thus the test conditions and results should be considered highly conservative.
 Reference: Mieure, J.P., Waters, J., Holt, M., and Matthijs, E. 1990. Terrestrial safety assessment of LAS. Chemosphere 21:251-262.

Reliability: 2 Valid with restrictions

(e)

Type : Artificial soil []; Filter paper []; Other [X] **Natural soil**
Species: *Folsomia fimetaria* (Collembola; springtails)
Endpoint: Mortality [X]; Weight [X]; Other [X] **molting rate, reproduction**
Exposure period: 21 days
Results: Results are shown in the following table. All results are in mg/kg dry weight of soil.

Endpoint	NOEC	LOEC	LC ₁₀ or EC ₁₀	LC ₅₀ or EC ₅₀
Adult survival	>1000	>1000	>1000	>1000
Juvenile survival	500	700	196	570
Reproductive output	500	1000	147	737
Juvenile growth	<200	200	163	896
Molting frequency	<300	300	185	923

Method: No internationally accepted guideline was available. Adult and juvenile collembola were exposed to LAS mixed with 30 g of a natural, commonly available moist soil. Concentration levels for survival and reproduction of adults were control, 100, 150, 300, 500, 700 and 1000 mg/kg dry weight. Concentrations for survival and growth of juveniles were control, 200, 3000, 500, 700, and 1000 mg/kg dry weight. Concentrations for molting of juveniles were control, 300 and 600 mg/kg dry weight. In all cases, four replicates per concentration were used. For measurement of molting frequency, juveniles were held singly on a compressed surface of soil in multidishes with 24 circular holes. The multidishes were assessed every second day and exuviae were recorded and removed for a period of 20 days. Deviations from the subsequently developed ISO 11267 protocol included use of adults, use of 20 individuals instead of 10 per test chamber, and an exposure of 21 days instead of 28 days.

GLP: Yes [] No [X] ? []

Test substance: Marlon A350 (CAS #68411-30-3) C₁₀₋₁₃ LAS; 50% active substance; mean chain length of C_{11.53}; mean molecular weight 344

Remarks: The most sensitive endpoint was reproduction (EC₁₀ = 147 mg/kg dry weight). Nominal concentrations are derived from tables and figures since values were not listed directly in the text. While there were some deviations from the subsequently developed ISO 11267 protocol, the procedures are considered reliable.

Reference: Holmstrup, M. and Krogh, P.H. 1996. Effects of an anionic surfactant, linear alkylbenzene sulfonate, on survival, reproduction and growth of the soil-living collembolan, *Folsomia fimetaria*. Environ. Toxicol. Chem. 15:1745-1748.

Reliability: 2 Valid with restrictions. Well documented publication, no GLP, EC_x calculation not fully detailed.

(f)

Type : Artificial soil []; Filter paper []; Other [X] **Natural soil**
Species: *Folsomia candida* (Collembola; springtails) and *Enchytraeus albidus* (potworm)

Endpoint: Mortality [X]; Weight []; Other [X] **reproduction**
 Exposure period: 4 to 6 weeks
 Results: The resulting EC₅₀ values were very similar for the two species, as shown in the following table. All values are mg/kg dry weight.

Species	Endpoint	NOEC	LOEC	LC ₁₀ or EC ₁₀	LC ₅₀ or EC ₅₀
<i>F. candida</i>	Adult survival	1000	2500	750	1338
	Reproduction	500	1000	480	1437
<i>E. albidus</i>	Adult survival	<750	750	511	1400
	Reproduction	750	1500	447	1143

The EC₅₀ values for nitrification and CH₄ production were 431 and 277 mg/kg, respectively, for LAS. Aerobic respiration and denitrification were not inhibited at the test concentrations.

Method: LAS may enter the soil environment during sludge application. The toxic effects of LAS and nonylphenol (NP) to two soil invertebrates (*Folsomia candida* and *Enchytraeus albidus*) and five microbial processes (aerobic respiration, nitrification, denitrification, anaerobic CH₄ production, and anaerobic CO₂ production) were assessed in sludge-soil mixtures.

A coarse sandy soil collected from the upper 20 cm of an agricultural field in Jyndevad, Denmark was used for the laboratory experiments and mineralization controls. The Jyndevad soil consisted of 76.8% coarse sand, 12.2% fine sand, 4.1% silt, 3.9% clay, 3.0% organic matter and a pH of 6.0. A similar soil was collected in Lundgaard for use in the microbiological tests was 63.1% coarse sand, 26.6% fine sand, 3.8 silt, 4.3% clay, 2.2% organic matter and pH of 6.1. Dewatered activated sludge collected from a WWTP in Lundtofte was used in all experiments. LAS was applied to the sludge in a demineralized water solution and allowed to sorb for 24 hours at 4°C in at N₂ atmosphere before mixing the sludge with soil. The soil insects were exposed to a sludge:soil ratio of 1:20 on a dry weight basis. In the microbiological tests, 0.5 mL aliquot solutions of LAS in methanol were added to 1 g of sand, the methanol allowed to evaporate, and sorbed for 24 hours at 4°C under an N₂ atmosphere to minimize biodegradation during the sorption period. The sand was then mixed with sludge (0.3 g dry weight) and finally soil was mixed in with the sludge and sand. A sludge:soil ratio of 1:100 (dry weight basis) was used to avoid depletion of oxygen.

Folsomia candida

The springtail reproduction test was initiated with 10- to 12-day old juveniles and lasted 4 weeks. The resulting nominal LAS concentrations were 125, 250, 500, 1000, 2500, and 4000 mg/kg sludge-soil mixture. Ten juvenile *F. candida* were added to each of the 4 replicate vials per concentration containing 30 g (wet weight) of the sludge-soil mixture. The numbers of surviving adults and offspring were counted after 4 weeks.

Enchytraeus albidus

The enchytraeid worm reproduction test was initiated by introducing 10 worms with visible clitellum to each of 4 replicate vials per concentration. Nominal LAS concentrations were 750, 1500, 2250, and 3000 mg/kg

sludge-soil mixture (dry weight). Test duration was six weeks. After 3 weeks, the adult worms were removed and counted. At 6 weeks, the number of offspring hatched from cocoons were counted.

Microbiological tests

The toxicity of LAS to microbiological processes was evaluated using one aerobic system (simultaneous determination of aerobic respiration and nitrification) and two anaerobic systems (denitrification and methanogenesis). Nominal LAS concentrations were 0, 125, 250, 500, 1000, and 2500 mg/kg mixture (dry weight) for both the aerobic and methanogenic systems, and 0, 250, 500, 1500, 300, and 5000 mg/kg (dry weight) in the denitrifying system.

GLP: Yes No ?

Test Substance: The test substance for soil invertebrates consisted of an aqueous sodium salt solution containing 14% (w/w) of ¹⁴C-labeled C₁₀₋₁₃ LAS (EniChem Augusta Industriale; purity 95%). The average alkyl chain length of C₁₀₋₁₃ LAS was 11.6 and the distribution was C₁₀ 14%, C₁₁ 34%, C₁₂ 32% and C₁₃ 20%. A pure C₁₂ LAS, 4-(2-dodecyl)benzene sulfonate sodium salt, was used in the microbiological tests.

Remarks: Reproduction of *E. albidus* was the most sensitive endpoint (EC₁₀ = 447 mg/kg dry weight). Danish laws stipulate a maximum cut-off value of 1300 mg/kg for LAS in sludge for agricultural use.

Reference: Gejlsbjerg, B., Klinge, C., Samsoe-Petersen, L., and Madsen, T. 2001. Toxicity of linear alkylbenzene sulfonates and nonylphenol in sludge-amended soil. *Environmental Toxicology and Chemistry* 20:2709-2716.

Reliability: 2 Valid with restrictions. Well documented publication, comparable to ISO.

4.6.2 TOXICITY TO TERRESTRIAL PLANTS

(a)
 Species/Endpoints: See table
 Results: The following table shows the available NOEC, EC₁₀ and EC₅₀ values for twelve terrestrial plant species (in mg/kg dry weight).

Species	Endpoint	EC ₁₀	EC ₅₀
<i>Brassica rapa</i>	Growth		134
<i>Brassica rapa</i>	Growth	90	200
<i>Malvia pusila</i>	Growth		204
<i>Solanum nigrum</i>	Growth		169
<i>Chenopodium album</i>	Growth		164
<i>Amaranthus retroflexus</i>	Growth		142
<i>Nigella arvensis</i>	Growth		132
<i>Galinsoga parviflora</i>	Growth		90
<i>Sorghum bicolor</i>	Growth		137
<i>Helianthus annuus</i>	Growth		289
<i>Phaseolus aureus</i>	Growth		316
<i>Avena sativa</i>	Growth	50	300
<i>Sinapis alba</i>	Growth	200	300

Remarks: Values are extracted from a variety of original references and compiled for this article. NOEC values were extrapolated by applying an assessment factor of 10 to the EC₅₀. This is considered an unreliable assumption as measured acute-to-chronic ratios for LAS in plants vary between 2 and 6.

Reference: Jensen, J., Lokke, H., Holmstrup, M., Krogh, P.H. and Elsgaard, L. 2001. Effects and risk assessment of linear alkylbenzene sulfonates in agricultural soil. 5. Probabilistic risk assessment of linear alkylbenzene sulfonates in sludge-amended soils. *Environmental Toxicology and Chemistry* 20:1690-1697.

Reliability: 4 This study was given a reliability score of 4 because the original reports reviewed by the authors were not directly reviewed in the compilation of this robust summary.

(b)

Species: Grass, beans, radishes, potatoes

Radiolabel: Yes

Results: No adverse effects on plant biomass were observed at the concentrations tested [initial concentrations in soil = 27.2 mg/kg (grass, beans, radishes); = 16.2 mg/kg (potatoes)]

Temperature: Room temperature

Method: Soil cores taken from two ecosystems were collected and placed in a climate controlled "plant metabolism box". Ecosystem Section I consisted of a heavy, clay-like soil. Radiolabeled LAS (a defined mixture) absorbed to digested sludge was incorporated into the soils, after which the soils were planted with either grass, bush beans and radishes (Section I) or potatoes (Section II). The test systems were maintained under a defined standard climate (i.e., an average day in June in Northern Germany) for the vegetative period (76 and 106 days, respectively for Sections I and II). At the end of the growing season samples were collected from plants and soil and subjected to radioanalysis.

GLP: Yes [] No [] ? [X]

Test Substance: LAS. The authors state that they tested a defined mixture of LAS, but do not report the composition in this paper.

Reference: Figge, K. and Schoberl, P. 1989. LAS and the application of sewage sludge in agriculture. *Tenside Surf. Det.* 26:122-128.

Reliability: 2 Valid with restrictions

(c)

Species: radish, tomato, oats

Endpoint: Emergence []; Growth [X]; Other []

Exposure period: 14 day

Results: EC₅₀ >77.1 mg/kg soil dw
NOEC = 25.7 mg/kg soil dw

Method: OECD Guide-line 208 "Terrestrial Plants, Growth Test".

GLP: Yes [] No [] ? [X]

Test substance: Commercial LAS with an average carbon chain length of C_{11.8}.

Remarks: Information as cited in IUCLID Data Sheet for CAS #68411-30-3. The substance was tested in the range of 2.57 to 257 mg MBAS/kg Nominal concentrations, synthetic soil, static, pH 5.0-7.5, temperature 20-25 °C. Results are expressed as mg MBAS per kg soil. First Observed Effect Concentration (FOEC) is 77.1 mg MBAS/kg, EC50 is about 77.1 but below 257 mg MBAS/kg.

Reference: European Commission. 2000b. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished data (Registry No. 5929).

Reliability: 4 Not assignable

(d)

Species: *Brassica rapa* (Dicotyledon)
Endpoint: Emergence ; Growth ; Other emergence of seedlings
Exposure period: 21 day
Results: NOEC = 50 mg/kg soil dw
FOEC = 150 mg/kg soil dw

Method: EEC Directive 79/831, Annex V; EEC Ring Test C (L1) 3: Higher Plant, 1986.

GLP: Yes No ?

Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = 11.6

Remarks: Data refer to 100% active ingredient

Reference: European Commission. 2000b. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).

Reliability: 4 Not assignable

(e)

Species: *Lycopersicon esculentum* (tomato)
Endpoint: Emergence ; Growth ; Other emergence of seedlings
Exposure period: 21 day
Results: NOEC = 50 mg/kg soil dw
FOEC = 150 mg/kg soil dw

Method: EEC Directive 79/831, Annex V; EEC Ring Test C (L1) 3: Higher Plant, 1986.

GLP: Yes No ?

Test substance: Marlon A350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = 11.6

Remarks: Data refer to 100% active ingredient

Reference: European Commission. 2000b. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).

Reliability: 4 Not assignable

(f)

Species: *Avena sativa* (Monocotyledon)
Endpoint: Emergence ; Growth ; Other emergence of seedlings
Exposure period: 21 day
Results: NOEC = 50 mg/kg soil dw
FOEC = 150 mg/kg soil dw

Method: EEC Directive 79/831, Annex V; EEC Ring Test C (L1) 3: Higher Plant, 1986.

GLP: Yes No ?

Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = 11.6

Remarks: Data refer to 100% active ingredient

Reference: European Commission. 2000b. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing Henkel KGaA, unpublished results (Registry No. 5929).

Reliability: 4 Not assignable

(g)

Species: *Sorghum bicolor* (crop sorghum), *Helianthus annuus* (sunflower), *Phaseolus aureus* (mung bean)

Endpoint: Emergence []; Growth [X]; Other []

Exposure period: Emergence: 7 days; Growth: 21 days

Results: NOEC = 100 mg/kg soil dw (all three species)
EC₅₀ = 167, 289, and 316 mg/kg dw (for listed above listed species, respectively)

Method: A laboratory standard operating procedure based on OECD Guideline 208 (OECD 1984) was used. The test was conducted in an artificial soil consisting of commercially available potting compost (WC-B) and washed, sieved (1 mm mesh), dried (temperature and time not reported) silver sand (1:9 potting soil:washed sand). Concentrations tested were control, 1, 10, 100 and 1000 mg/kg dw. Deviations from the guideline included: % particles less than 20 µg not given; 4.1% instead of maximum 3% organic carbon in the test soil; sand was sieved with 1 mm mesh instead of 0.5 mm mesh; weight and variability of seeds not reported. The EC₅₀ values were calculated with probit analysis according to Finney (1971).

GLP: Yes [X] No [] ? []

Test substance: C₁₁ LAS (57.3% activity; average MW = 343)

Remarks: The NOEC is for the most sensitive endpoint, which was growth (shoot fresh weight). Nominal concentrations not measured.

Reference: Windeat, A.J. 1987. Effects on the growth of *Sorghum bicolor*, *Helianthus annuus*, *Phaseolus aureus*. Unilever study report BL/B/3078 (R118). Unilever Research Port Sunlight Laboratory, Sunlight, UK.

Reliability: 2 Valid with restrictions.

(h)

Species: *Araucaria heterophylla*

Endpoint: Foliar penetration of NaCl

Exposure period: 4 weeks

Methods: The influence of surfactants on foliar NaCl uptake was examined in Norfolk Island Pines (*Araucaria heterophylla*). Plants were exposed to seawater with different concentrations of LAS by spraying with a handheld sprayer three times a week for four weeks. Plants were sprayed until the foliage was wetted sufficiently for the spray to run off.

Results: At 10 mg/L of LAS, which corresponds with a reduced surface tension of 32 mN/m, the Na⁺ content in the foliage increased almost tenfold to a level of approximately 500 µmol/g dw and damage symptoms were recorded.

Remarks: The potential for LAS and other surfactants to influence defoliation in coastal trees was reviewed in a literature review sponsored by ERASM in 2002. In laboratory studies in which young trees are exposed to artificial sea spray, it has been demonstrated that the presence of surfactants at a concentration that causes a dynamic surface tension < 30 mN/m lead to an increased foliar penetration of NaCl via the stomata. It was found that a low surface tension increases the contact angle with the leaf and makes it

possible for an aqueous solution to enter the stomata. This is a hypothesized mechanism of defoliation.

- Reference: 1) Grieve, A.M. and Pitman, M.G. 1978. Salinity damage to Norfolk Island pines caused by surfactants. III. Evidence for stomatal penetration as the pathway of salt entry to leaves. *Aust. J. Plant. Physiol.* 5:397-413.
 2) Hamwijk, C. 2002. Literature study: Exposure and possible indirect effects of aerosol borne surfactants on coastal vegetation. TNO Chemistry report, Study number 02-4077/01. Prepared for CEFIC ERASM.
- Reliability: 4 Not assignable. Original studies were not directly reviewed.

(i)

- Species: *Pinus halepensis* (pine tree)
 Endpoint: Accumulation of LAS in plant tissues
 Exposure period: 2 minutes
 Method: The concentration of LAS tested was 1.7×10^{-4} mol/kg, which is equivalent to 58 mg/L based on a C_{11.6} LAS with a molecular weight of 342. Three batches of ten 2-year old pine trees were immersed for 2 minutes in distilled water alone (batch 1), in LAS-distilled water (batch 2), or LAS-synthetic sea water (batch 3). The objective was to simulate the exposure to severe storms on the seashore. Only the aerial parts of the plants were immersed in a large volume container (50 x 50 x 8 cm) containing 1 L of the respective solutions. The root system was isolated with a plastic bag enclosure and not exposed directly to the solutions. Controls were run on ten plants. Trees were removed from the solutions and gently shaken to eliminate liquid droplets. Radiolabeled and control trees were kept in a greenhouse for 48 hours under a 16:8 day:night photoperiod, temperatures of 22° C (day):16° C (night), and a constant relative humidity. Before analysis, the trees were washed twice in distilled water for 1 minute while shaking to simulate rainfall. Trees were cut back at the soil surface and the aerial part divided into several parts for analysis (epicuticular wax from needles, dewaxed needles, and remaining plant material consisting of branches without needles and tree stem). Respective samples were extracted, prepared, and the radioactivity measured in each fraction using a liquid scintillation cocktail and spectrometer. For scanning electron microscopy, 48 hours after the exposure period ten needles from each of the five replicates of treated and control plants were cut into small segments and air dried, fixed on aluminium stubs with conductive glue and carbon coated. Axial surfaces were examined with a Stereoscan 90B electron microscope with 15 kV acceleration voltage.
- GLP: Yes No ?
- Test substance: Stated by authors as LAS with an alkyl chain length of 12 carbons, with no further information. For our review, we have assumed this to be the typical European C_{11.6} LAS (average MW = 342). LAS was radiolabeled with ³⁵S in the sulphonate group attached to the phenyl ring and had a specific radioactivity of 8712 µCi/mol.
- Results: The amount of LAS (as a percent of total in solution; mean ± SD) found in each analyzed fraction of the trees and the wash water is shown below:

Fraction	Distilled water only (Batch 1)	LAS-distilled water (Batch 2)	LAS-seawater (Batch 3)
Epicuticular waxes	~ 0	0.18 ± 10	1.48 ± 1.03

Dewaxed needles	~ 0	~ 0	0.09 ± 0.16
Remaining plant material	~ 0	~ 0	~ 0
Distilled water wash	~ 0	0.17 ± 0.06	1.41 ± 0.57

The amount of uptake and percent of the total radioactivity incorporated in each fraction for the LAS-seawater and LAS-distilled water treatments are shown below:

	LAS-seawater treatment	LAS-distilled water treatment
³⁵ S LAS uptake µg/mg dw		
Epicuticular waxes	9.96 ± 3.10	4.90 ± 1.10
Dewaxed needles	0.007 ± 0.001	~ 0
Remaining plant material	0.006 ± 0.001	~ 0
% of total radioactivity		
Epicuticular waxes	89.70 ± 6.02	96.90 ± 2.34
Dewaxed needles	9.90 ± 5.95	~ 0
Remaining plant material	0.37 ± 0.23	3.10 ± 0.02

Remarks: After LAS exposure in seawater or distilled water, alterations of the epicuticular wax fine structure were observed by SEM. Forty-eight hours after exposure, half of the radioactivity detected was in the epicuticular waxes, with nearly all the rest in the washing solution. LAS was absorbed to a much greater extent in the seawater treatment (surface tension = 29 mN/m) than in the distilled water treatment (surface tension = 45 mN/m). LAS accumulated mainly in the epicuticular wax of the needles. Very little accumulated in the dewaxed needles or remaining plant material. More dramatic changes in epicuticular wax fine structure were observed following LAS treatment in seawater than in distilled water. These observations are in agreement with the studies reported by Hamwijk (2002) and Grieve and Pitman (1978), who demonstrated that low surface tension (<30 mN/m) could increase foliar penetration of salts from sea spray (dossier section 4.6.2h).

Reference: Richard, B., Grieve, P., Badot, P.M., and Garrec, J.P. 1996. Influence of marine salts on the localization and accumulation of surfactant in the needles of *Pinus halepensis* Mill. Ann. Sci. For. 53:921-930.

Reliability: 2 Valid with restrictions

4.6.3 TOXICITY TO OTHER NON MAMMALIAN TERRESTRIAL SPECIES (INCLUDING AVIAN)

Species: Chicken (Leghorn hens)
 Endpoint: Mortality [X]; Reproduction rate []; Weight []; Other [X] egg quality
 Exposure period: 45 day
 Results: NOEC = 200 mg/kg diet
 Method: Ten month old Leghorn chicken hens were given a dosage of 200 mg/kg in drinking water for 45 days. Four groups of six hens were used in the treatment group, with an additional six hens used as a control group.

GLP: Yes [] No [X] ? []
 Test substance: Commercial LAS

Remarks: No mortality or adverse effects on egg quality occurred at 200 mg/kg. While this is a non-standard study, it does indicate that up to 200 mg/kg in the drinking water does not adversely affect hens or egg laying.

Reference: Lopez-Zavalla, A., de Aluja, A.S., Elias, B.L., Manjarrez, L., Buchmann, A., Mercado, L., and Caltenco, S. 1975. The effects of ABS, LAS and AOS detergents on fish, domestic animals and plants. Prog. Water Technol. 7:73-82.

Reliability: 2 Valid with restrictions

4.7 BIOLOGICAL EFFECTS MONITORING (INCLUDING BIOMAGNIFICATION)

(a)

Type of test: static ; semi-static ; flow-through ; other outdoor experimental streams

Analytical monitoring: open-system ; closed-system Yes No ?

Method and Results: An integrated model stream ecosystem fate and effects study of a C₁₂LAS homologue, with a high content (35.7%) of its most hydrophobic and toxic 2-phenyl isomer, was performed in the summer and fall of 1996 in Procter and Gamble's Experimental Stream facility. The study addressed responses of periphytic microbes, immature benthic fauna including abundance and drift, and emergence of adult insects in a 56-day exposure. Exposures ranged from 126 to 2978 µg/L and were continuously presented in a single-pass, flow through test system. Microbial heterotrophs acclimated to C₁₂LAS exposure quickly (14 days) and biodegraded C₁₂LAS at all concentrations. Blue-green algae responded by increasing in abundance with increasing C₁₂LAS concentration. Invertebrates responded by increased drift and reduced benthic abundances at concentrations exceeding 293 µg/L. Emergence at 927 µg/L also declined relative to the control. Adverse responses for mayflies and chironomids were indicated using univariant statistical techniques. Multivariant techniques indicated these taxa plus molluscs, aquatic worms, caddisflies, and stoneflies were impaired at some concentrations. Bioavailability of C₁₂LAS was investigated in streams as a function of the total suspended solids load in the water column driven by local weather and watershed patterns. A continuous bioavailability model indicated exposure was reduced by an average of 8.5 ± 8.9%. A model ecosystem NOEC (no-observed-effect-concentration) was concluded to be 293 µg/L based on measured water column exposure and adjusted to 268 µg/L by the bioavailability model. A summary of selected population and community responses at 8 weeks from the current study is shown in the following table:

Community/Measure	Dose Response	Temporal	NOEC (µg/L)
Heterotrophic microbial			
Biomass (total lipid phosphate/mm ²)	NS		
Amino acid uptake (³ H dpm/mm ² /min)	NS		
Phospholipid fatty acid (PLFA) distr.		Shift at >293 µg/L	
Surfactant mineralization (% CO ₂)		Acclimation at all conc.	
Autotrophic microbial			

Bicarbonate uptake (^{14}C dpm/mm ² /min)	+	+	
Algal density (cells/mm ²)	NS		
Algal biovolume ($\mu\text{m}^3/\text{mm}^2$)	NS		
Blue-green algal density (cells/mm ²)	++	++	
Green algal density (cells/mm ²)	NS		
Diatom algal density (cells/mm ²)	NS		
Algal richness	-	-	927
Dominant taxa (cells/mm ²)			
<i>Cocconeis placentula</i>	-	-	927
<i>Melosira varians</i>	NS		
<i>Chroococcus</i> sp.	+	+	
<i>Nitzschia dissipata</i>	NS		
<i>Navicula salinarum</i> v. <i>intermedia</i>	NS		
<i>Pleurosira</i> (= <i>Biddulphia</i>) <i>laevis</i>	NS		
<i>Nitzschia inconspicua</i>	++	++	
<i>Nitzschia palea</i>	+	+	
<i>Diatoma vulgare</i>	--	--	927
<i>Gyrosigma acuminatum</i>	-	-	927
Invertebrates			
Richness	NS		
Diversity (Shannon-Weaver)	NS		
Total abundance (No./m ²)	--	--	293
Insect abundance (No./m ²)	NS		
EPT abundance (No./m ²)	-	-	927
Mayfly abundance (No./m ²)	NS		
Caddisfly abundance (No./m ²)	NS		
True fly abundance (No./m ²)	NS		
Chironomid abundance (No./m ²)	NS		
Mollusk abundance (No./m ²)	NS		
Oligochaete abundance (No./m ²)	--	--	293
Dominant populations (No./m ²)			
<i>Baetis</i> sp. (mayfly)	b		
<i>Isonychia</i> sp. (mayfly)	---	---	927
<i>Stenonema</i> sp. (mayfly)	NS		
<i>Thienemannimyia</i> sp. (chironomid)	---	---	293
<i>Tanytarsus</i> sp. (chironomid)	++	++	
<i>Cricotopus</i> sp. (chironomid)	+	+	
<i>Polypedilum</i> sp. (chironomid)	+	+	
<i>Reotanytarsus</i> sp. (chironomid)	NS		
<i>Naididae</i> (aquatic worm)	NS		

^a Plus (+) and minus (-) signs indicate whether the response significantly increased or decreased from the control condition ($\alpha = 0.05$). The strength to the response was graded as slight (+/-), moderate (++/-), or great (+++/-) based on statistical analyses. NS indicates not significant. ^b Taxon too low in abundance, emerged.

A literature review of 13 available model ecosystem studies was conducted and NOEC conclusions were adjusted by a structure-activity-relationship to dodecyl chain length (sulfophenyl position and distribution being ignored due to lack of information in the reviewed studies). Lentic studies (n = 7) were found to have higher NOECs than lotic studies (n = 6) and were more variable. Mean NOEC \pm standard deviations for all studies, lentic studies only, and lotic studies only were 3320 \pm 6040, 5720 \pm 7640, and 530 \pm 430 $\mu\text{g/L}$, respectively. Interpretation of results for anomalies from specific studies suggest the

importance of experimental design, use of laboratory versus natural surface water, biological complexity of the test system, and physical test system design as relevant factors for consideration.

GLP: Yes No ?

Test substance: Dodecyl linear alkylbenzene sulfonate (C₁₂LAS) (CAS# 25155-30-0)

Remarks: The mesocosm studies indicate that the lower limits of field studies can be considered between 0.12 to 0.5 mg/L. It should be noted that the lowest NOEC value (0.12 mg/L) was observed in an artificial stream study (Tattersfield et al. 1995, 1996) in which river water was seeded from field collections and a hydrocyclone used to prevent colonization of biota throughout the study. Drift therefore comprised only emigration and not immigration. Thus, the Tattersfield et al. study is an ecologically restrictive study design that ignores the importance of recovery vectors present in natural systems. The current study (Belanger et al. 2002) did not have these design limitations. The current critical review of all field studies, including the Tattersfield et al. study, concluded that a NOEC = 0.27 mg/L for a C₁₂LAS homologue (0.37 mg/L if normalised to C_{11.6} LAS) is the most reliable, robust and defensible value for the aquatic freshwater ecosystem. This is a key study for aquatic toxicity in model ecosystems (see SIAR Table 15).

Reference: Belanger, S.E., Bowling, J.W., Lee, D.M., LeBlanc, E.M., Kerr, K.M., McAvoy, D.C., Christman, S.C., and Davidson, D.H. 2002. Integration of aquatic fate and ecological responses to linear alkyl benzene sulfonate (LAS) in model stream ecosystems. *Ecotoxicology and Environmental Safety*. 52:150-171.

Reliability: 1 Valid without restriction

(b)

Results: Time averaged mean measured concentrations over the 28 day exposure period were 0.03, 0.06, 0.12, 0.32, 0.52, 1.0 and 3.0 mg/L in the artificial streams and 0.24, 0.81, and 2.0 mg/L in the downstream pools. Problems were experienced with dosing LAS into the streams after day 45 due to extreme weather conditions causing freezing of stock solutions of LAS in the delivery tubes. Results should therefore be treated with caution as exposure data is extrapolated from 45 to 56 days. Time averaged mean measured concentrations over the first 45 days of the 56 day study period were 0.03, 0.06, 0.12, 0.32, 0.52, 1.0 and 3.0 mg/L in the artificial streams and 0.22, 0.69, and 1.6 mg/L in the downstream pools. A total of 65 taxa were identified in the artificial streams and downstream pools. Effects data were generated for 24 endpoints, which included ten invertebrate taxa, two fish species and algae. The inclusion of downstream pool sections increased the range of taxa investigated. The downstream pool community appeared generally less sensitive to the LAS than the stream channel community. Individual taxa were found to differ in susceptibility to LAS depending on their location in the stream channels. The same taxa were generally more susceptible when in the riffle section than in the pool sections. This may have been the result of differences in exposure or physiological state of the organism. Results from the first 28 days of the study concluded that there were no NOECs below 0.12 mg/L. Extending the study to 56 days resulted in no change in NOECs for the majority of endpoints. NOECs determined in the artificial streams were in the range of 0.03 to >3.0 mg/L although the

most reliable NOECs were in the range 0.12 to 3.0 mg/L. In the downstream pools the NOECs ranged from 0.69 to >1.6 mg/L. Only two NOECs were below 0.12 mg/L; the population density of *Gammarus pulex* in the riffle and population density of *Baetis sp.* Some uncertainty is associated with the extended 56 day study due to three main factors. First, lack of exposure data between days 45 and 56. Further uncertainty is associated with the low NOECs for some end points, particularly for *G. pulex* in the riffle section of the artificial streams. Second, the uncertainty in the lower NOEC is due to the times dependent effects (increased susceptibility at 56 compared to 28 days) only being observed for individuals of *G. pulex* in the riffle but not the pool section of the artificial streams. Reduction in NOEC from 28 to 56 days for *G. pulex* in the riffle was not evident in the pool section of the artificial streams where the 28 and 56 day NOEC was 0.52 mg/L. The exact cause of the difference between riffle and pool is unknown. Third, uncertainty was due to variability of the *G. pulex* data and sensitivity to statistical transformation.

Conclusion:

The lowest NOEC value observed in this artificial stream study was 0.12 mg/L. However, the river water was seeded from field collections and a hydrocyclone used to prevent colonization of biota throughout the study. Drift therefore comprised only emigration and not immigration. This is an ecologically restrictive study design that ignores the importance of recovery vectors present in natural systems.

Method:

Communities of freshwater organisms were established in eight artificial streams and four downstream pools over a 10 day period, five weeks prior to the onset of dosing. Seven nominal LAS exposure concentrations (0.02, 0.05, 0.1, 0.3, 0.5, 1.0 and 3.0 mg/L) and an untreated control were randomly allocated to the eight artificial streams to yield a regression model experiment design. Four of the streams (Control, 0.3, 1.0 and 3.0 mg/L) were connected to downstream pools. Effects measurements were taken after 28 and 56 days.

The streams were operated as once-through systems with a residence time of three minutes. Each individual stream was divided approximately equally into a slow flowing pool section (0.20 m water depth, ~3 cm/s flow velocity) and a faster flowing riffle section (0-0.02 m water depth, ~30 cm/s flow velocity). The total volume of water in each stream system was 240 litres. The downstream pools consisted of plastic cylindrical tanks (1.04 m diameter and 510L capacity) with a residence time of approximately 2 days.

The water used in the study was from a local chalk stream with a hardness of 194 to 392 and 280 to 378 mg/L CaCO₃ in the streams and downstream pools, respectively. The water temperature was relatively low ranging from 3.0 to 13.2°C and 3.2 to 13.1°C in the streams and downstream pools, respectively. Concentrations of suspended solids and total organic carbon (TOC) in the artificial streams were low (suspended solids 1.8 to 3.0 and 0.4 to 6.6 mg/L and TOC 1.5 to 5.2 and 1.9 to 7.9 mg/L in the streams and downstream pools, respectively). The low concentrations of suspended solids and TOC would have tended to maximise the availability of LAS in the system. Also Goyer et al. showed that toxicity of surfactants to daphnia correlated with water hardness. Given the above conditions, effects data generated in this system should be judged to be at the most sensitive end of the distribution.

Remarks: Effects measurements were taken after 28 and 56 days. The streams were operated as once-through systems with a residence time of three minutes. Each individual stream was divided approximately equally into a slow flowing pool section and a fast flowing riffle section. There is uncertainty connected to the extended 56 day study having to do with the lack of exposure data between days 45 and 56. Uncertainty is also linked with the low NOEC values for some of the end points. This is related to time dependant effects (increased susceptibility at 56 compared to 28 days) being observed only for individuals of *G. pulex* in the riffle but not the pool section of the artificial streams and variability of the *G. pulex* data and sensitivity to statistical transformation. The NOEC values of ≥ 0.12 mg/L determined after 28 days of exposure are considered estimates of no effect for LAS. For the previously mentioned reasons, the indications of NOECs below 0.12 mg/L from the extended 56 day study appear to be outliers and are not considered reliable for assessment purposes (i.e., would be Klimish 3). Also, it should be noted that a hydrocyclone was used to prevent colonization of biota throughout the study. Thus, drift comprised only emigration and not immigration. Therefore, the study design is ecologically restrictive in that it ignores the importance of recovery vectors present in natural systems. In a review of 13 model ecosystem studies, including this one, Belanger et al. (2002) concluded that a NOEC of 0.27 mg/L for a C₁₂LAS homologue is the most reliable, robust and defendable value for aquatic freshwater ecosystems.

Test Substance: C₁₀₋₁₃ LAS, 38.3% active matter, average carbon chain length = 11.52

Reference: 1) Tattersfield, L.J., Holt, M., Girling, A.G., Mitchell, G.C., Pearson, N., and Ham, L. 1995. The fate and effects of linear alkylbenzene sulfonate (LAS) in outdoor artificial streams and pools. External report. Shell Research Limited, Sittingbourne Research Centre. Document No. SBER.95.009.
 2) Tattersfield, L.J., Mitchell, C.G., Holt, M., Girling, A.G., Pearson, N., and Ham, L. 1996. Linear alkylbenzene (LAS): Fate and effects in outdoor artificial streams and pools – An extended study. Internal report. Shell Research and Technology Centre, Thornton. Document No. TNER.96.005.

Reliability: 4 Not assignable due to restrictive test design and inconsistencies in the data

(c)
 Type: Aquatic []; Field []; Soil [X]; Other []
 Results: LAS had no effect on heterotrophic respiration in the sludge compartment but stimulated activity and metabolic quotient (microbial activity per unit of biomass) in the surrounding soil. Basal respiration (BR) was significantly stimulated up to 60 days in the 0-10 mm compartment, but only after 60 and 82 days in the 10-30 mm compartment. No significant stimulation in BR was observed at 30-60 mm. Substrate-induced respiration stimulation was variable and restricted only to soil in the 0-10 mm compartment. Autotrophic ammonia oxidation was initially inhibited in the LAS-spiked sludge, which led to dramatic but transient increases of NH₄⁺ availability in the sludge and surrounding soil, subsequently stimulating soil ammonia oxidizers. As judged from a bioluminescence toxicity assay, however, LAS or other sludge components never

accumulated to toxic levels in the soil and the LAS tolerance of the indigenous microbes further remained unchanged following LAS exposure. Bioluminescence was slightly, but not significantly, reduced in the 0-10 mm compartment at the first sampling, but not thereafter and not in the 10-30 or 30-60 mm compartments. LAS effects on the microbial populations largely occurred during the first two months and were confined to soil closer than 30 mm from LAS-spiked sludge.

Test Substance: C₁₀₋₁₃ LAS, sodium salt; average alkyl chain length C_{11.6}

Method:

Well-defined bands of sewage sludge spiked with 0 (control), 7.1, or 31.3 g LAS/kg dry weight were applied to loamy sand soil in an agricultural field in Lundgaard, Denmark using a random block design. To each block, three sludge bands (one per LAS treatment) were carefully applied such that the bands were eventually applied at a specific soil depth of approximately 6 to 10 cm and covered by soil. All treatments were replicated five times. A few days after sludge application, the entire experimental site was sown with oats in order to make experimental conditions as realistic as possible. Sampling for microbial parameters was done on a weekly to monthly basis for the first 100 days, with the last samples being taken approximately one year after the start of the experiment. A rectangular corer providing a 40 mm wide cross section of the sludge bands and the surrounding soil was sectioned into four compartments representing various distances (0-10, 10-30, 30-60 mm) from the sludge. At each sampling date, two replicate cores from each sludge band were sampled and the corresponding samples from the two cores were pooled. Microbial parameters measured included basal respiration (BR), substrate-induced respiration (SIR), potential ammonia oxidation (PAO), and pollution-induced community tolerance. Bioluminescence toxicity tests were also conducted and correlated with ammonia oxidation activity as a measure of the physiological state of the cells. Two-way analysis of variance statistics were used for each sampling date, followed by Dunnett's test. Data were transformed if necessary.

Remarks:

Measured LAS concentrations were 0.069 (control), 7.1 and 31.3 g/kg dw sludge. Results strongly suggest that disposed of LAS-contaminated sludge will not produce a significant adverse effect on the function of the soil microbial community under field conditions. Measured effects generally lasted two months or less and were confined to soil closer than 30 mm from the LAS-spiked sludge. No signs of long-term selection due to toxicity were noted. According to the authors, the study should be considered a worst-case due to the application of high LAS concentrations only occasionally encountered in sewage sludge, the use of LAS-spiked sludge possible overestimating the actual bioavailability relative to aged surfactants in natural sludge, the application of relatively large (4 x 4 cm) two dimensional sludge bands possible retarding oxygen intrusion and consequently LAS degradation in the sludge relative to smaller spherical sludge clumps present under more realistic field conditions, and the use of a coarse, sandy soil with relatively low organic matter content. While NOEC, LOEC, EC50, etc. were not calculated, significant differences from the control sludge were detected in the high (31.3 g/kg dw sludge) and in the low (7.1 g/kg dw sludge) treatments. Therefore, the NOEC should be <7.1 g/kg dw sludge.

Reference:

Brandt, K.K., Krogh, P.H., and Sorensen, J. 2003. Activity and population dynamics of heterotrophic and ammonia-oxidizing microorganisms in soil

surrounding sludge bands spiked with linear alkylbenzene sulfonate: a field study. *Environ. Toxicol. Chem.* 22:821-829.

Reliability: 2 Valid with restriction. Well documented publication, no GLP, concentrations only measured at start of experiment

(d)

Type: Aquatic []; Field []; Soil [X]; Other []

Results: No short-term or long-term (4 years) adverse effects on 9 different microbial functions/processes or the abundance or diversity of microarthropods and earthworms were observed after sludge application of up to 21 t dw/ha dry weight, corresponding to a LAS dose of approximately 35 kg/ha, or approximately 15 mg/kg dry weight.

Method: This study was conducted by the Danish EPA to assess the effects of using sewage sludge applications on soil fauna and microbial processes in winter-wheat and barley undersown with clover grass. Three levels of sludge and cow dung (3.5, 7, and 21 t dw/ha) were tested along with control fields. Concentrations of LAS in sludges from the two waste water treatment plants were 1,100 and 1,700 mg/kg.

Test Substance: LAS (unspecified)

Reference: 1) Jensen, J., Lokke, H., Holmstrup, M., Krogh, P.H. and Elsgaard, L. 2001. Effects and risk assessment of linear alkylbenzene sulfonates in agriculture soil. 5. Probabilistic risk assessment of linear alkylbenzene sulfonates in sludge-amended soils. *Environ. Toxicol. Chem.* 20:1690-1697.

2) Jensen, J. and Krogh, P.H. 1999. Ecological assessment of sewage sludge application. *Proceedings, Nordiska Jordbruks forskares Forening, Seminar 292. Jokionen, Finland, November 23-25, 1998, pp. 98-100.*

3) Krogh, P.H., Holmstrup, M., Jensen, J., and Peterssen, S.O. 1997. Ecotoxicological assessment of sewage sludge in agricultural soil. Working Report No. 69. Ministry of Environment and Energy, Danish Environmental Protection Agency.

Reliability: 2 Valid with restrictions

(e)

Type: Aquatic []; Field []; Soil [X]; Other []

Species: cellulolytic bacteria, fungi and actinomycetes and microbial parameters

Exposure Period: up to 8 weeks

Results: EC₅₀ = 17 to 128 mg/kg dry weight for all parameters other than as indicated.

EC₁₀ = < 8 to 22 mg/kg dry weight for all parameters other than as indicated.

Except for β-glucosidase activity, basal respiration, and total PLFA content, all soil parameters were sensitive to LAS, with EC₁₀ values in the range of less than 8 to 22 mg/kg dry weight. The authors indicate that this probably reflected a similar mode of LAS toxicity, ascribed to cell membrane interactions, and showed than sensitivity to LAS was common for various soil microorganisms. The extracellular β-glucosidase activity was rather insensitive to LAS (EC₁₀, 47 mg/kg dry weight), whereas the basal soil respiration was not inhibited even at 793 mg/kg dry weight. This was interpreted as a combined response of inhibited and stimulated compartments of the microbial community. The PLFA content showed no decrease even at 488 mg/kg.

Analytical monitoring:	Yes <input checked="" type="checkbox"/> No <input type="checkbox"/> ? <input type="checkbox"/>
Method:	The short-term effects of aqueous LAS on microbial parameters was tested in a sandy agricultural soil that was incubated for up to 11 days. The assays included 10 microbial soil parameters: ethylene degradation; potential ammonium degradation; potential dehydrogenase activity; β -glucosidase activity; iron reduction; populations of cellulolytic bacteria, fungi and actinomycetes; the basal soil respiration; and the phospholipid fatty acid (PLFA) content. Soil from the plough layer was sampled at an agricultural field at Lundgaard, Denmark. The soil consisted of coarse sand (67%), fine sand (16%), silt (8.6%), clay (6.2%), humus (2.7%) and had a total carbon content of 1.5%. The soil had not been treated with sewage sludge and had not been sprayed with pesticides in the last two years. For the experiments with aqueous LAS, triplicate soil incubations were amended with the appropriate LAS solutions to produce the LAS contents. The soils were carefully mixed and incubated in the dark and duplicate soil samples for LAS analyses were frozen at the beginning of the incubation period. EC ₁₀ and EC ₅₀ values were calculated by a linear-interpolation analysis (IC _p), which was based on bootstrapping. The NOECs and LOECs were determined by Dunnett's test using a SAS analysis-of-variance procedure. Nominal concentrations were control, 8, 22, 62, 174 and 488 mg/kg dw soil, except for BR (control, 0.8, 8, 79 and 793 mg/kg dw soil). On average, 84 to 95% of the nominal concentrations were initially recovered by the chemical analysis. Nominal levels were used for the calculation of effect concentrations.
GLP:	Yes <input type="checkbox"/> No <input type="checkbox"/> ? <input checked="" type="checkbox"/>
Test substance:	C ₁₀₋₁₃ LAS obtained as an aqueous sodium salt solution with a LAS content of 16.1% (w/w), Na-LAS average molecular weight = 342 g/mol, distribution: C ₁₀ 14%, C ₁₁ 34%, C ₁₂ 31%, and C ₁₃ 21%
Remarks:	The study demonstrated that LAS inhibited specific compartments of the soil microbial community. The lowest EC ₁₀ values for microbial soil parameters were slightly higher than the predicted no-effect concentrations recently derived for plants and soil fauna (~5 mg/kg dry weight). A subsequent study (Elsgaard et al. 2001, Part 2) further indicated that the short-term effects observed for aqueous LAS on soil microbiology were modified by the dosage of LAS with sewage sludge and by a prolonged incubation time. The data suggest that a terrestrial risk assessment based on short-term effects of aqueous LAS fully encompasses the risk that may occur when LAS is applied to agricultural soil by means of sewage sludge.
Reference:	1) Elsgaard, L., Petersen, S.O., and Deboz, K. 2001a. Effects and risk assessment of linear alkylbenzene sulfonates in agricultural soil. 1. Short-term effects on soil microbiology. <i>Environmental Toxicology and Chemistry</i> 20:1656-1663. 2) Elsgaard, L., Petersen, S.O., and Deboz, K. 2001b. Effects and risk assessment of linear alkylbenzene sulfonates in agricultural soil. 2. Effects on soil microbiology as influenced by sewage sludge and incubation time. <i>Environmental Toxicology and Chemistry</i> 20:1664-1672.
Reliability:	2 Valid with restrictions. Well documented publication, no GLP, EC _x calculation not fully detailed.
(f) Type:	Outdoor experimental stream

Methods: Species or ecosystem studied. Taxonomic groups tested were periphyton, detritus, invertebrates, snails, amphipods, and fish. One concentration was tested in triplicate. The substance was dosed eight times at seven day intervals. Test duration 45 days.

Results: Effects monitored: Population and community effects.
The best estimated NOEC is >0.36 mg/L.
For fish (caged larval fathead minnows) no effects on mortality occurred at the only concentration tested. However growth was significantly decreased in the dosed systems. The authors state that this is caused by the better food and light conditions in the controls.
Growth of periphyton, the degradation rate of detritus, population and community growth of invertebrates and the population density of snails were not inhibited at the only concentration tested.
For Amphipods no NOEC could be determined. Mortality was highest (45%) at a control location.
Chemical analysis: Concentrations were measured.

Test Substance: LAS; average chain length C_{11,9} (representative of homologues found in typical sewage effluents in the U.S.).

Reference: Fairchild, J.F., Dwyer, F.J., La Point, T.W., Burch, S.A., and Ingersoll, C.G. 1993. Evaluation of a laboratory-generated NOEC for linear alkylbenzene sulfonate in outdoor experimental streams. *Environ. Toxicol. Chem.* 12:1763-1775.

Reliability: 2 Valid with restrictions

(g)

Type: Outdoor ponds.

Methods: Species or ecosystem studied: Outdoor ponds were used with three compartments (700 L each) with sediment. Taxonomic groups tested were phytoplankton, plants, cyclopedia, and cladocerans. Two concentrations were tested with no duplicates. Test duration 56 days and one year.
Effects monitored: Population and community effects.
Chemical analysis: The substance was dosed according to need.

Results: For phytoplankton slight inhibition of photosynthesis and chlorophyll occurred at 5.0 mg/L. The best estimated NOEC was determined at slightly below 5 mg/L.
For plants reduction of species number and composition and for cladocerans inhibition of development occurred at the highest concentration tested. The best estimated NOEC for these groups was 5.0 mg/L. The best estimated LOEC was 10 mg/L.
For cyclopedia egg production was reduced at 5 mg/L. The best estimated NOEC was determined at 3.5 mg/L. The best estimated LOEC was 5 mg/L.
For midge no NOEC could be determined. Survival was strongly reduced due to low oxygen concentrations and a high level of suspended solids.

Substance: LAS; chain length is probably C₁₂.

Remarks: A concentration-effect relationship was found. Concentrations were measured. Data as reported by BKH, Huls, and Henkel in IUCLID dataset for CAS #90194-45-9 dated 19 February 2000.

Reference: Huber, W., Zieris, F.J., Feind, D., and Neugebauer, K. 1987. Ecotoxicological evaluation of environmental chemicals by means of aquatic model ecosystems (Translation). Bundesministerium fuer Forschung und Technologie, Research Report (03-7314-0).

Reliability: 4 Not assignable. Original report not available for review.

(h)

Type: Laboratory aquaria

Methods: The effect of LAS on the structure and function of microbial communities was studied in a flow-through model ecosystem containing several trophic levels. The LAS was applied to 19-L glass aquaria in either well water (Phase I) or sewage effluent (Phase II). In Phase I, duplicate chambers contained water and 2.5 cm lake sediment (Winton Lake, Cincinnati, OH) and several trophic levels (bacteria, algae, macrophytes [*Elodea canadensis*, *Lemna minor*], macroinvertebrates [*Daphnia magna*, *Paratanytarsus parthenogenica*], and fish [*Lepomis macrochirus*]). Chambers were allowed to equilibrate for about four weeks and then were exposed to 0.5 and 5.0 mg/L LAS. Flow rate in the proportional diluter delivered 6 to 10 replacement volumes per day. In Phase II, the aquaria were supplied with LAS in sewage effluent to simulate more closely the situation in an actual receiving stream. Sewage effluent was generated in a continuous activated sludge (CAS) unit and was adjusted to maintain 50 percent LAS degradation. Effluent from the CAS unit was then supplied to the test chambers at sewage dilutions of 3 and 30 percent to achieve nominal undegraded LAS concentrations of 0.5 and 5.0 mg/L, respectively. Test duration was 28 days. Microbial structure was estimated by measurements of total viable bacterial biomass as CFU/mL. Microbial function was estimated in Phase I by measuring the rates of oxygen consumption during the degradation of glucose and LAS. In Phase II, microbial function was assayed by radiochemical methods.

Results: In Phase I, the structure of microbial communities was not affected, and no significant differences were reported in mean biomass or number of colony-forming units between the microorganisms exposed at the two levels. The mean total biomass calculated for all tanks and across all sampling points was about 3×10^5 CFU/mL. The function of the microbial communities was reduced only at 5.0 mg/L. In Phase II, no effect was seen on the structure of the microbial community, with mean CFU/mL in the low and high dose aquaria (0.9×10^5 and 1.4×10^5 , respectively) similar to the control aquaria (1.4×10^5). Also, no effects were observed microbial function in Phase II, which was measured only as the degradation of LAS. Therefore, the NOEC based on the most sensitive endpoint (microbial community function) is 0.5 mg/L.

Substance: LAS; radiolabeled C^{14} -LAS with chain length C_{12} (91% purity) plus unlabeled LAS with average chain length $C_{11.6}$ (C_{10} 9.7%, C_{11} 27.9%, C_{12} 54.4%, C_{13} 8.0%; 95% purity)

Remarks: Function assays in Phase II were based on LAS degradation only, since the Phase I results indicated that LAS degradation was the most sensitive indicator of toxic effect levels.

Reference: Larson, R.J. and Maki, A.W. 1982. Effect of linear alkylbenzene sulfonate on the structure and function of microbial communities in model ecosystems. Aquatic Toxicology and Hazard Assessment: Fifth Conference, ASTM STP 766, Pearson, J.G., Foster, R.B., and Bishop, W.E., Eds., American Society for Testing and Materials, pp. 120-136.

Reliability: 2 Valid with restrictions

(i)

Type: Laboratory aquaria

Methods: Two exposures were conducted. Phase I was designed to develop basic toxicological information. Phase II introduced partially degraded LAS contained within the effluent of a continuous activated sludge unit and was designed to simulate real-world fate and effects for LAS. In Phase I, duplicate 19-L glass aquaria containing model ecosystems at four nominal concentrations (0.5, 1.0, 2.0, 4.0 mg/L) contained water and 2.5 cm lake sediment (Winton Lake, Cincinnati, OH) and several trophic levels (bacteria, algae, macrophytes [*Elodea canadensis*, *Lemna minor*], macroinvertebrates [*Daphnia magna*, *Paratanytarsus parthenogenica*], and fish [*Lepomis macrochirus*]). Flow rate in the proportional diluter delivered approximately 8 replacement volumes per day. Additionally, at each cycle of the diluter, 1.5 mL of a *Daphnia* food suspension diluted with a culture of *Selenastrum* was added to each chamber. Following an initial 3 day acclimation period to analytically confirm test concentrations, 4 glass periphyton slides (5 x 5 cm), 8 vegetative shoots of *Elodea*, 10 early instar *Daphnia*, 25 midge eggs and 5 pre-weighed juvenile bluegills (2.5-5.0 cm length) were added to each aquarium. Fish were screened from access to the macroinvertebrates by a 60 mesh stainless steel screen and were fed a daily supplement of frozen brine shrimp. In Phase II, the aquaria were supplied with LAS in sewage effluent to simulate more closely the situation in an actual receiving stream. Sewage effluent was generated in a continuous activated sludge (CAS) unit and was adjusted to maintain 50 percent LAS degradation. Effluent from the CAS unit was then supplied to the test chambers at continuous dilutions of 3.75, 7.5, 15, and 30 percent sewage concentrations to simulate sewage dilutions existing in natural receiving waters. Test duration was 28 days. Effects monitored included population and community effects. Nominal concentrations were confirmed with MBAS analysis.

Results: Dissolved oxygen concentrations ranged between 7.0 and 9.0 mg/L during Phase I. Dissolved oxygen concentrations in Phase II ranged between 3.1 and 7.3 mg/L, with the lowest readings consistently observed in the aquaria receiving the 30% sewage concentrations, as would be expected. Temperature was maintained at 2°C, mean pH was 8.1 ± 0.2 in Phase I and 7.5 ± 0.3 in Phase II. MBAS analysis confirmed the nominal concentrations.

No significant effects on microbial community structure occurred in Phase I, with biomass levels in the high dose (4.0 mg/L) comparable to or greater than the biomass levels in the controls. Similarly, no significant effects on microbial community structure were observed in Phase II. The function of microbial communities in Phase I was affected at the high dose (4.0 mg/L), as evidenced by significant depression in the rates of both glucose and LAS degradation. No effects on microbial function were observed in Phase II. No dose response correlation in overall productivity was evident for the periphyton (aufwuchs) community in Phase I. In Phase II, the introduction of the sewage effluent produced a generally higher turbidity level and the higher organic concentrations were conducive to the growth of thick sheets of bacterial and fungal communities. Very little direct periphytic plant growth was observed. The stimulatory effect of the increasingly higher sewage concentration is evident in the progressively higher aufwuchs production observed between 3.75 and 30% effluent. No effects on *Elodea* production were observed in Phase I. However, in

Phase II, *Elodea* and *Lemna* plant growth was inhibited at all concentrations except 3.75% by the increased bacterial and fungal periphyton growth as periphytic sheaths tended to cover the leaves and vegetative tips of the macrophytes. Evaluation of the *Daphnia magna* data from Phase I is confounded by unexpected poor control survival, although productivity appeared to be lower in the 1.0, 2.0 and 4.0 mg/L concentrations than at 0.5 mg/L. In Phase II, all *Daphnia* died in the 30% sewage concentration but production reached much greater numbers in the other concentrations than they did in Phase I. The midge species had an apparent reduction in numbers at 4.0 mg/L compared to the controls in Phase I. In Phase II, erratic growth in the controls and all exposures led to no meaningful midge survival at the end of Phase II. Bluegill fish growth at the end of Phase I was reduced at the 2.0 and 4.0 mg/L concentrations but not at 0.5 or 1.0 mg/L. In Phase II, fish in all wastewater dilution concentrations from 3.75 to 30% grew less than the controls. The following table summarizes the NOEC values (mg/L) for Phase I and Phase II, as determined from the data described above.

Species	Phase I	Phase II
Bacterial Structure	4.0	30%
Bacterial Function	1.41 ^a	30%
Periphyton/Algae	4.0	3.75%
<i>Elodea</i>	4.0	3.75%
Duckweed (<i>Lemna minor</i>)	Not Reported	3.75%
<i>Daphnia magna</i>	-- ^b	-- ^c
Midge (<i>Paratanytarsus parthenogenica</i>)	2.0	-- ^c
Bluegill (<i>Lepomis macrochirus</i>)	1.0	<3.75%

^a Data available only for the low and high concentrations (0.5-4.0 mg/L). Value is geometric mean.

^b Poor control survival precludes calculation of a NOEC.

^c Data not interpretable

Therefore, the NOEC based on the most sensitive endpoint (bluegill growth) is 1.0 mg/L.

Substance: LAS; C¹⁴-LAS chain length C₁₂ (91% purity) plus unlabeled LAS with average chain length C_{11.6} (C₁₀ 9.7%, C₁₁ 27.9%, C₁₂ 54.4%, C₁₃ 8.0%; 95% purity)

Reference: Maki, A.W. 1981. A laboratory model ecosystem approach to environmental fate and effects studies. Unpublished Internal Report, Environmental Safety Department Procter & Gamble Company, Cincinnati, Ohio.

Reliability: 2 Valid with restrictions

(j)

Type: *In situ* river exposures

Methods: Species or ecosystem studied: Test system was rectangular plexiglass plates (108 cm²) suspended in river water. Plates were colonized with periphyton for four weeks before testing at locations above and below the Xenia sewage treatment plant outfall in the Little Miami River (Ohio). Studies below the outfall assessed the toxicity of LAS in the presence of 2030% treated municipal effluent. Colonized plates were then placed in five submerged plexiglass tubes (1 cm thick, 1 m long), which were then

attached to an aluminium frame supported by rubber floats. LAS stock solutions were stored in 80-L polypropylene containers on the river bank and solutions were pumped daily at a delivery rate adjusted based on river flow to maintain four concentrations (0.2, 1.1, 9.8 and 28.1 mg/L) and a control. Duplicate set ups were used as replicates. Identical set ups were used above and below the outfall.

Test duration 21 days.

Effects monitored: Phytoplankton inhibition of photosynthesis and reduction of the number of taxa was determined.

Chemical analysis: Concentrations were measured.

Results: For inhibition of photosynthesis the best estimated NOEC and LOEC were 9.8 and 28.1 mg/L respectively. At the highest concentration tested the number of taxa was not reduced. For this effect parameter the best estimated NOEC was >28.1 mg/L.

Substance: Dodecyl LAS (CAS #25155-30-3); average chain length C_{11.9}; average molecular weight 346; C₁₀ 9%, C₁₁ 30%, C₁₂ 34%, C₁₃ 19%, C₁₄ 9%

Remarks: A concentration-effect relationship was found. Suspended solids were measured.

Reference: Lewis, M.A., Pittinger, C.A., Davidson, D.H., and Ritchie, C.J. 1993. *In situ* response of natural epiphyton to an anionic surfactant and an environmental safety assessment for phytotoxic effects. *Environ. Toxicol. Chem.* 12:1803-1812.

Reliability: 2 Valid with restrictions

(k)

Type: Laboratory bottles/lake exposures

Methods: Species or ecosystem studied: Bottles (300 ml) filled with lake water were used as the test systems. Nine concentrations were tested in triplicate. Once a month the bottles were suspended in a lake at 1 m depth for three hours. The experiment was repeated for six months. Effects monitored: photosynthetic response of phytoplankton was determined.

Results: Chemical analysis: Concentrations were measured. Mean EC₅₀ values were 3.4 and 1.9 mg/L for C₁₂ and C₁₃, respectively. The ranges of EC₅₀ values for the different tests were 0.5-8.0 and 0.2-8.1 mg/L for C₁₂ and C₁₃, respectively.

Substance: Dodecyl LAS; average chain length C_{11.8} (CAS #25155-30-3); and Tridecyl LAS, average chain length C_{13.3} (CAS #26248-24-8).

Remarks: A concentration-effect relationship was found. Suspended solids were not measured. The wide range of EC₅₀ values is in part due to seasonal differences in temperature and community dynamics.

Reference: Lewis, M.A. and Hamm, B.G. 1986. Environmental modification of the photosynthetic response of lake plankton to surfactants and significance to a laboratory-field comparison. *Water. Res.* 20:1575-1582.

Reliability: 2 Valid with restrictions

4.8 BIOTRANSFORMATION AND KINETICS

(a)

Type: Animal []; Aquatic []; Plant []; Terrestrial []; Other []

Methods: In a flow-through system a 21 day uptake and 14 day elimination experiment was conducted with *Lepomis macrochirus*. The LSC-measured exposure concentration was 0.5 mg/L (1.45 uM). Flow-through; water renewal rate: 20 renewals per day; hardness as CaCO₃ 35 mg/L; pH 7.1; water/fish ratio: 0.04 L/g.

Results: k_1/k_2 concentration factors were for muscle 36, liver 171, blood 237, carcass 64, gall bladder 5224, gill and viscera 282 (on wet weight basis). k_2 values are almost the same (0.24-0.28 d-L) in all tissues but the liver. The value for the liver is double (0.48 d-L) of the other tissues. This indicates biotransformation in the liver. The value of k_1 in the gall bladder (1461 L/kg/d) is at least 16 times higher than those in the other tissues (9-82 L/kg/d).
 t_{95} in the different tissues range between 150 and 300.
 No information on metabolism was given.

Test Substance: Na-alkylbenzenesulfonate average chain length: C_{11.7} (C₁₁ 45%, C₁₂ 36.5%, C₁₃ 18.5%) radiolabeled material (¹⁴C uniformly labelled benzene ring) is spiked with unlabelled LAS (identical homologue composition), spiking ratio 4:1 (unlabeled:¹⁴C labeled).

Remarks: Whole body concentration factors are reliable. Steady state attained within 24 h.

Reference: Kimerle, R.A., Macek, K.J., Sleight, B.H., and Burrows, M.E. 1981. Bioconcentration of linear alkylbenzene sulfonates (LAS) in bluegill (*Lepomis macrochirus*). Water. Res. 15:251-256.

Reliability: 2 Valid with restrictions

(b)

Type: Animal []; Aquatic [X]; Plant []; Terrestrial []; Other []

Methods: Fathead minnows (*Pimephales promelas*) were exposed to LAS in a flow-through system according to OECD guideline 305E.

Results: Steady state uptake was achieved by 96-h of exposure. Uptake constants (k_1) range from 4.3 to 642.2 L/kg/day with C₁₁ and C₁₃ having the lowest and highest, respectively. The elimination rate constants (k_2) range from 0.5 and 1.5 days. k_1 was dependent on hydrophobicity and alkyl chain length. k_2 did not vary with hydrophobicity.

Reference: Tolls, J., Haller, M., De Graaf, I., Thijssen, M.A.T.C., and Sijm, D.T.H.M. 1997. Bioconcentration of LAS: Experimental determination and extrapolation to environmental mixtures. 31:3426-3431.

Reliability: 2 Valid with restrictions

(c)

Type: Animal []; Aquatic [X]; Plant []; Terrestrial []; Other []

Method: In a flow-through system a 14 day uptake experiment was conducted with fish (*Pimephales promelas*). Two concentrations of 0.100 and 0.135 mg/L (0.3 and 0.4 uM) were tested resulting in concentration factors. Tissue extracts of the fish exposed to LAS were analysed by desulfonation-GC. Flow-through; water renewal rate: 3-4 renewals per day; no feeding; hardness 250 mg/L; well water. Chemical analyses by LSC.

Results: On the basis of the concentration in fish tissues (wet weight) and the two concentrations in water concentration factors in muscle were 4 and 3, and in the gall bladder 13,700 and 7,500. It remains unclear whether the observed variation of the concentration factors indicates a concentration dependence of bioaccumulation. The results indicate metabolic

transformation, since the parent compound radioactivity could not account for all the radioactivity in the fish. Percentages of radioactivity accounted for by parent LAS in muscle 50-70%, other organs 50-80%, gall bladder <1%. The authors report clearance of "substantially all ¹⁴C activity" within 3 days.

Test Substance: Linear LAS, chain length C₁₂; uniformly ¹⁴C-labelled benzene ring, 2-phenyl-isomer-content: 17%.
 Remarks: Steady state attained within 144 h.
 Reference: Kimerle, R.A., Swisher, R.D., and Schroeder-Comotto, R.M. 1975. Surfactant structure and aquatic toxicity. Proc. IJC Symposium of Structure Activity Correlations in studies on toxicity and bioconcentration with aquatic organisms, pp. 22-35.
 Reliability: 2 Valid with restrictions

(d)

Type: Animal [X]; Aquatic []; Plant []; Terrestrial []; Other []
 Methods: Fish (*Ictalurus punctatus* 250-450 g) were dosed with LAS via liquid gavage, gavage of food impregnated with LAS, and by intraperitoneal injection. Amount dosed: 425 ug (1.22 umole). Metabolism and elimination pathways were investigated. No aqueous exposure; metabolism chamber operated in a static mode; water was exchanged twice per day; 1 g of food, no feeding of the fish dosed by liquid gavage. Chemical analysis via LSC.
 Results: Percentage eliminated from the different tissues 48 h after the end of the 24 exposure period:

	Percent Eliminated:		
	i.p. injection	Gavage	
		Food	Fluid
Total	42	68	71
Gills	4	49	33
Urine	26	7	11
Faeces+ Skin	12	12	27
CO ₂	<1	<1	<1

Test substance: Linear LAS, chain length C₁₂; uniformly isomer distribution not specified.
 Remarks: Elimination pathways for the three modes of dosing differed. As a considerable fraction of radiolabel administered via gavage is excreted via the gills it can be concluded that:
 a) LAS is resorbed readily in the GI-tract.
 b) The compounds excreted via the gills (LAS or its metabolites) are able to pass the gill membrane.
 Reference: Schmidt, E.J. and Kimerle, R.A. 1981. New design and use of a fish metabolism chamber. In: Branson, D.R. and Dickson, K.L. (eds.). Aquatic Toxicology and Hazard Assessment: Fourth Conference, ASTM STP 737, 436-448.
 Reliability: 2 Valid with restrictions

(e)

Type: Animal []; Aquatic [X]; Plant []; Terrestrial []; Other []
 Method: Fish (*Cyprinus carpio* 34.6 g) were exposed to combinations of LAS,

Results:	<p>polyoxyethylene and sorbitan monooleate to assess the influence of these substances on the uptake of LAS. Exposure period was 3 h. Exposure concentration was 1×10^{-5} M LAS. LSC was used for tissue specific analysis, HPLC was used for measurement of gill adsorption. Dechlorinated tap water, filtered over active carbon; hardness 63 mg/L CaCO₃; water/fish ratio: 0.14 g/L; no feeding during experiment.</p> <p>Concentration factors for specific tissues (measured by LSC) were for blood 4.2, hepatopancreas 4.0, spleen 1.0, kidney 3.3, heart 1.7, brain 0.6, muscles 0.2, gill 11, gall bladder 21. Adsorption of LAS to gills (C_{gill}/C_{water}), measured by HPLC, isomer specific: 2-phenyl 16; 3-phenyl 5.4; 4-phenyl, 2.6; 5- and 6-phenyl 1.9.</p> <p>C₁₂-LAS associated radiolabel is taken up by gills rapidly. It reaches the highest body level in the gall bladder after only three hours. The adsorption to the gills is related to the phenyl-substitution of the alkane. The closer the benzenesulfonate-group is attached to the terminal carbon atom of the alkyl chain, the higher the adsorption to the gills. As the gills are an important organ in the uptake of xenobiotic compounds, it seems reasonable to expect that those isomers which sorb strongly to the gills will also be taken up preferentially.</p>
Test substance:	Labelled linear LAS, isomer distribution not specified. (CAS #25155-30-3)
Remarks:	No whole body concentration factors.
Reference:	Toshima, S., Moriya, T., and Yoshimura, K. 1992. Effects of polyoxyethylene (20) sorbitan monooleate on the acute toxicity of linear alkylbenzenesulfonate (C ₁₂ -LAS) to fish. <i>Ecotox. Environ. Saf.</i> 24:26-36.
Reliability:	2 Valid with restrictions

4.9 ADDITIONAL REMARKS

(a) Remarks:	<p>Pre-1993 published data and company owned aquatic toxicity test data are collected in a common data base (BKH, 1993). Statistical analysis showed:</p> <ul style="list-style-type: none"> -After removal of outliers, the dataset contains 586 records covering 93 species. Algae, crustaceans and fish together make 68% of the 93 species. Dominating species (genera) are: <i>Scenedesmus</i>, <i>Selenastrum</i>, <i>Daphnia</i>, <i>Gammarus</i>, <i>Lepomis</i>, <i>Pimephales</i> and <i>Carassius</i> (71% of the 586 records). 34 of the 93 species are marine species. -LAS does not have a specific mode of action for different species. The variability in the sensitivity between species is comparable to the variability within a species. -The acute to chronic ratio is approximately a factor 5. -The mean LC₅₀ of all species is 5.5 mg/L. -The mean NOEC (chronic) of all species is 0.8 mg/L. -A quantitative structure activity relationship for chain length was determined for fish and crustaceans, longer chain lengths corresponding to higher toxicity. The bioavailability of adsorbed LAS molecules for aquatic organisms as midge larvae, daphnids and fish is assumed to be low, because observed toxicity thresholds in the presence of adsorbing material correspond to the calculated concentration of the fraction LAS
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dissolved and the toxicity data for the completely dissolved substance (Pittinger et al., 1989).

Reference: 1) BKH. 1993. The use of existing toxicity data for estimation of the Maximum Tolerable Environmental Concentration of Linear Alkyl Benzene Sulfonate, Part I: Main report; Part II: Data base. Study carried out for ECOSOL, BKH Consulting Engineers, Delft, NL.
2) Hand, V.C., Rapaport, R.A., Pittinger, C.A. 1990. First validation of a model for the adsorption of linear alkylbenzene sulfonate to sediment and comparison to chronic effects data. *Chemosphere* 21(6):741-750.
3) Pittinger, C.A., Woltering, D.M., and Masters, J.A. 1989. Bioavailability of sediment sorbed and aqueous surfactants to *Chironomus riparius* (midge). *Environm. Toxicol. Chem.* 8:1023-1033.

Reliability: 4 Not assignable

(b)

Results: Neither LAS nor its sulfophenylcarboxylate degradation products displayed any estrogenic effects.

Remarks: A recombinant yeast estrogen screen using *Saccharomycea cerevisiae* was employed to determine the potential estrogenic activity of LAS and its degradation products.

Reference: Routledge, E.J. and Sumpter, J.P. 1996. Estrogenic activity of surfactants and some of their degradation products assessed using a recombinant yeast screen. *Environ. Toxicol. Chem.* 15:241-248.

Reliability: 2 Valid with restrictions

(c)

Type: Endocrine Disruption

Results: In the yeast screen, no statistical differences in absorbance were induced in any concentration of either of the three substances. In the hepatocyte assay, no increase in the concentration of Vg over the concentration of the controls was observed for LAS or any other three test substances. Results of both assays indicate that no estrogenic effects occurred after exposure to LAS or two of its biodegradation intermediates (SPC5 and SPC11).

Method: LAS, SPC5, or SPC11 were prepared in sterile distilled water and added to the culture media. Two *in vitro* screening assays for measurement of estrogenic activity were used: 1) the ER assay, with the recombinant yeast screen, and 2) the vitellogenin assay, with hepatocytes. ER was used as the positive control. Serial dilutions of the test compounds were used, with the maximum concentrations used in the hepatocytes assay were 150 µM (50 mg/L) for LAS, 25 µM (7.4 mg/L) for SPC5, and 200 µM (72.8 mg/L) for SPC11.

Test Substance: 1) LAS-C₁₁, 47% a.i., supplied by Petroquimica Espanola S.A.
2) Sulfophenylcarboxylic acids (SPC5 and SPC11) (formed from successive oxidation of terminal methyl groups on the alkyl chain)

Reference: Navas, J.M., Gonzalez-Mazo, E., Wenzel, A., Gomez-Parra, A., and Segner, H. 1999. Linear alkylbenzene sulfonates and intermediate products from their degradation are not estrogenic. *Marine Pollution Bulletin* 38:880-884.

Reliability: 2 Valid with restrictions

(d)

Results: The final predicted no-effect concentration (PNEC) for C_{11.6} LAS was 250 µg/L based on a single species PNEC of 320 µg/L and the range of field NOECs of 250-500 µg/L. All data values are expressed as dissolved concentrations.

Method: Predicted no-effect concentrations (PNECs) were derived for LAS and three other surfactants using three stages in an aquatic effects assessment. In the Initial stage, assessment factors are applied to available short-term toxicity data. In the Refined stage, statistical extrapolation based on long-term (i.e., chronic) toxicity data are employed. In the Comprehensive stage of effects assessment, a wide variety of laboratory and field model ecosystem studies are incorporated into the analysis. To determine the PNEC for LAS, all data types were compiled and evaluated. Since toxicity is related to carbon chain length, all data were normalized to LAS with a mean carbon chain length of 11.6, the structure typically present in the environment based on the monitoring study described by Matthijs et al. 1999.

Remarks: For LAS, the predicted environmental concentrations (PECs) in the environment are about 50 to 100 times lower than the PNECs. This PNEC determination is part of an extensive monitoring program executed jointly by the Dutch Soap Association (NVZ) and the Dutch Ministry of Housing, Spatial Planning and the Environment (VROM).

Reference: van de Plassche, E.J., de Bruijn, J.H.M., Stephenson, R.R., Marshall, S.J., Feijtel, T.C.J., and Belanger, S.E. 1999. Predicted no-effect concentrations and risk characterization of four surfactants: Linear alkylbenzene sulfonate, alcohol ethoxylates, alcohol ethoxylated sulfates, and soap. *Environmental Toxicology and Chemistry* 18:2653-2663.

Reliability: 2 Valid with restrictions

(e)

Results: A realistic worst-case estimation of the LAS concentration in sludge-amended soil is predicted to be 7 mg/kg dry weight, which is compared to the PNEC of 4.6 mg/kg. The LAS concentration will drop to a level below the PNEC within 6 to 24 days after sludge application, depending on the degradation rate of LAS.

Methods: LAS can be found in high concentrations in sewage sludge and may enter the soil compartment as a result of sludge application. To evaluate the effects and risk to soil organisms, a probabilistic (log-normal) distribution model was used to predict a no effect concentration (PNEC) for soil fauna, flora, and a combination of these. By extrapolation, the method determines a lower statistical tolerance limit. The preferred inputs to the current model are EC₁₀ data from laboratory studies. By use of the log-normal distribution, a concentration (K_p) is found, for which the EC₁₀ or NOEC values for 95% of all species in the community are greater. The value of K_p is used as the estimate of the PNEC. The soil concentration after sludge application was predicted by a number of scenarios and used as the predicted environmental concentration (PEC) in the risk characterization and calculation of risk quotients (RQ = PEC/PNEC). A LAS concentration of 4.6 mg/kg was used as the current best estimate of PNEC in all RQ calculations. The exposure scenarios included three levels of LAS contamination (530, 2,600 and 16,100 mg/kg), three half-lives (10, 25 and 40 days) and five different sludge loads (2, 4, 6, 8 and 10 t/ha).

Remarks: Even in the most extreme scenarios, the level of LAS is expected to be far below the estimated PNEC one year after application.

Test Substance: LAS (various, based on each study used)

Reference: Jensen, J., Lokke, H., Holmstrup, M., Krogh, P.H., and Elsgaard, L. 2001. Effects and risk assessment of linear alkylbenzene sulfonates in agricultural soil. 5. Probabilistic risk assessment of linear alkylbenzene sulfonates in sludge-amended soils. Environmental Toxicology and Chemistry 20:1690-1697.

Reliability: 2 Valid with restrictions

5. TOXICITY

5.1 ACUTE TOXICITY

5.1.1 ACUTE ORAL TOXICITY

(a) (Rat)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LDL₀ []; Other []
Species/strain: rat
Value: 1080 mg/kg bw
Method: OECD Guide-line 401 "Acute Oral Toxicity" 1981. Five male and five female rats were given LAS doses of 1075, 1220, 1360, 1710 or a control by gavage. Body weight and other signs were measured on days 7 and 14. Temperature was maintained at 20⁺/₋₁°C with a 12 hr light-dark cycle. Animals were observed for 14 days after dosing.
GLP: Yes [] No [X] ? []
Test substance: Marlon A 386 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}; Activity: 86%
Remarks: Symptoms beginning about 30 minutes past application included diarrhea, squatting attitude, breathing difficulties, nose bleeding, ataxia, and lethargy. These symptoms had disappeared in surviving animals by 120 hours. Virtually all animals died in doses of 1220 mg/kg and above. Note that all doses are corrected for 86% activity. The original doses were 1250, 1415, 1580 and 1990 mg/kg.
Reference: Murmann, P. 1984a. Akute orale Toxizitat von Marlon A 386 fur Ratten. Huels Report No. 0191.
Reliability: 2 Valid with restrictions

(b)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LDL₀ []; Other []
Species/strain: rat
Value: 1630 mg/kg bw
Method: OECD Guide-line 401 "Acute Oral Toxicity" 1981. Five male and five female rats were given LAS doses of 1260, 1580, 1785, and 1990 or a control by gavage. Body weight and other signs were measured on days 7 and 14. Temperature was maintained at 20⁺/₋₁°C with a 12 hr light-dark cycle. Animals were observed for 14 days after dosing.
GLP: Yes [] No [X] ? []
Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}; Activity: 50%
Remarks: Symptoms beginning about 1-4 hours past application included diarrhea, squatting attitude, breathing difficulties, nose bleeding, ataxia, and lethargy. The symptoms in the lower doses disappeared with 24 to 48 hours. Symptoms disappeared in the 1785 mg/kg dose and higher within 8 days. Virtually all animals died in doses of 1785 mg/kg and above. Note that all doses are corrected for 50% activity. The original doses were 2510, 3160, 3570 and 3980 mg/kg.
Reference: Murmann, P. 1984c. Akute orale Toxizitat von Marlon A 350 fur Ratten. Huels Report No. 209.
Reliability: 2 Valid with restrictions

(c)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LD_{L0} []; Other []

Species/strain: rat

Value: 1410 mg/kg bw

Method: OECD Guide-line 401 "Acute Oral Toxicity" 1981. Five male and five female rats were given LAS doses of 1190, 1500 and 1890 or a control by gavage. Body weight and other signs were measured on days 7 and 14. Temperature was maintained at 20[±]-1°C with a 12 hr light-dark cycle. Animals were observed for 14 days after dosing.

GLP: Yes [] No [X] ? []

Test substance: Marlon A 330 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}; Activity: 30%.

Remarks: Symptoms beginning about 90 minutes past application included diarrhea, squatting attitude, breathing difficulties, nose bleeding, ataxia, and lethargy. These symptoms had disappeared in surviving animals by 72 hours. Virtually all animals died in doses of 1500 mg/kg and above. Note that all doses are corrected for 30% activity. The original doses were 3980, 5010 and 6310 mg/kg.

Reference: Murmann, P. 1984a. Akute orale Toxizität von Marlon A 330 für Ratten. Huels Report No. 0186.

Reliability: 2 Valid with restrictions

(d)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LD_{L0} []; Other []

Species/strain: rat

Value: LD₅₀ for male animals: 1460 mg/kg
LD₅₀ for female animals: 1470 mg/kg

Method: Male and female rats were given a single dose of LAS by gavage and observed for mortality.

GLP: Yes [] No [X] ? []

Test substance: C₁₀₋₁₃ LAS, sodium salt (CAS #68411-30-3)
<C₁₀ 0.1%, C₁₀ 10.1%, C₁₁ 33.7%, C₁₂ 31%, C₁₃ 25.1%.; average alkyl chain length = C_{11.7}; activity: 99.5%

Remarks: Information as reported in IPCS document.

Reference: Ito, R., Kawamura, H., Chang, H.S., Kudo, K., Kajiwara, S., Toida, S., Seki, Y., Hashimoto, M., and Fukushima, A. 1978. Acute, subacute and chronic toxicity of magnesium linear alkylbenzene sulfonate (LAS-Mg). J. Med. Soc. Toho, Japan. 25 (5-6):850-875 (in Japanese). Referenced in IPCS, Environmental Health Criteria 169. Linear Alkylbenzene Sulfonates and Related Compounds, WHO.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document, therefore it is considered to be reliable.

(e)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LD_{L0} []; Other []

Species/strain: Rat, CFY (Sprague-Dawley origin); male and female

Value: 1980 mg/kg bw

Method: OECD Guideline 401. Five male and five female rats were given single doses by gavage at 1500, 2350 and 3760 mg/kg bw. Rats were housed in cages grouped by sex and given standard laboratory diet and water *ad*

libitum. Mean daily temperature was maintained at 21-22°C at a mean relative humidity of 56%. Lighting was on a 12 hrs dark and 12 hrs light photoperiod. Animals were observed for 14 days after dosing.

GLP: Yes No ?
Test substance: Alkylbenzene sulfonate, sodium salt (designated as P-500 N-Na). Activity 47%. Average alkyl chain length = C_{11.2}. Clear yellow liquid.
Remarks: Four rats from each of the two lowest concentrations and all rats from the highest concentration died. All deaths occurred between 6 and 23 hours after dosing. Signs of reaction to treatment included pilo-erection, hunched posture, abnormal gait (waddling), lethargy, decreased respiratory rate, ptosis, pallor of the extremities, and diarrhea. All surviving animals appeared to recover completely by day 4. Autopsy of rats that died revealed isolated cases of pallor of the kidneys or spleen. Terminal necropsy findings for survivors were normal. Note that all doses are corrected for 47% activity. The original doses were 3200, 5000, and 8000 mg/kg.
Reference: Kynoch, S.R. 1986a. Acute oral toxicity to rats: P-500 N-Na. Huntingdon Research Center Report. No. 86546D/PEQ 7/AC.
Reliability: 1 Valid without restriction

(f)

Type: LD₀ ; LD₁₀₀ ; LD₅₀ ; LD_{L0} ; Other
Species/strain: Sprague-Dawley strain albino male and female rats
Value: 1320 mg/kg (lower limit: 1220 mg/kg; upper limit: 1430 mg/kg)
Method: Acute Oral Minimal Lethal Dose Test
The test substance was applied as a 20% aqueous solution by stomach tube to Sprague-Dawley strain albino male and female rats. After the approximate LD₅₀ was determined, groups of male and female rats were fed in increasing doses at increments of 0.1 fractional log intervals at three levels (1000, 1260 and 1580 mg/kg) designed to blanket the toxicity range thereby supplying data for calculation of the LD₅₀ which was performed according to the method of E.J. de Beer. Observations of toxic signs were recorded and the viscera of the test animals were examined macroscopically.

GLP: Yes No ?
Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-225); C₉ 1%; C₁₀ 7%, C₁₁ 25%, C₁₂ 48%, C₁₃ 19%, C₁₄ 1%; average alkyl chain length = C_{11.9}.
Remarks: The compound was classified as mildly toxic by oral ingestion in male and female rats. All rats at 1000 mg/kg and 4 of 5 rats at 1260 mg/kg survived. Survival time was one to two days for rats that died at 1580 mg/kg. The toxic signs included reduced appetite and activity (one to two days in survivors), diarrhoea, increasing weakness, collapse, and death. The autopsy revealed hemorrhagic lungs, liver discoloration, and acute gastrointestinal inflammation. Surviving animals were sacrificed seven days after dosing. In these animals the viscera appeared normal by macroscopic examination.
Reference: Monsanto Company. 1971. Linear alkylbenzene sodium sulfonate – Alkylate 225 Lot CC 6450 – Acute toxicity screen. Project No. Y-71-119. Unpublished report.
Reliability: 2 Valid with restrictions

(g)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LDL₀ []; Other []
Species/strain: Sprague-Dawley strain albino male and female rats
Value: 1430 mg/kg (lower limit: 1300 mg/kg; upper limit: 1570 mg/kg)
Method: Acute Oral Minimal Lethal Dose Test
The test substance was applied as a 10% aqueous solution by stomach tube to Sprague-Dawley strain albino male and female rats. After the approximate LD₅₀ was determined, groups of male and female rats were fed in increasing doses at increments of 0.1 fractional log intervals at four levels (1000, 1260, 1580 and 2000 mg/kg) designed to blanket the toxicity range thereby supplying data for calculation of the LD₅₀ which was performed according to the method of E.J. de Beer. Observations of toxic signs were recorded and the viscera of the test animals were examined macroscopically.

GLP: Yes [] No [X] ? []
Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-215); C₉ 1%, C₁₀ 16%, C₁₁ 43%, C₁₂ 40%, C₁₃ 1%, C₁₄ <1%; average alkyl chain length = C_{11.35}.
Remarks: The compound was classified as mildly toxic by oral ingestion in male and female rats. All rats survived at 1000 mg/kg. Three of 5 and 2 of 5 survived 1260 and 1580, respectively. Survival time was sixteen hours to two days in the rats that died. The toxic signs included reduced appetite and activity (two to three days in survivors), increasing weakness, slight tremors, collapse, and death. The autopsy revealed lung and liver hyperaemia and gastrointestinal inflammation. Surviving animals were sacrificed seven days after dosing. In these animals the viscera appeared normal by macroscopic examination.

Reference: Monsanto Company. 1972a. Linear alkylbenzene sodium sulfonate – Alkylate 215 Lot CC 6772S – Acute toxicity screen. Project No. Y-72-274. Unpublished report.

Reliability: 2 Valid with restrictions

(h)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LDL₀ []; Other []
Species/strain: Sprague-Dawley strain albino male and female rats
Value: 1360 mg/kg (lower limit: 1240 mg/kg; upper limit: 1500 mg/kg)
Method: Acute Oral Minimal Lethal Dose Test
The test substance was applied as a 10% aqueous solution by stomach tube to Sprague-Dawley strain albino male and female rats. After the approximate LD₅₀ was determined, groups of male and female rats were fed in increasing doses at increments of 0.1 fractional log intervals at four levels (1000, 1260, 1580 and 2000 mg/kg) designed to blanket the toxicity range thereby supplying data for calculation of the LD₅₀ which was performed according to the method of E.J. de Beer. Observations of toxic signs were recorded and the viscera of the test animals were examined macroscopically.

GLP: Yes [] No [X] ? []
Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-222L); average alkyl chain length = C_{11.5}.
Remarks: The compound was classified as slightly toxic by oral ingestion in male and female rats. Survival time was sixteen hours to three days in the rats that died. The toxic signs included reduced appetite and activity (one to three days in survivors), increasing weakness, collapse, and death. The autopsy revealed lung and liver hyperaemia and gastrointestinal

inflammation. Surviving animals were sacrificed seven days after dosing. In these animals the viscera appeared normal by macroscopic examination.

Reference: Monsanto Company. 1972b. Linear alkylbenzene sodium sulfonate – Alkylate 222L Lot CC 6773S – Acute toxicity screen. Project No. Y-72-275. Unpublished report.

Reliability: 2 Valid with restrictions

(i) (Mouse)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LD_{L0} []; Other []

Species/strain: mouse

Value: LD₅₀ male animals: 2160 mg/kg
LD₅₀ female animals: 2250 mg/kg

Method: Male and female mice were given a single dose of LAS and observed for mortality.

GLP: Yes [] No [X] ? []

Test substance: C₁₀₋₁₃ LAS, sodium salt (CAS #68411-30-3).
<C₁₀ 0.1%, C₁₀ 10.1%, C₁₁ 33.7 %, C₁₂ 31%, C₁₃ 25.1%; average alkyl chain length = C_{11.7}; activity: 99.5%

Remarks: Information as cited in IPCS document.

Reference: Ito, R., Kawamura, H., Chang, H.S., Kudo, K., Kajiwara, S., Toida, S., Seki, Y., Hashimoto, M., and Fukushima, A. 1978. Acute, subacute and chronic toxicity of magnesium linear alkylbenzene sulfonate (LAS-Mg). J. Med. Soc. Toho, Japan. 25 (5-6):850-875 (in Japanese). Referenced in IPCS, Environmental Health Criteria 169. Linear Alkylbenzene Sulfonates and Related Compounds, WHO.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

5.1.2 ACUTE INHALATION TOXICITY

Type: LC₀ []; LC₁₀₀ []; LC₅₀ []; LCL₀ []; Other [X] Approximate lethal concentration (ALC)

Species/strain: Rat/Male Ctrl: CD (SD) BR

Exposure time: single 4-hour period

Value: 310 mg/m³ of particulate

Method: Groups of six male 8-week old rats were restrained in perforated, stainless steel cylinders with conical nose pieces. Exposure was nose-only to an aerosol atmosphere for 4 hours. After exposure, rats were returned to their cages and observed for clinical signs for 14 days. Mean measured concentrations in the test chambers were 65, 120, 260, and 310 mg/m³. Chamber temperature ranged between 25-26°C.

GLP: Yes [] No [] ? [X]

Test substance: LAS (CAS #25155-30-0); activity 98%

Remarks: The ALC is defined as the lowest atmospheric concentration generated that caused death in 1 or more rats either on the day of exposure or within 14 days post exposure. No mortality occurred at concentrations up to 260 mg/m³. At 310 mg/m³, one rat died during exposure and 2 rats died one day post exposure. The test material is considered moderately toxic by inhalation. However, it is important to note that this laboratory exposure is not representative of the possible LAS exposure during actual use. In

this study, animals were given high exposures to respirable-sized particles (MMAD at 310 mg/m³ = 2.5 microns). Spray products containing LAS are designed to produce large particle sizes. These large particles are needed for efficient delivery of the spray to the surface being cleaned. This results in particle sizes that are much larger than the respirable particle sizes used in testing and therefore would not be able to reach far into the lungs where effects could occur. Given this lack of relevance to real-world exposure potential, the use of this study for risk assessment purposes is limited.

Reference: Kinney, L.A. 1985. Approximate lethal concentrations (ALCs) by inhalation of sodium lauryl sulfate & sodium dodecylbenzene sulfonate. Dupont Haskell Laboratory Report No. 474-84.

Reliability: 2 Valid with restrictions

5.1.3 ACUTE DERMAL TOXICITY

(a)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LDL₀ []; Other []

Species: Rat, CFY (Sprague-Dawley origin); male and female

Value: >2000 mg/kg bw

Method: OECD Guideline 402. Five male and five female rats were exposed to 2000 mg/kg in a limit test. The test substance was applied to clipped intact skin in the dorso-lumbar region and covered with gauze held in place with an impermeable dressing. The dressing was removed after 24 hours and the treated area of the skin washed with warm water and blotted dry. Observations for dermal irritation were made daily for 14 days.

GLP: Yes [X] No [] ? []

Test substance: Alkylbenzene sulfonate, sodium salt (designated as P-500 N-Na). activity 47%. Average alkyl chain length = C_{11.2}. Yellow, viscous liquid.

Remarks: There were no deaths or signs of a systemic reaction following a single dermal application at 2000 mg/kg bw. Well defined or slight erythema and slight oedema were observed at all test sites after removal of the occlusive dressing on Day 2. All test sites were entirely covered by scab formation from Day 7. Sloughing from the scabbed skin began at various times between Day 7 and Day 12 and was completed before termination. Low bodyweight gains or loss of body weight were recorded for one male and three females in Day 8. Two of the same females and a third female also showed low bodyweight gain between Days 8 and 15.

Reference: Kynoch, S.R. 1986b. Acute dermal toxicity to rats of P-500 N-Na. Huntingdon Research Center. Report No. 86718D/PEQ 8/AC.

Reliability: 1 Valid without restriction

(b)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LDL₀ []; Other []

Species/strain: New Zealand white rabbit

Value: > 200 mg/kg and < 316 mg/kg

Method: Acute Skin Absorption Minimal Lethal Dose Test

The test substance was applied as a 30% aqueous solution and the doses of the solution administered were 126, 200, 316, 501, 794, 1260, 2000, 3160 and 5010 mg/kg. The doses were administered to a closely clipped area of intact skin of male and female rabbits (1 animal/dose). The treated areas

were covered with plastic strips and the animals were placed in wooden stocks for up to twenty-four hours. After the twenty-four hours the animals were assigned to individual cages. Observations of toxic signs were recorded daily and the viscera of the test animals were examined macroscopically.

GLP: Yes [] No [X] ? []
Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-225; C₉ 1%; C₁₀ 7%, C₁₁ 25%, C₁₂ 48%, C₁₃ 19%, C₁₄ 1%; average alkyl chain length = C_{11.9})
Remarks: The test substance was classified as moderately toxic. Animals exposed dermally to 126 and 200 mg/kg survived for the 14 day study duration. Survival time for animals receiving all other doses ranged from two to eight days. The toxic signs included reduced appetite and activity, increasing weakness, collapse and death. The autopsy revealed hemorrhagic lungs, liver discoloration, enlarged gall bladder, and gastrointestinal inflammation. The animals that survived were sacrificed fourteen days after dosing. In these animals the viscera appeared normal by macroscopic examination.
Reference: Monsanto Company. 1971. Linear alkylbenzene sodium sulfonate – Alkylate 225 Lot CC 6450 – Acute toxicity screen. Project No. Y-71-119. Unpublished report.
Reliability: 4 (insufficient animals per dose of mixed sex, etc.)

(c)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LDL₀ []; Other []
Species/strain: New Zealand white rabbit
Value: > 631 mg/kg and < 1000 mg/kg
Method: Acute Skin Absorption Minimal Lethal Dose Test
The test substance was applied as a 20% aqueous solution and the doses of the solution administered were 200, 316, 631, 1000, 1260, 2000 and 3160 mg/kg. The doses were administered to a closely clipped area of intact skin of male and female rabbits (1 animal/dose). The treated areas were covered with plastic strips and the animals were placed in wooden stocks for up to twenty-four hours. After the twenty-four hours the animals were assigned to individual cages. Observations of toxic signs were recorded daily and the viscera of the test animals were examined macroscopically.

GLP: Yes [] No [X] ? []
Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-215; C₉ 1%, C₁₀ 16%, C₁₁ 43%, C₁₂ 40%, C₁₃ 1%, C₁₄ <1%; average alkyl chain length = C_{11.35})
Remarks: The test substance was classified as moderately toxic. Survival time ranged from one to two days. The toxic signs included reduced appetite and activity, increasing weakness, collapse and death. The autopsy revealed lung hyperemia, areas of liver discoloration and gastrointestinal inflammation. The animals that survived were sacrificed fourteen days after dosing. In these animals the viscera appeared normal by macroscopic examination.
Reference: Monsanto Company. 1972a. Linear alkylbenzene sodium sulfonate – Alkylate 215 Lot CC 6772S – Acute toxicity screen. Project No. Y-72-274. Unpublished report.
Reliability: 4 (insufficient animals per dose of mixed sex, etc.)

(d)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LDL₀ []; Other []

Species/strain: New Zealand white rabbit
 Value: > 631 mg/kg and < 1000 mg/kg
 Method: Acute Skin Absorption Minimal Lethal Dose Test
 The test substance was applied as a 20% aqueous solution and the doses of the solution administered were 200, 398, 631, 1000, 1260, 2000 and 3160 mg/kg. The doses were administered to a closely clipped area of intact skin of male and female rabbits (1 animal/dose). The treated areas were covered with plastic strips and the animals were placed in wooden stocks for up to twenty-four hours. After the twenty-four hours the animals were assigned to individual cages. Observations of toxic signs were recorded daily and the viscera of the test animals were examined macroscopically.

GLP: Yes No ?
 Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-222L; average alkyl chain length = C_{11.5})
 Remarks: The test substance was classified as moderately toxic. Survival time was two days. The toxic signs were reduced appetite and activity, increasing weakness, collapse and death. The autopsy revealed lung hyperemia, areas of liver discoloration and gastrointestinal inflammation. The animals that survived were sacrificed fourteen days after dosing. In these animals the viscera appeared normal by macroscopic examination.
 Reference: Monsanto Company. 1972b. Linear alkylbenzene sodium sulfonate – Alkylate 222L Lot CC 6773S – Acute toxicity screen. Project No. Y-72-275. Unpublished report.
 Reliability: 4 (insufficient animals per dose of mixed sex, etc.)

5.1.4 ACUTE TOXICITY, OTHER ROUTES OF ADMINISTRATION

(a)
 Type: LD₀ ; LD₁₀₀ ; LD₅₀ ; LDL₀ ; Other
 Species/strain: rat
 Administration: i.m. ; i.p. ; i.v. ; infusion ; s.c. ; other
 Value: Females = 810 mg/kg; males = 840 mg/kg bw
 Method: Rats were given subcutaneous injections of LAS
 GLP: Yes No ?
 Test substance: C₁₀₋₁₃ LAS, sodium salt (CAS #68411-30-3)
 <C₁₀ 0.1%, C₁₀ 10.1%, C₁₁ 33.7 %, C₁₂ 31%, C₁₃ 25.1%; average alkyl chain length = C_{11.7}; activity: 99.5%
 Remarks: Information as cited in IPCS document.
 Reference: Ito, R., Kawamura, H., Chang, H.S., Kudo, K., Kajiwara, S., Toida, S., Seki, Y., Hashimoto, M., and Fukushima, A. 1978. Acute, subacute and chronic toxicity of magnesium linear alkylbenzene sulfonate (LAS-Mg). J. Med. Soc. Toho, Japan. 25 (5-6):850-875 (in Japanese). Referenced in IPCS, Environmental Health Criteria 169. Linear Alkylbenzene Sulfonates and Related Compounds.
 Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(b)
 Type: LD₀ ; LD₁₀₀ ; LD₅₀ ; LDL₀ ; Other
 Species/strain: mouse

Administration: i.m. []; i.p. []; i.v. []; infusion []; s.c. [X]; other []
Value: Females = 1400 mg/kg; males = 1250 mg/kg bw
Method: Mice were given subcutaneous injections of LAS.
GLP: Yes [] No [X] ? []
Test substance: C₁₀₋₁₃ LAS, sodium salt (CAS #68411-30-3)
<C₁₀ 0.1%, C₁₀ 10.1%, C₁₁ 33.7 %, C₁₂ 31%, C₁₃ 25.1%; average alkyl
chain length = C_{11.7}; activity: 99.5%
Remarks: Information as cited in IPCS document.
Reference: Ito, R., Kawamura, H., Chang, H.S., Kudo, K., Kajiwara, S., Toida, S.,
Seki, Y., Hashimoto, M., and Fukushima, A. 1978. Acute, subacute
and chronic toxicity of magnesium linear alkylbenzene sulfonate (LAS-
Mg). J. Med. Soc. Toho, Japan. 25 (5-6):850-875 (in Japanese).
Referenced in IPCS, Environmental Health Criteria 169. Linear
Alkylbenzene Sulfonates and Related Compounds.
Reliability: 4 This study is assigned a reliability score of 4 because the original
report was not available for review. However, the study was evaluated
by IPCS prior to inclusion in their criteria document.

(c)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LD_{L0} []; Other []
Species/strain: rat
Administration: i.m. []; i.p. []; i.v. [X]; infusion []; s.c. []; other []
Value: Females = 126 mg/kg; males = 119 mg/kg bw
Method: Rats were given intravenous injections of LAS.
GLP: Yes [] No [X] ? []
Test substance: C₁₀₋₁₃ LAS, sodium salt (CAS #68411-30-3)
<C₁₀ 0.1%, C₁₀ 10.1%, C₁₁ 33.7 %, C₁₂ 31%, C₁₃ 25.1%; average alkyl
chain length = C_{11.7}; activity: 99.5%
Remarks: Information as cited in IPCS document.
Reference: Ito, R., Kawamura, H., Chang, H.S., Kudo, K., Kajiwara, S., Toida, S.,
Seki, Y., Hashimoto, M., and Fukushima, A. 1978. Acute, subacute
and chronic toxicity of magnesium linear alkylbenzene sulfonate (LAS-
Mg). J. Med. Soc. Toho, Japan. 25 (5-6):850-875 (in Japanese).
Referenced in IPCS, Environmental Health Criteria 169. Linear
Alkylbenzene Sulfonates and Related Compounds.
Reliability: 4 This study is assigned a reliability score of 4 because the original
report was not available for review. However, the study was evaluated
by IPCS prior to inclusion in their criteria document.

(d)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LD_{L0} []; Other []
Species/strain: mouse
Administration: i.m. []; i.p. []; i.v. [X]; infusion []; s.c. []; other []
Value: Females = 298 mg/kg; males = 207 mg/kg bw
Method: Mice were given intravenous injections of LAS.
GLP: Yes [] No [X] ? []
Test substance: C₁₀₋₁₃ LAS, sodium salt (CAS #68411-30-3)
<C₁₀ 0.1%, C₁₀ 10.1%, C₁₁ 33.7 %, C₁₂ 31%, C₁₃ 25.1%; average alkyl
chain length = C_{11.7}; activity: 99.5%
Remarks: Information as cited in IPCS document.
Reference: Ito, R., Kawamura, H., Chang, H.S., Kudo, K., Kajiwara, S., Toida, S.,
Seki, Y., Hashimoto, M., and Fukushima, A. 1978. Acute, subacute

and chronic toxicity of magnesium linear alkylbenzene sulfonate (LAS-Mg). J. Med. Soc. Toho, Japan. 25 (5-6):850-875 (in Japanese). Referenced in IPCS, Environmental Health Criteria 169. Linear Alkylbenzene Sulfonates and Related Compounds.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(e)

Type: LD₀ []; LD₁₀₀ []; LD₅₀ [X]; LD_{L0} []; Other []
Species/strain: mouse, albino, Harlan strain
Administration: i.m. []; i.p. []; i.v. [X]; infusion []; s.c. []; other []
Exposure time: 24 hours
Value: 105 mg/kg bw (C₁₂ LAS); 115 mg/kg bw (C₁₀ LAS)
Method: Mice in groups of 10 were given intravenous injections of C₁₀ or C₁₂ LAS.
GLP: Yes [] No [X] ? []
Test substance: C₁₂ LAS homologue; C₁₀ LAS homologue
Remarks: Varying doses were given with an increasing increment between doses of 20% or less.
Reference: Hopper, S.S., Hulpieu, H.R. and Cole, V.V. 1949. Some toxicological properties of surface active agents. J. Am. Pharmacol. Assoc. 38:428-432.
Reliability: 2 Valid with restrictions

5.2 CORROSIVENESS/IRRITATION

5.2.1 SKIN IRRITATION/CORROSION

(a)

Species: New Zealand albino rabbits
Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating []; Moderate irritating []; Slightly irritating []; Not irritating []; Moderate to severe irritating [X]
Method: OECD Guideline 404. A 0.5 ml aliquot of P-500 N-Na was applied under a 2.5 cm² gauze pad to an approximate 10 cm² area of clipped intact skin of 3 rabbits. Each treatment site was occluded with an elastic adhesive dressing for four hours, after which the dressing was removed and the area washed with distilled water. Examination of the treated skin was made approximately 30 minutes after removal of the patch and daily through 14 days. Grading and scoring of the dermal reactions was performed using the standard numerical scoring system.
GLP: Yes [X] No [] ? []
Test substance: Alkylbenzene sulfonate, sodium salt (designated as P-500 N-Na). Activity 47%. Average alkyl chain length = C_{11.2}. Clear yellow liquid.
Remarks: Well defined to moderate skin reactions were observed in all three animals following removal of the bandages. Desquamation of the stratum corneum was observed in all three animals. The reaction in all three animals gradually ameliorated from Days 5, 10 and 11, respectively, and had all resolved completely in one animal by Day 12.
Reference: Liggett, M.P. and Parcell, B.I. 1986a. Irritant effects on rabbit skin of P-500 N-Na. Huntingdon Research Center. Report No. 86400D/PEQ 9/SE.
Reliability: 1 Valid without restriction

(b)

Species/strain: New Zealand albino rabbits

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating [X]; Moderate irritating []; Slightly irritating []; Not irritating []

Method: OECD Guideline 404. A single dose of 0.5 g was applied to the clipped intact skin of six rabbits and secured with a gauze pad covered by an impermeable covering. The patch was removed after a 4 hour exposure and the site washed with water. Animals were examined for signs of erythema and oedema and the responses scored at 60 minutes, 24, 48 and 72 hr, and daily as needed up to 7 days. Skin irritation was scored using the standard rating system and averaged.

GLP: Yes [] No [X] ? []

Test substance: LAS activity 50%; average alkyl chain length = C_{11.6}

Remarks: Average score after 72 hours were 2.4 and 2.83 for erythema and oedema, respectively.

Reference: Biolab SGS. 1989a. Primary skin irritation. Report No. T00428/4.

Reliability: 2 Valid with restrictions

(c)

Species/strain: rabbit

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating [X]; Moderate irritating []; Slightly irritating []; Not irritating []

Classification: Highly corrosive (causes severe burns) []; Corrosive (causes burns) []; Irritating [X]; Not irritating []

Method: OECD Guide-line 404 "Acute Dermal Irritation/Corrosion" 1984

GLP: Yes [X] No [] ? []

Test substance: C₁₀ LAS (CAS #1322-98-1) and C₁₂ LAS (CAS #25155-30-0).

Remarks: Reference reports the results of many experiments conducted on LAS and other surfactants.

Reference: Kaestner, W. 1997. Local tolerance (animal tests): mucous membranes and skin. In: Anionic Surfactants: Biochemistry, Toxicology, Dermatology. 2nd Edition.

Reliability: 4 Not assignable

(d)

Species/strain: rabbit

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating [X]; Moderate irritating []; Slightly irritating []; Not irritating []

Classification: Highly corrosive (causes severe burns) []; Corrosive (causes burns) []; Irritating [X]; Not irritating []

Method: OECD Guide-line 404 "Acute Dermal Irritation/Corrosion" 1981. Three male and female rabbits received 0.5 ml of 50% active material to the shaved intact skin.

GLP: Yes [] No [X] ? []

Test substance: Marlon A 350 (CAS # 68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}; activity: 50%.

Remarks: Mean irritation index: 5.1 out of 8. Individual scores: edema: 2.28, erythema: 3.0

Reference: Murmann, P. 1983a. Prufung der akuten Hautreizwirkung von Marlon A 350. Huels Report No. 0171.

Reliability: 2 Valid with restrictions

(e)

Species/strain: New Zealand albino rabbits

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating []; Moderately irritating [X]; Slightly irritating []; Not irritating []

Method: OECD Guideline 404. A single dose of 0.5 g was applied to the clipped intact skin of six rabbits and secured with a gauze pad covered by an impermeable covering. The patch is removed after a 4 hour exposure and the site washed with water. A second site with the skin abraded received the same treatment. Animals are examined for signs of erythema and oedema and the responses scored at 60 minutes, 24, 48 and 72 hr, and daily as needed up to 7 days. Skin irritation is scored using the standard rating system and averaged.

GLP: Yes [] No [X] ? []

Test substance: LAS (made from Sirene LAB); activity 5%; average alkyl chain length = C_{11.6}

Remarks: Average scores after 72 hours were 1.67 and 2.17 for erythema and oedema, respectively. The primary irritation index was calculated to be 3.82, which classifies 5% LAS as a moderate skin irritant. No differences were observed between intact and abraded skin.

Reference: Biolab SGS. 1989b. Primary skin irritation. Report No. T343.

Reliability: 2 Valid with restrictions

(f)

Species/strain: New Zealand albino rabbits

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating []; Moderate irritating []; Slightly irritating []; Not irritating [X]

Method: OECD Guideline 404. A single dose of 0.5 g was applied to the clipped intact skin of six rabbits and secured with a gauze pad covered by an impermeable covering. The patch is removed after a 4 hour exposure and the site washed with water. A second site with the skin abraded received the same treatment. Animals are examined for signs of erythema and oedema and the responses scored at 60 minutes, 24, 48 and 72 hr, and daily as needed up to 7 days. Skin irritation is scored using the standard rating system and averaged.

GLP: Yes [] No [X] ? []

Test substance: LAS (made from Sirene LAB); activity 2.5%; average alkyl chain length = C_{11.6}

Remarks: No signs of irritation were observed during the study.

Reference: Biolab SGS. 1989c. Primary skin irritation. Report No. T00430/2.

Reliability: 2 Valid with restrictions

(g)

Species/strain: New Zealand albino rabbits

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating []; Moderate irritating []; Slightly irritating []; Not irritating [X]

Method: OECD Guideline 404. A single dose of 0.5 g was applied to the clipped intact skin of six rabbits and secured with a gauze pad covered by an impermeable covering. The patch is removed after a 4 hour exposure and the site washed with water. A second site with the skin abraded received the same treatment. Animals are examined for signs of erythema and oedema and the responses scored at 60 minutes, 24, 48 and 72 hr, and daily as needed up to 7 days. Skin irritation is scored using the standard rating system and averaged.

GLP: Yes No ?

Test substance: LAS (made from Sirene LAB); activity 1%; average alkyl chain length = C_{11.6}

Remarks: No signs of irritation were observed during the study.

Reference: Biolab SGS. 1983. Primary skin irritation. Report No. T116/2.

Reliability: 2 Valid with restrictions

(h)

Species/strain: albino male and female rabbits

Results: Highly corrosive ; Corrosive ; Highly irritating ; Irritating ; Moderate irritating ; Slightly irritating ; Not irritating

Classification: Highly corrosive (causes severe burns) ; Corrosive (causes burns) ; Irritating ; Not irritating

Method: The clipped, intact and abraded skin of six albino male and female rabbits was exposed to 0.5 g of finely ground sample moistened with water under a one inch by one inch square patch, two single layers thick. The patches were positioned in place with adhesive tape. The trunk of each animal was wrapped with plastic strips to avoid contamination and retard evaporation for the 24 hour exposure period. Observations were made over a period of seven days for irritation. The data was scored according to the method of Draize, et al.

GLP: Yes No ?

Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-225); C₉ 1%, C₁₀ 7%, C₁₁ 25%, C₁₂ 48%, C₁₃ 19%, C₁₄ 1%; average alkyl chain length = C_{11.9}

Remarks: Primary Irritation Score: 6.2
Intact Skin: 6.8/8
Abraded Skin: 6.8/8
Slight to moderate erythema and edema were present after 24 hours. These symptoms persisted through 120 hours. The compound was classed as a primary skin irritant under the grading system as outlined in the Federal Hazardous Substance Act. The sample had a defatting effect on the skin causing the skin to slough off in ten to fourteen days. There was no injury in depth.

Reference: Monsanto Company. 1971. Linear alkylbenzene sodium sulfonate – Alkylate 225 Lot CC 6450 – Acute toxicity screen. Project No. Y-71-119. Unpublished report.

Reliability: 2 Valid with restrictions

(i)

Species/strain: New Zealand white male and female rabbits

Results: Highly corrosive ; Corrosive ; Highly irritating ; Irritating ; Moderate irritating ; Slightly irritating ; Not irritating

Classification: Highly corrosive (causes severe burns) [];
Corrosive (causes burns) []; Irritating [X]; Not irritating []

Method: The clipped, intact and abraded skin of three New Zealand white male and female rabbits was applied with 0.5 g of finely ground sample moistened with water under a one inch by one inch square patch, two single layers thick. The patches were positioned in place with adhesive tape. The trunk of each animal was wrapped with plastic strips to avoid contamination and retard evaporation for the 24 hour exposure period. Observations were made over a period of seven days for irritation. The data was scored according to the method of Draize, Woodard and Calvery (Journal of Pharm. and Exp. Therapeutics, Volume 82, December 1944).

GLP: Yes [] No [X] ? []

Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-215); C₉ 1%, C₁₀ 16%, C₁₁ 43%, C₁₂ 40%, C₁₃ 1%, C₁₄ <1%; average alkyl chain length = C_{11.35}

Remarks: Moderate to severe erythema and edema were present after 24 hours. These symptoms persisted through 120 hours. The average maximum score was 7.3 out of a possible 8 at 24 hours. The compound was classed as a severe skin irritant. The sample had a defatting effect on the skin causing the skin to slough off in ten to fourteen days. There was no injury in depth.

Reference: Monsanto Company. 1972a. Linear alkylbenzene sodium sulfonate – Alkylate 215 Lot CC 6772S – Acute toxicity screen. Project No. Y-72-274. Unpublished report.

Reliability: 2 Valid with restrictions

(j)

Species/strain: New Zealand white male and female rabbits

Results: Highly corrosive []; Corrosive []; Highly irritating [X]; Irritating []; Moderate irritating []; Slightly irritating []; Not irritating []

Classification: Highly corrosive (causes severe burns) [];
Corrosive (causes burns) []; Irritating [X]; Not irritating []

Method: The clipped, intact and abraded skin of three New Zealand white male and female rabbits was applied with 0.5 g of finely ground sample moistened with water under a one inch by one inch square patch, two single layers thick. The patches were positioned in place with adhesive tape. The trunk of each animal was wrapped with plastic strips to avoid contamination and retard evaporation for the 24 hour exposure period. Observations were made over a period of seven days for irritation. The data was scored according to the method of Draize, Woodard and Calvery (Journal of Pharm. and Exp. Therapeutics, Volume 82, December 1944).

GLP: Yes [] No [X] ? []

Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-222L); average alkyl chain length = C_{11.5}

Remarks: Moderate to severe erythema and edema were present after 24 hours. These symptoms persisted through 120 hours. The average maximum score was 7.0 out of a possible 8 at 24 hours. The compound was classed as a severe skin irritant. The sample had a defatting effect on the skin causing the skin to slough off in ten to fourteen days. There was no injury in depth.

Reference: Monsanto Company. 1972b. Linear alkylbenzene sodium sulfonate – Alkylate 222L Lot CC 6773S – Acute toxicity screen. Project No. Y-72-275. Unpublished report.
Reliability: 2 Valid with restrictions

5.2.2 EYE IRRITATION/CORROSION

(a)
Species/strain: New Zealand albino rabbits
Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating [X]; Moderate irritating []; Slightly irritating []; Not irritating []
Method: OECD Guideline 405. Nine rabbits received a 0.1 mL aliquot of P-500 N-Na placed into the lower everted lid of one eye per animal. For three rabbits the eyelids were then gently held together for one second before releasing. For three other rabbits the eyes were irrigated with water for 5 minutes following a 4-second exposure. For the remaining three rabbits the eyes were irrigated for 5 minutes following a 30-second exposure. Eyes were examined after 1 hour and 1, 2, 3, 4, 7, 14 and 21 days after exposure. Grading was performed using the standard scoring system.
GLP: Yes [X] No [] ? []
Test substance: Alkylbenzene sulfonate, sodium salt (designated as P-500 N-Na). Activity 47%. Average alkyl chain length = C_{11.2}. Clear yellow liquid.
Remarks: The following results were noted:
1) Three animals without any rinsing: averaged irritation scores (24, 48, 72 hours) for each animal: cornea 2.3, 1.7, 2; iris: 1.3, 0, 0; conjunctivae redness: 3, 1.7, 2; conjunctivae chemosis: 3, 2, 2. In the first animal the effects were persistent at day 14.
2) Three animals with rinsing for five minutes following a 30 second exposure: averaged scores: cornea 0.7, 1, 1.3; iris: 0, 0.7, 0.3; conjunctivae redness: 1.7, 2, 1.3; conjunctivae chemosis: 2, 1.3, 2. The eyes were normal 7 or 14 days after instillation.
3) Three animals with rinsing for five minutes following a 4 second exposure: averaged scores: cornea 0.7, 2.3, 0.7; iris: 0, 0, 0; conjunctivae redness: 1.7, 1.7, 1; conjunctivae chemosis: 1.3, 2, 1. The eyes were normal 7 days after instillation.
Findings lead to a definition of irritating for LAS at 47% applied without rinsing, irritating (even if with lower effects, mainly as cornea opacity and conjunctivae redness) with rinsing after 30 second of exposure, and not irritating with rinsing after 4 second of exposure.
Overall, instillation of P-500 N-Na into the eyes of rabbits elicited positive responses in all animals. Irrigation of the eyes only slightly reduced the irritation potential.
Reference: Liggett, M.P. and Parcell, B.I. 1986b. Irritant effects on the rabbit eye of P-500 N-Na. Huntingdon Research Ctr Report No. 86570D/PEQ 10/SE.
Reliability: 1 Valid without restriction

(b)
Species/strain: New Zealand albino rabbits
Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating [X]; Moderate irritating []; Slightly irritating []

Method: Not irritating []
OECD Guideline 405. Six rabbits were exposed to 0.1 ml of 50% LAS, which was placed into the conjunctival sac of one eye per animal. The eyelids were gently held together for one second and then released. No irrigation step was performed. The eyes were examined at 1, 24, 48 and 72 hours and at 7 days and scored for irritation using the standard system.

GLP: Yes [] No [X] ? []

Test substance: LAS (made from Sirene LAB); activity 50%; average alkyl chain length = C_{11.6}

Remarks: Average irritation scores were 1.3, 1.0, 2.6, and 2.7 for the cornea, iris, conjunctival redness, and conjunctival chemosis, respectively. Effects were persistent to Day 6. This classifies LAS at 50% as irritating.

Reference: Biolab SGS. 1989d. Acute eye irritation. Report No. 00428/13.

Reliability: 2 Valid with restrictions

(c)

Species/strain: rabbit

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating [X]; Moderate irritating []; Slightly irritating []; Not irritating []

Classification: Irritating [X]; Not irritating []; Risk of serious damage to eyes []

Method: OECD Guide-line 405 "Acute Eye Irritation/Corrosion" 1981

GLP: Yes [] No [X] ? []

Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}; activity: 50%.

Remarks: The mean irritation index was 26.5 out of 110. Individual scores: 1.0; iris: 0; conjunctivae chemosis: 1.11, conjunctivae redness: 2.39

Reference: Murmann, P. 1983b. Prufung der akuten Augen-und Schleimhautreiz Wirkung von Marlon A 350. Huels Report No. 0172.

Reliability: 2 Valid with restrictions

(d)

Species/strain: rabbit

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating [X]; Moderate irritating []; Slightly irritating []; Not irritating []

Classification: Irritating [X]; Not irritating []; Risk of serious damage to eyes []

Method: OECD Guide-line 405 "Acute Eye Irritation/Corrosion" 1984

GLP: Yes [X] No [] ? []

Test substance: LAS

Remarks: Possibility of persistent effects on the eye.

Reference: Kaestner, W., Henkel KGaA, unpublished data, Report No. 870553 (1987).

Reliability: 4 Not assignable

(e)

Species/strain: rabbit

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating [X]; Moderate irritating []; Slightly irritating []; Not irritating []

Classification: Irritating [X]; Not irritating []; Risk of serious damage to eyes []

Method: 0.1 mL solutions of LAS at 5 different concentrations ranging from 0.01 to 1.0 % were instilled in the eyes of rabbits (13 per group). The rabbits were observed for 24 hours after LAS application.

GLP: Yes No ?

Test substance: C₁₀₋₁₃ LAS, (CAS #68411-30-3). Molecular weight 346.5; average alkyl chain length = C_{11.9}

Remarks: Information as cited in IPCS document. The 0.01 % group showed no abnormalities, but the 0.05 % group showed slight congestion. The groups of 0.5 % and higher concentrations showed marked reactions such as severe congestion and oedema, increased secretion, turbidity of the cornea, and disappearance of corneal reflex.

Reference: Oba, K., Mori, A. and Tomiyama, S. 1968. Biochemical studies of n-alpha-olefin sulfonates (II) Acute toxicity, skin and eye irritation, and some other physical properties. Journ. Jap. Oil Chem. Soc. 17:628-634. (In Japanese) cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(f)

Species/strain: rabbit

Results: Highly corrosive ; Corrosive ; Highly irritating ; Irritating ; Moderate irritating ; Slightly irritating ; Not irritating

Classification: Irritating ; Not irritating ; Risk of serious damage to eyes

Method: LAS solutions at 6 different concentrations ranging from 0.01% to 5.0% were instilled in the eyes of rabbits (3 per group). The rabbits were observed for 168 hours after LAS application.

GLP: Yes No ?

Test substance: LAS (chain length distribution C₁₀₋₁₄; 80.9% of C₁₁₋₁₃) (CAS #69669-44-9); average alkyl chain length (based on LAS SIDS Consortium Survey, 2000) = C_{11.7}

Remarks: Information as cited in IPCS document. The 0.01% group showed no reaction. Within 2 hours, the 0.05% group showed slight congestion and the 0.1% group showed considerable congestion or oedema, which disappeared at 24 hours. In the group of 0.5% and higher, marked reactions such as severe congestion an oedema, increased secretion, turbidity of the cornea, and disappearance of the corneal reflex were observed for 24 hours. The effects disappeared completely at 120 hours.

Reference: Iimori, M., Ogata, T. and Kudo, K. 1972. Eye irritation testing of surface active agents in experimental animals. Jour. Jap. Oil Chem. Soc. 22:807-813 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(g)

Species/strain: New Zealand albino rabbits

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating []; Moderate irritating []; Slightly irritating []; Not irritating [X]

Method: OECD Guideline 405. Six rabbits were exposed to 0.1 ml of 1% LAS, which was placed into the conjunctival sac of one eye per animal. The eyelids were gently held together for one second and then released. No irrigation step was performed. The eyes were examined at 1, 24, 48 and 72 hours and at 7 days and scored for irritation using the standard system.

GLP: Yes [] No [X] ? []

Test substance: LAS (made from Sirene LAB); activity 1%; average alkyl chain length = C_{11.6}

Remarks: Average irritation scores were 0, 0, 0.1, and 0.1 for the cornea, iris, conjunctival redness, and conjunctival chemosis, respectively. This classifies LAS at 1% as not irritating.

Reference: Biolab SGS. 1984. Acute eye irritation. Report No. T3R/27.

Reliability: 2 Valid with restrictions

(h)

Species/strain: New Zealand albino rabbits

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating []; Moderate irritating [X]; Slightly irritating []; Not irritating []

Methods: OECD Guideline 405. Six rabbits were exposed to 0.1 ml of 5% LAS, which was placed into the conjunctival sac of one eye per animal. The eyelids were gently held together for one second and then released. No irrigation step was performed. The eyes were examined at 1, 24, 48 and 72 hours and at 7 days and scored for irritation using the standard system.

GLP: Yes [] No [X] ? []

Test substance: LAS (made from Sirene LAB); activity 5%; average alkyl chain length = C_{11.6}

Remarks: Average irritation scores were 0, 0, 1.83, and 1.16 for the cornea, iris, conjunctival redness, and conjunctival chemosis, respectively. This classifies LAS at 5% as moderately irritating.

Reference: Biolab SGS. 1988. Acute eye irritation. Report No. T343.

Reliability: 2 Valid with restrictions

(i)

Species/strain: albino male and female rabbits

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating []; Moderate irritating [X]; Slightly irritating []; Not irritating []

Classification: Irritating [X]; Not irritating []; Risk of serious damage to eyes []

Method: A total of 100 mg of finely ground sample was administered in the conjunctival sac of the right eye of each of six albino male and female rabbits. Observations for inflammation were made over seven days. The treated eyes were washed with sodium chloride solution USP after the 24 hour reading. The left eye served as a control. The data was scored according to the method of Draize, et al. Tests were conducted in accordance with the Federal Hazardous Substance Act.

GLP: Yes [] No [X] ? []

Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-225) C₉ 1%, C₁₀ 7%, C₁₁ 25%, C₁₂ 48%, C₁₃ 19%, C₁₄ 1%; average alkyl chain length = C_{11.9}

Remarks: The compound is classified as an eye irritant under the grading system as outlined in the Federal Hazardous Substances Act. Slight to moderate erythema was present after 10 minutes and persisted through 72 hours. Edema was present after 1 hour and persisted through 24 hours. The average maximum irritation score was 19.3 out of a possible 110.

Reference: Monsanto Company. 1971. Linear alkylbenzene sodium sulfonate – Alkylate 225 Lot CC 6450 – Acute toxicity screen. Project No. Y-71-119. Unpublished report.

Reliability: 2 Valid with restrictions

(j)

Species/strain: albino male and female rabbits

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating []; Moderate irritating []; Slightly irritating [X]; Not irritating []

Classification: Irritating [X]; Not irritating []; Risk of serious damage to eyes []

Method: A total of 100 mg of finely ground sample was administered in the conjunctival sac of the right eye of each of three albino male and female rabbits. Observations for inflammation were made over seven days. The treated eyes were washed with isotonic saline solution after the 24 hour reading. The left eye served as a control. The data was scored according to the method of Draize, et al. Tests were conducted in accordance with the Federal Hazardous Substance Act.

GLP: Yes [] No [X] ? []

Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-215) C₉ 1%, C₁₀ 16%, C₁₁ 43%, C₁₂ 40%, C₁₃ 1%, C₁₄ <1%; average alkyl chain length = C_{11.35}

Remarks: Slight erythema and a copious discharge were present 10 minutes after application. A slight erythema persisted for 72 hours after which the eyes returned to normal. The average maximum score was 10 out of a possible 110 after 24 hours. This compound is classified as a slight eye irritant.

Reference: Monsanto Company. 1972a. Linear alkylbenzene sodium sulfonate – Alkylate 215 Lot CC 6772S – Acute toxicity screen. Project No. Y-72-274. Unpublished report.

Reliability: 2 Valid with restrictions

(k)

Species/strain: albino male and female rabbits

Results: Highly corrosive []; Corrosive []; Highly irritating []; Irritating []; Moderate irritating []; Slightly irritating [X]; Not irritating []

Classification: Irritating [X]; Not irritating []; Risk of serious damage to eyes []

Method: A total of 100 mg of finely ground sample was administered in the conjunctival sac of the right eye of each of three albino male and female rabbits. Observations for inflammation were made over seven days. The treated eyes were washed with isotonic saline solution after the 24 hour reading. The left eye served as a control. The data was scored according to the method of Draize, et al. Tests were conducted in accordance with the Federal Hazardous Substance Act.

GLP: Yes [] No [X] ? []

Test substance: Sodium sulfonate of linear alkylbenzene (Alkylate-222L; average alkyl chain length = C_{11.5})

Remarks: Moderate erythema and slight edema and a copious discharge were present 10 minutes after application. A slight erythema persisted for 72 hours after which the eyes returned to normal. The average maximum score was 18 out of a possible 110 after 24 hours. This compound is classified as a mild eye irritant.

Reference: Monsanto Company. 1972b. Linear alkylbenzene sodium sulfonate – Alkylate 222L Lot CC 6773S – Acute toxicity screen. Project No. Y-72-275. Unpublished report.

Reliability: 2 Valid with restrictions

5.3 SKIN SENSITIZATION

(a)

Type: Sensitisation test

Species/strain: Guinea pig (males & females, albino)

Results: Sensitising []; Not sensitising [X]; Ambiguous []

Classification: Sensitising []; Not sensitising [X]

Method: Buehler Test; OECD Guideline 406 "Skin Sensitization" 1981; Directive 84/449, EEC B.6. "Acute Toxicity - Skin Sensitization".

GLP: Yes [X] No [] ? []

Test Substance: LAS; activity: 6.7%; average alkyl chain length = C_{11.6}

Remarks: The purpose of this study was to assess the allergenic potential of LAS when administered to the skin. 10 animals (5M/5F) remained untreated and were used as controls to be treated at first challenge. 10 animals (5M/5F) remained untreated and were used as additional controls to be treated at second challenge; 20 animals (10M/10F) were treated with LAS. Induction concentration was 1.0% in water; first and second challenge concentrations were 0.8% in water. 0/20 animals responded in the treated group; 0/10 animals responded in the control group.

Reference: European Commission. 2000o. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition, citing The Procter & Gamble Company, unpublished data, Reports No. RCC-2315547.

Reliability: 4 Not assignable

(b)

Type: Guinea pig maximisation test

Species/strain: guinea pig, females

Results: Sensitising []; Not sensitising [X]; Ambiguous []

Classification: Sensitising []; Not sensitising [X]

Method: OECD Guide-line 406 "Skin Sensitisation" 1981

GLP: Yes [] No [X] ? []

Test substance: Marlon A 350 (CAS #68411-30-3) C₁₀₋₁₃ LAS, average alkyl chain length = C_{11.6}; activity: 50%.

Remarks: 0.1% intracutaneous and 3% epidermal doses. No sensitizing effects were observed.

Reference: Murmann, P. 1988. Prufung auf hautsensibilisierende Wirkung am Meerschweinchen von Marlon A 350. Huels Report No. 1387.

Reliability: 2 Valid with restrictions

(c)

Type: Maximization test

Species/strain: Guinea pig, Hartley
 Results: Sensitising []; Not sensitising [X]; Ambiguous []
 Classification: Sensitising []; Not sensitising [X]
 Method: OECD Guideline 406, 1981; Directive 179/831 Annex, Part B.
 GLP: Yes [X] No [] ? []
 Test substance: LAS, activity: 50%; average alkyl chain length = C_{11,6}
 Remarks: Solutions of LAS were applied intracutaneously and epicutaneously to 10 male and 10 female animals. Induction concentration was 25% in water; the challenge concentration was 12.5%. No positive responses were observed.
 Reference: RBM. 1985. Test di sensibilizzazione cutanea nella cavia. RBM Exp. No. 2076.
 Reliability: 1 Valid without restriction

5.4 REPEATED DOSE TOXICITY

(a)
 Species/strain: Rat (FDRL)
 Sex: Female []; Male []; Male/Female [X]; No data []
 Administration: Oral feed
 Exposure period: 12 weeks
 Dose: 50 or 250 mg/kg bw d
 Control group: Yes [X]; No []; No data [];
 Concurrent no treatment [X]; Concurrent vehicle []; Historical []
 NOAEL: 50 mg/kg bw d
 LOAEL: 250 mg/kg bw d
 Results: No behavioural abnormalities were noted during the test period. Growth responses were equal in all groups. There were no differences in food intake or in efficiency of food utilization. The clinical data showed no abnormal variations in any of the dose groups. The relative organ weights and the histopathological evaluation did not show significant differences among the dose groups except a liver weight increase in female animals of the high dose group.
 Method: Based on Fitzhugh and Schouboe (1959) Subacute toxicity in: Assoc. Food Drug Offices of the U.S., Austin, Texas, p. 26-35. Weanling rats were distributed into 5 groups of 15 male and 15 female animals per dose group. All rats were given standard diet daily. Doses were 0, 50 and 250 mg/kg bw d in the diet. Daily observations of behavior and signs of toxicity were made. Food consumption and blood and urine chemistries were also measured periodically. Organ weights and gross pathological findings were measured at the end of the study in the liver, kidneys, spleen, heart, adrenals, pituitary, and cecum.
 GLP: Yes [] No [X] ? []
 Test substance: C₁₀₋₁₃ LAS, sodium salt, activity: 39.5%; average molecular weight 346; average alkyl chain length = 11.9
 Reference: Oser, B.L. and Morgareidge, K. 1965. Toxicological studies with branched and linear alkyl benzene sulfonates in rats. Toxicol. Appl. Pharmacol. 7:819-825.
 Reliability: 2 Valid with restrictions

(b)

Species/strain: rat (Sprague-Dawley)
Sex: Female []; Male []; Male/Female [X]; No data []
Administration: oral feed
Exposure period: 90 days
Frequency of treatment: *Ad libitum*
Dose: 0.02/0.1/0.5% (corresponding to 8.8, 44 and 220 mg/kg bw d)
Control group: Yes [X]; No []; No data [];
Concurrent no treatment [X]; Concurrent vehicle []; Historical []
NOAEL: 0.5% (220 mg/kg bw d)
Results: No adverse effects were found upon the following parameters: growth, food efficiency, survival, haematologic values, and urinary analytical values. In addition, no treatment-related adverse effects were observed in absolute and relative organ weights, nor were gross histopathological changes observed in any of the organs examined.
Method: Groups of 10 male and 10 female weanling rats were fed levels of 0.02, 0.1 or 0.5% LAS in a commercial chow for 90 days. Body weights, food consumption, mortality, and several blood parameters were measured periodically during the study and at termination. Autopsy and microscopic examination of the organs was performed at test termination. Organ weights and gross pathological findings were recorded for the liver, kidneys, spleen, gonads, heart, and brain.
GLP: Yes [] No [X] ? []
Test substance: C₁₀₋₁₄ LAS, sodium salt (CAS #69669-44-9), activity: 87.9%; C₁₀ 1.8%, C₁₁ 43.2%, C₁₂ 32.2%, C₁₃ 16.0%, C₁₄ 5.3%, C₁₅ 1.5%; average alkyl chain length = C_{11.8}; mean molecular weight 346.
Remarks: Two male rats at the 0.2% level died in the early stages of the study. These deaths were attributed to respiratory illness and were not considered to be treatment related.
Reference: Kay, J.H., Kohn, F.E. and Calandra, J.C. 1965. Subacute oral toxicity of a biodegradable linear alkylbenzene sulfonate. *Toxicol. Appl. Pharmacol.* 7:812-818.
Reliability: 2 Valid with restrictions

(c)

Species/strain: Rat/Sprague-Dawley
Sex: Female []; Male []; Male/Female [X]; No data []
Administration: gavage
Exposure period: one month
Frequency of treatment: daily
Dose: 125, 250, 500 mg/kg bw d.
Control group: Yes [X]; No []; No data [];
Concurrent no treatment [X]; Concurrent vehicle []; Historical []
NOAEL: 125 mg/kg bw d
LOAEL: 250 mg/kg bw d
Results: Diarrhea was observed in the 500 mg/kg group and soft stools were observed in the other 2 groups. Body weight gain was suppressed in all the male groups and in the female 500 mg/kg group. Haematological examinations revealed no abnormalities. Serum-biochemical examinations revealed several differences among the mid and high dose group compared to the control group. The weight of the spleen and the heart significantly decreased in the male high dose group. In the female high dose group, the weight of the liver increased while the weight of the

heart and thymus decreased. Histological findings of the liver revealed no abnormalities.

GLP: Yes No ?

Remarks: Information as cited in IPCS document. 12 animals per dose group.

Test substance: C₁₀₋₁₃ LAS, sodium salt (CAS #68411-30-3)
<C₁₀ 0.1%, C₁₀ 10.1%, C₁₁ 33.7%, C₁₂ 31.0%, C₁₃ 25.1%; average alkyl chain length = C_{11.7}; activity: 99.5%

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Ito, R., Kawamura, H., Chang, H.S., Kudo, K., Kajiwara, S., Toida, S., Seki, Y., Hashimoto, M. and Fukushima, A. 1978. J. Med. Soc. Toho, Japan, 25:850-875 (in Japanese). cited in IPCS. 1996. Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates and Related Compounds. World Health Organization, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(d)

Species/strain: Rat (Wistar)

Sex: Female ; Male ; Male/Female ; No data

Administration: oral feed

Exposure period: 9 months

Frequency of treatment: Daily in feed

Dose: 0.6% and 1.8% (260 and 780 mg/kg bw d)

Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical

LOAEL: = 0.6% (260 mg/kg bw d)

Results: In the 1.8% dose group, the body weight gain was suppressed and haematological and serum-biochemical adverse effects were observed in both treatment groups of both sexes. The weight of the cecum of the male rats and the weight of the liver and cecum of the females in the high dose groups were significantly increased. Enzymatic examinations of the liver and kidneys revealed changes in different enzyme activities in the 1.8% groups. The intake of LAS was 230 mg/kg bw d in the male 0.6% group and 290 mg/kg bw d in the female 0.6% group.

GLP: Yes No ?

Test substance: C₁₀₋₁₄ LAS, sodium salt (CAS #69669-44-9) C₁₀ 10.6%, C₁₁ 34.1%, C₁₂ 27.7%, C₁₃ 19.0%, C₁₄ 8.7%; average alkyl chain length = C_{11.8}; mean molecular weight: 345.8.

Remarks: Information as cited in IPCS document. 8 rats were used per dose group.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Yoneyama, M., Mabuchi, Y., Ikawa, M., Kobayashi, H. and Ichikawa, H. 1976. Subacute toxicity of linear alkyl benzene sulfonate. Ann. Rep. Tokyo Metr. Res. Lab. P.H. 27(2): 105-112 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(e)

Species/strain: Rat (Wistar)
Sex: Female []; Male []; Male/Female []; No data []
Administration: oral feed
Exposure period: 2, 4, 12 weeks
Frequency of treatment: Daily in feed
Dose: 1.5% (750 mg/kg bw d)
Control group: Yes []; No []; No data [];
Concurrent no treatment []; Concurrent vehicle []; Historical []
LOAEL: 1.5% (750 mg/kg bw d)
Results: LAS suppressed body weight gain, and the relative liver weight was increased from 2 weeks of LAS administration. Serum biochemical examinations revealed significant increases in ALP and GTP at each observation period and significant decreases in cholesterol and protein in 4 weeks. Enzymatic examinations of the liver revealed decreases in G6Pase and G6PDH and an increase in isocitrate dehydrogenase (IDH) at each observation period. Enzymatic examinations of the renal cortex revealed decreases in G6Pase and 5'-nucleotidase at each observation period, an increase in LDH at 12 weeks, and an increase in IDH at 2 and 4 weeks. Enzymatic examinations in the renal medulla revealed a decrease in NA,K-ATPase, an increase in LDH at 12 weeks, a decrease in IDH at 2 weeks, and an increase in IDH at 12 weeks.

GLP: Yes [] No [] ? []

Test substance: LAS (unspecified)

Remarks: Information as cited in IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Ikawa, M., Yoneyama, M., Nakao, T. and Hiraga, K. 1978. Uptake of organic acid and organic base by renal cortical slices of rats treated with LAS and ABS. Ann. Rep. Tokyo Metr. Res. Lab. P.H. 29:51-54 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(f)

Species/strain: Rat (Wistar)
Sex: Female []; Male []; Male/Female []; No data []
Administration: oral feed
Exposure period: 6 months
Frequency of treatment: Daily in feed
Dose: 0.07, 0.2, 0.6, 1.8% (40, 115, 340, 1030 mg/kg bw d)
Control group: Yes []; No []; No data [];
Concurrent no treatment []; Concurrent vehicle []; Historical []
NOAEL: = 40 mg/kg bw d
LOEL: = 115 mg/kg bw d

Results: The 1.8% group showed diarrhoea, markedly suppressed growth, increased weight of the cecum, and remarkable degeneration of the renal tubes. The 0.6% group showed slightly suppressed growth, increased weight of the cecum, increased activity of serum alkaline phosphatase (ALP), a decrease in serum protein and degeneration of the renal tubes. The 0.2% group showed increased weight of the cecum and slight degeneration of the renal tubes, the 0.07% group showed no adverse effects related to the administration of LAS. The intake of LAS in the 0.07% group was about 40 mg/kg bw d.

GLP: Yes No ?

Test substance: C₁₀₋₁₄ LAS, sodium salt (CAS #69669-44-9). C₁₀ 10.6%, C₁₁ 34.1%, C₁₂ 27.7%, C₁₃ 19.0%, C₁₄ 8.7%; average alkyl chain length = C_{11.8}; mean molecular weight: 345.8.

Remarks: Information as cited in IPCS document. This is a key study for repeated dose toxicity because it represents the lowest LOAEL (see SIAR Table 6).

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Yoneyama, M. Fujii, T., Ikawa, M., Shiba, H., Sakamoto, Y., Yano, N., Kobayashi, H., Ichikawa, H. and Hiraga, K. 1972. Studies on the toxicity of synthetic detergents. (II) Subacute toxicity of linear and branched alkyl benzene sulfonates in rats. Ann. Rep. Tokyo Metrap. Res. Lab. Public Health. 24:409-440. (In Japanese). cited in IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(g)

Species/strain: Rat

Sex: Female ; Male ; Male/Female ; No data

Administration: gavage

Exposure period: 10 weeks

Doses: 50, 100 or 250 mg/kg bw d

Control: Yes ; No ; No data ; Concurrent no treatment ; Concurrent vehicle ; Historical

LOEL: 50 mg/kg bw d

Results: Histopathology was evaluated in the females only. At the highest dose level, the kidneys showed mild degeneration and desquamation of the tubular epithelium and there was a moderate degree of fatty change in the liver as well as proteinaceous degeneration. Sections of the intestine did not exhibit any significant histologic variation. Adenosine triphosphatase activity was inhibited with increasing dose in both sexes while alkaline phosphatase and acid phosphatase activities were increased with increasing dose. The activity of lactate dehydrogenase was significantly inhibited at all dose levels in females but was not measured in males. SGOT and SGPT were significantly decreased in females at 100 and 250 mg/kg/day and SGPT was inhibited in males at 250 mg/kg/day.

Method: Twenty four male (50+/-5g) and 36 female (40+/-5g) rats were given daily oral (cannula) doses of LAS detergent solution (0, 50, 100 and 250 mg/kg) by gavage for 10 weeks. Animals were maintained on standard pellet diets and drinking water *ad libitum*. After 10 weeks, animals were

fasted for 24 hours and sacrificed. Liver, kidney, heart, and intestine were removed immediately, weighed, and parts sectioned for histological examination. The remaining parts of the liver and kidney homogenized in ice cold 0.25 M sucrose solution using Potter-Elvehjem type homogenizer and 10% w/v homogenates were prepared for histopathology and enzyme analysis. The activities of adenosine triphosphatase (ATPase), acid phosphatase (ACP), alkaline phosphatase (ALP), glutamic oxaloacetic transaminase (GOT), and glutamic pyruvic transaminase (GPT) were determined in the homogenates.

GLP: Yes No ?

Remarks: All of the effects observed are related to energy metabolism, so given the lack of morphological and structural changes, the reduced food intake and body weight gain may have compromised energy dynamics and affected the enzyme levels. The reliability and use of this study for risk assessment purposes is limited and the 50 mg/kg/day level should be considered a LOEL rather than a LOAEL.

Test Substance: Commercial LAS synthetic detergent solution

Reference: 1) Gupta, B.N., Mathur, A.K., Agarwal, C., and Singh, A. 1986. Effect of synthetic detergent on certain enzymes in liver and kidney in male rats. *Arogya-J. Health Sci.* 12:50-54.
2) Mathur, A.K., Gupta, B.N., Singh, A., and Shanker, R. 1986. Toxicological evaluation of a synthetic detergent after repeated oral ingestion in rats. *Biol. Mem.* 12:187-191.

Reliability: 2 Valid with restrictions

(h)

Species/strain: Rat (Wistar)

Sex: Female ; Male ; Male/Female ; No data

Administration: drinking water

Exposure period: 9 months

Frequency of treatment: Daily in drinking water.

Dose: 0.07, 0.2%, 0.6% (85, 145, 430 mg/kg bw d; average of male and female)

Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL: 0.07% (85 mg/kg bw d)

LOAEL: 0.2% (145 mg/kg bw d)

Results: Body weight gain was suppressed in the male 0.6% group. Hematological examination revealed no significant change in any of the experimental groups, but a dose-related decrease in cholesterol level was seen in males. Significant decreases in the activities of glutamate-oxalate transaminase and lactate dehydrogenase were seen in males at 0.2% and a dose-related increase in the activity of glutamate-oxalate transaminase in females. A significant decrease in renal Na,K-ATPase was seen in the group given 0.2%. No organ weight changes were observed. The intake of LAS was 50 mg/kg bw d in the male 0.07% group and 120 mg/kg bw d in the female group. The values for the 0.2% group were 120 and 170 mg/kg bw d for males and females, respectively.

Method: Groups of 8-9 male and 8-9 female rats were given LAS for 9 months.

GLP: Yes No ?

Remarks: Information as cited in IPCS document. This is a key study for repeated dose toxicity because it represents the highest NOAEL below the lowest LOAEL (see SIAR Table 6).

Test substance: C₁₀₋₁₄ LAS (CAS #69669-44-9); average alkyl chain length (based on LAS SIDS Consortium Survey, 2000) = C_{11.7}

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Yoneyama, M., Mabuchi, Y., Ikawa, M., Kobayashi, H. and Ichikawa, H. 1976. Subacute toxicity of linear alkylbenzene sulfonate. Ann. Rep. Tokyo Metr. Res. Lab. Public Health 27:105-112 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(i)

Species/strain: mouse (DDY)

Sex: Female ; Male ; Male/Female ; No data

Administration: drinking water

Exposure period: 6 months

Frequency of treatment: Daily

Observation period: 2 months post exposure

Dose: 100 ppm in the drinking water (20 mg/kg bw d)

Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical

Results: Atrophy of the golgi apparatus, degeneration of the mitochondria, and increased appearance of lysosomes were observed. The severity of these adverse effects were dependent on the length of the administration. After six months, some cells showed degenerative cytoplasm and indications of cell necrosis. Effects on the rough endoplasmic reticulum were observed. Some animals still showed cellular effects after the two months post administration period while other animals showed full recovery. Given the unknown significance of the effects for the health of the animals and the reversibility of the effects, as well as the consistent lack of adverse effects in other studies at similar or higher doses, the dose tested in this study is considered a LOEL rather than a LOAEL.

Method: LAS was administered up to 6 months. The animals were sacrificed at 1, 2, 3, and 6 months. Some animals were observed an additional 2 months without test substance administration. Liver slices were investigated using electron microscopy.

GLP: Yes No ?

Remarks: The reliability and usefulness of this study for risk assessment purposes is limited. The study employed only a single dose (i.e., no dose response information). In addition, there is the likelihood of dehydration of the animals. Because of these study deficiencies, it was determined that it is inappropriate to derive a LOAEL from this study.

Test substance: LAS (unspecified)

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Watari, N., Torizawa, K., Kanai, M. and Suzuki, Y. 1977. Ultrastructural observations of the protective effect of glycyrrhizin for mouse liver injury caused by oral administration of detergent ingredient (LAS) J. Clin. Electron. Microscopy 10:121-139 (in Japanese) cited in IPCS. 1996. Environmental Health Criteria 169: Linear Alkylbenzene

Sulfonates and Related Compounds. World Health Organization, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. While the study was evaluated by IPCS prior to inclusion in their criteria document, the deficiencies noted above make its usefulness in risk assessment questionable.

(j)

Species/strain: mouse (ICR)
Sex: Female []; Male []; Male/Female [X]; No data []
Administration: Oral feed or water
Exposure period: 9 months
Frequency of treatment: Daily
Dose: Diet: 0.6 and 1.8% (corresponding to 500 and 1000 mg/kg bw d).
Diet: Drinking water: 0.07, 0.2, and 0.6% (100, 250, and 600 mg/kg bw d for males and 100, 250, and 900 mg/kg bw d for females)
Control group: Yes [X]; No []; No data [];
Concurrent no treatment [X]; Concurrent vehicle []; Historical []
NOAEL: 250 mg/kg bw d in drinking water
LOAEL: 500 mg/kg bw d in diet
Results: In the mice given 0.6% in their diet, body weight gain was not suppressed, but the weight of the liver increased in male and female mice. Enzymatic examinations revealed significant decreases in LDH of the liver and in acid phosphatase of the kidneys in the male mice. For mice given LAS in drinking water, body weight was depressed at the highest dose for males and females. This dose also elicited an increase in liver weight in females and significant decreases in renal Na and K-ATPase.
Method: Groups of 8 or 9 mice were given diets containing LAS at concentrations of 0.6 and 1.8% or drinking water containing LAS at concentrations of 0.07, 0.2, and 0.6% for 9 months.
GLP: Yes [] No [X] ? []
Test substance: LAS
Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Yoneyama, M., Mabuchi, Y., Ikawa, M., Kobayashi, H. and Ichikawa, H. 1976. Subacute toxicity of linear alkylbenzene sulfonate. Ann. Rep. Tokyo Metr. Res. Lab. P.H. 27(2):105-112 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.
3) HERA. 2002. HERA-LAS Human and Environmental Risk Assessment: Linear Alkylbenzene Sulphonates, LAS. CAS No. 68411-30-3, Draft #6, May 2002.
Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(k)

Species/strain: Rat: Charles River
Sex: Female []; Male []; Male/Female [X]; No data []
Administration: oral feed
Exposure period: 2 years
Frequency of treatment: continuous in feed

Doses: 0.02, 0.1, 0.5% (10, 50, 250 mg/kg bw d)
Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical
Results: Gross examination of all animals for pathology did not reveal any abnormalities. No consistent dietary induced changes that could be considered a toxic response were observed. Animals that showed significant loss of weight, development of tumors, or other evidence of abnormalities were sacrificed and tissues examined. The incidence of tumors and the common incidental diseases were similar in all dieting groups. No treatment-related adverse histological effects were observed in any of the tissue sections examined.
Method: Four groups of Charles River weanling rats, divided by sex, were given 0.5, 0.1, and 0.02% LAS in their food for 2 years. Following completion of those studies, five male and five female rats from each of the parental groups (F_{1b} and F_{2b}) and all survivors were selected for necropsy. Livers and kidneys were removed and weighed. Body weight and organ to body weight ratios were recorded, and routine hematology and histology were performed. Sections for histological examination were taken from the liver, kidney, thyroid, trachea, esophagus, lung, heart, spleen, pancreas, adrenals, stomach, small intestine, urinary bladder, gonads and mesenteric lymph nodes. Weanling animals for the F_{3a} generation were similarly treated.
GLP: Yes No ?
Test substance: C₁₀₋₁₄ LAS, sodium salt; activity: 98.1% on an anhydrous basis (41.9% active)
Reference: Buehler, E.V., Newmann, E.A., and King, W.R. 1971. Two year feeding and reproduction study in rats with linear alkylbenzene sulfonate (LAS). Toxicol. Appl. Pharmacol. 18:83-91.
Reliability: 2 Valid with restrictions

(1)
Species/strain: Rat/Wistar
Sex: Female ; Male ; Male/Female ; No data
Administration: Drinking water
Exposure period: 2 years
Frequency of treatment: Daily
Doses: 0.01%, 0.05%, 0.1% (20,100, 200 mg/kg bw d)
Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical
Results: There were no changes due to the administration of LAS in regard to growth, mortality, the weight of major organs, or histopathological findings. The intake of LAS was about 200 mg/kg bw d in the 0.1% group.
Method: Groups of 20 male Wistar rats were given LAS in drinking water daily for 2 years.
GLP: Yes No ?
Test substance: LAS, activity: 34.55%
Remarks: Information as cited in the IPCS document.
Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Tiba, S. 1972. Studies on the acute and chronic toxicity of linear alkylbenzene sulfonate. J. Food Hyg. Soc. Jpn. 16:66-71 (in Japanese);

cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(m)

Species/strain: Rhesus monkey (*Macaca mulatta*)
Sex: Female []; Male []; Male/Female []; No data []
Administration: Simultaneous oral and subcutaneous
Exposure period: 28 days
Frequency of treatment: daily
Dose: 30, 150, 300 mg/kg/day oral via gavage given simultaneously with 0.1, 0.5, 1.0 mg/kg/day subcutaneous administration
Control group: Yes []; No []; No data [];
Concurrent no treatment []; Concurrent vehicle []; Historical []
NOAEL: 150 mg/kg/day (oral) with 0.5 mg/kg/day (sc)
Results: At 300 (oral) and 1.0 (sc) mg/kg/day, the monkeys vomited frequently and usually within 3 hours of administration. An increased frequency of loose or liquid faeces was recorded for animals receiving 150 (oral) and 0.5 (sc) mg/kg. These effects are probably related to the inherent irritative effects of LAS rather than its systemic toxicity. Fibrosis of the injection sites was found among all the test group, the incidence and severity being dose related. Ophthalmoscopy, laboratory examination of blood and urine, organ weight analysis and histopathological investigation did not detect any further treatment-related responses.
Method: Three male and 3 female monkeys were given simultaneous oral and subcutaneous administration doses daily for 28 days. Animals were observed for physical and behavioral signs of toxicity. Analysis of blood, biochemistry and urine were conducted. Monkeys were held in individual wall-mounted cages at a room temperature of 22⁺/₋₁°C and normal daylight. Food consisted of 300 g dry diet and bread daily, and fresh fruit on alternate days. Tap water for drinking was freely available.
GLP: Yes [] No [] ? []
Test substance: C₁₀₋₁₃ LAS, activity: 20.5%
Remarks: The SDA/NOTOX report sets a systemic NOAEL of 30 mg/kg/day (oral) with 0.1 mg/kg/day (sc). However, based on the information provided in the article and reiterated above, the actual systemic NOAEL is considered to be 150 mg/kg/day (oral) with 0.5 mg/kg/day (sc). According to the authors, the three combined doses correspond to 100, 500 and 1000 times the estimated maximum human daily intake.
Reference: Heywood, R., James, R.W., and Sortwell, R.J. 1978. Toxicology studies of linear alkylbenzene sulphonate (LAS) in rhesus monkeys. I. Simultaneous oral and subcutaneous administration for 28 days. Toxicology 11:245-250.
Reliability: 2 Valid with restrictions

(n)

Species/strain: Rat (Wistar)
Sex: Female []; Male []; Male/Female []; No data []
Administration: Dermal

2) Sunakawa, T., Inoue, K. and Okamoto, K. 1981. Studies on the mutagenicity of surfactants, mutagenicity of surfactants following activation with various liver homogenates (S-9) and mutagenicity in the presence of norharman. Hyg. Chem. 27:204-211. (In Japanese); Cited in IPCS. 1996. Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates and Related Compounds. World Health Organization, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(d)

Type: *Bacillus subtilis* recombination assay

System of testing: H17 (rec+) and M45 (rec-)

Concentration: Up to 50 µg/plate

Metabolic activation: With []; Without []; With and Without [X]; No data []

Results:

Cytotoxicity conc: With metabolic activation: > 50 µg/plate

Without metabolic activation: > 50 µg/plate

Genotoxic effects: + ? -

With metabolic activation: [] [] [X]

Without metabolic activation: [] [] [X]

Method: *Bacillus* recombination assay

GLP: Yes [] No [X] ? []

Test substance: C₁₀₋₁₄ LAS, sodium salts (CAS #69669-44-9); average alkyl chain length (based on LAS SIDS Consortium Survey, 2000) = C_{11.7}; activity: 99.5%

Remarks: Information as cited in IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.

2) Inoue, IK. and Sunakawa, T. 1979. Mutagenicity tests of surfactants. Jpn Fragrance J. 38:67-75 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: for Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(e)

Type: *Escherichia coli* reverse mutation assay.

System of testing: WP23 uvr A

Concentration: Not specified

Metabolic activation: With []; Without []; With and Without [X]; No data []

Results:

Cytotoxicity conc: With metabolic activation: Not specified

Without metabolic activation: Not specified

Genotoxic effects: + ? -

With metabolic activation: [] [] [X]

Without metabolic activation: [] [] [X]

Method: *E. coli* assay

GLP: Yes [] No [X] ? []

Test substance: C₁₀₋₁₄ LAS, sodium salt; activity: 99.5%

Remarks: Information as cited in IPCS document.

Method: Chromosomal aberrations were examined 6, 24, 48 hours after administration. Mytomycin C was used as a positive control and appropriately induced severe chromosomal aberrations.

GLP: Yes No ?

Test substance: C₁₀₋₁₄ LAS, sodium salt (CAS #69669-44-9); average alkyl chain length (based on LAS SIDS Consortium Survey, 2000) = 11.7

Remarks: Besides the pure LAS, commercial preparation containing 19% LAS and another containing 17.1% LAS were given to mice as single doses only by gavage at 800, 1600 or 3200 mg/kg bw d and 1000, 2000 or 4000 mg/kg bw d, respectively. The highest doses were 50% of the respective LD₅₀ values. No significant differences in the incidence of chromosomal aberrations were observed in any LAS treatment relative to the controls. Information as cited in the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Inoue, K., Shibata, T., Hamano, Y., Oda, Y., Kuwano, A., Yamamoto, H., Mitsuda, B. and Kunita, N. 1977. *In vivo* cytogenetic tests of some synthetic detergents in mice. Ann Res. Osaka Prefect Inst. Public Health. 8:17-24 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document

(b)

Type: Mammalian bone marrow cytogenetic assay

Species/strain: rat (Wistar, SD); mouse (ICR)

Sex: Female ; Male ; Male/Female ; No data

Administration: oral feed

Exposure period: 9 months

Doses: 0.9% (450 mg/kg bw d in rats; 1170 mg/kg bw d in mice)

Results: There were no significant differences in the incidences of chromosomal aberrations between the experimental and control groups.

Method: Groups of 5 male Wistar rats, Sprague-Dawley rats, and ICR mice were given a diet containing 0.9% LAS for 9 months after which the chromosomes of the bone marrow cells were examined.

GLP: Yes No ?

Test substance: LAS

Remarks: Information as cited in the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Masabuchi, M., Takahashi, A., Takahashi, O. and Hiraga, K. 1976. Cytogenetic studies and dominant lethal tests with long term administration of butylated hydroxytoluene (BHT) and linear alkylbenzene sulfonate (LAS) in mice and rats. Ann. Rep. Tokyo Metrop. Res. Lab. Public Health. 27:100-104 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(c)

Type: Dominant lethal assay
Species/strain: mouse (ICR: JCL)
Sex: Female []; Male [X]; Male/Female []; No data []
Administration: oral feed
Exposure period: 9 months
Doses: 0.6% (300 mg/kg bw d)
Results: There were no significant differences in fertility, the mortality of ova and embryos, the number of surviving fetuses, or the index of dominant lethal induction between the experimental groups and the control group.
Method: Seven male mice received LAS in the diet for 9 months. Each of the male mice was then mated with 2 female mice that had not been given LAS. The pregnant mice were laparotomized on day 13 of gestation to determine the numbers of luteal bodies, implantations, surviving fetuses, and dead fetuses.
GLP: Yes [] No [X] ? []
Test substance: LAS
Remarks: Information as cited in the IPCS document.
Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Masabuchi, M., Takahashi, A., Takahashi, O. and Hiraga, K. 1976. Cytogenetic studies and dominant lethal tests with long term administration of butylated hydroxytoluene (BHT) and linear alkylbenzene sulfonate (LAS) in mice and rats. Ann. Rep. Tokyo Metrop. Res. Lab. Public Health. 27:100-104 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.
Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(d)

Type: Micronucleus assay
Species/strain: mouse: (ddy)
Sex: Female []; Male [X]; Male/Female []; No data []
Administration: Intraperitoneal injection
Exposure period: single dose
Doses: 100 mg/kg bw
Results: There were no differences in the incidences of polychromatic erythrocytes with micronuclei in the bone marrow cells between the treated group and the control group.
Method: Three male ddy mice were each given a single i.p. injection of 100 mg/kg bw LAS.
GLP: Yes [] No [X] ? []
Test substance: LAS
Remarks: Information as cited in the IPCS document.
Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Kishi, M., Satoh, S., Horiguchi, Y. and Ito, K. 1984. Effects of surfactants on bone marrow cells. Bull. Kanagaw Public Health Lab. 14:57-58. (In Japanese); cited in: IPCS (1996); Environmental Health Criteria 169:

Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

5.7 CARCINOGENICITY

(a)

Species/strain: Rat: Charles River
Sex: Female ; Male ; Male/Female ; No data
Administration: oral feed
Exposure period: 2 years
Frequency of treatment: continuous in feed
Doses: 0.02, 0.1, 0.5% (10, 50, 250 mg/kg bw d)
Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical
Results: Gross examination of all animals for pathology did not reveal any abnormalities. No consistent dietary induced changes that could be considered a toxic response were observed. Animals that showed significant loss of weight, development of tumors, or other evidence of abnormalities were sacrificed and tissues examined. The incidence of tumors and the common incidental diseases were similar in all dieting groups.
Method: Four groups of Charles River weanling rats, divided by sex, were given 0.5, 0.1, and 0.02% LAS in their food for 2 years. Following completion of those studies, five male and five female rats from each of the parental groups (F_{1b} and F_{2b}) and all survivors were selected for necropsy. Body weight and organ to body weight ratios were recorded, and routine hematology and histology were performed. Weanling animals for the F_{3a} generation were similarly treated.
GLP: Yes No ?
Test substance: C₁₀₋₁₄ LAS; activity: 98.1% on an anhydrous basis (41.9% active)
Reference: Buehler, E.V., Newmann, E.A., and King, W.R. 1971. Two year feeding and reproduction study in rats with linear alkylbenzene sulfonate (LAS). Toxicol. Appl. Pharmacol. 18:83-91.
Reliability: 2 Valid with restrictions

(b)

Species/strain: Rat/Wistar
Sex: Female ; Male ; Male/Female ; No data
Administration: Drinking water
Exposure period: 2 years
Frequency of treatment: Daily
Doses: 0.01%, 0.05%, 0.1% (20,100, 200 mg/kg bw d)
Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical
Results: There were no changes due to the administration of LAS in regard to growth, mortality, the weight of major organs, or histopathological findings. The intake of LAS was about 200 mg/kg bw d in the 0.1% group. There is no description of tumors.

Method: Groups of 20 male Wistar rats were given LAS in drinking water daily for 2 years.

GLP: Yes No ?

Test substance: LAS, activity: 34.55%

Remarks: Information as cited in the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Tiba, S. 1972. Studies on the acute and chronic toxicity of linear alkylbenzene sulfonate. J. Food Hyg. Soc. Jpn. 16:66-71 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document

(c)

Species/strain: Rat/Wistar

Sex: Female ; Male ; Male/Female ; No data

Administration: drinking water

Exposure period: up to 26 months

Frequency of treatment: Daily

Doses: 0.1% (140 mg/kg bw d)

Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical

Results: The administration of LAS had no effect on the intake of water, mortality, body weight gain, or general condition. In pathological examination, looseness, atrophy, and fatty change of the hepatic cells in the liver were found in the experimental group at 6 months. The experimental group showed significant increases in GOT, GTP and bilirubin at 6 months and thereafter. In haematological examinations no effects due to LAS were observed.

Method: A group of 62 male and 62 female rats were given drinking water treated with LAS and a control group of 37 male and 37 female rats were given pure water. Five to 12 of the rats in the experimental group and 3 to 12 rats in the control group at 3, 6, 12, and 18 months, respectively, and all surviving rats between 24 and 26 months, were sacrificed for pathological, biochemical, and haematological examinations.

GLP: Yes No ?

Test substance: LAS; mean molecular weight 348; average alkyl chain length = C_{12.0}; activity: 38.74%

Remarks: Information as cited in the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Endo, T., Furuido, Y., Namie, K., Yamamoto, N., Hasunuma, H. and Ueda, K. 1980. Studies of the chronic toxicity and teratogenicity of synthetic surfactants. Ann. Rep. Tokyo Metrop. Res. Inst. Environ. Prot. 236-246 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(d)

Species/strain: Rat/Wistar
Sex: Female ; Male ; Male/Female ; No data
Administration: oral feed
Exposure period: 1, 3, 6, 24, or more months
Frequency of treatment: Daily
Doses: 0.04, 0.16, 0.6% (20, 80, 300 mg/kg bw d)
Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical
Results: The 0.6% group showed slight increases in the weight of liver and cecum, and increased activity of GPT and ALP in serum. LAS administration had no adverse effects upon the intake of food, body weight gain, general condition, mortality or mean survival. At one month, proliferation of hepatic cells in the liver and slight swellings of the renal tubes and narrowing of the tubular lumen in the kidneys were found in the 0.16% and 0.6% groups. These findings later disappeared, and are considered to be adaptation phenomena to the administration of LAS. There were no histopathological lesions attributable to LAS administration in any of the organs in rats fed for 24 months or more. Various types of tumors were observed in various organs, but findings suggestive of tumorigenicity of LAS were not present. Therefore, the authors concluded that the diet containing LAS at a concentration of 0.6% (300 mg/kg bw d) did not have any adverse effects on rats.
Method: Groups of 50 male and 50 female rats were given diets containing LAS at 0.04, 0.16 and 0.6%. In each group, 5 rats of each sex were fed for 1, 3, 6, and 12 months, respectively, and groups of 15 rats of each sex were fed for 24 months or more. Detailed histopathological examinations were made on the rats.
GLP: Yes No ?
Test substance: C₁₀₋₁₄ LAS; C₁₀ 10.6%, C₁₁ 34.1%, C₁₂ 27.7%, C₁₃ 19.0%, C₁₄ 8.7%; average alkyl chain length = C_{11.8}; mean molecular weight 345.8
Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Fujii, T., Sakamoto, Y., Abe, Y., Mikurita, H., Yuzawa, K. and Hiraga, K. 1977. Pathological examination of rats fed with linear alkylbenzene sulfonate for their lifespan. Ann. Rep. Tokyo Metrop. Res. Lab. Public Health. 28:85-108 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.
3) Yoneyama, M., Masubuchi, M., Oishi, S., Takahashi, O., Ikawa, M., Yoshida, S., Oishi, H., Mikuriya, H., Yuzawa, K. and Hiraga, K. 1977. Subacute toxicity of linear alkylbenzene sulfonate. Ann. Rep. Tokyo Metrop. Res. Lab. Public Health. 28:73-84 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.
Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

5.8 TOXICITY TO REPRODUCTION

(a)

Type: Fertility []; One-generation study []; Two-generation study [];
Other [X] : 3-generation reproduction study

Species/strain: Rat: Charles River

Sex: Female []; Male []; Male/Female [X]; No data []

Administration: oral feed

Exposure period: 2 years

Frequency of treatment: continuous in feed

Premating
exposure period: male: 84 days, female: 84 days

Duration of the test: 3 generations

Doses: 0.02, 0.1, 0.5% (14, 70, 350 mg/kg bw d)

Control group: Yes [X]; No []; No data [] ;
Concurrent no treatment [X]; Concurrent vehicle []; Historical [X]

NOAEL Parental: = 0.5%. (350 mg/kg bw d)

NOAEL F1 Offspring: = 0.5% (350 mg/kg bw d)

NOAEL F2 Offspring: = 0.5% (350 mg/kg bw d)

Results: General reproduction including fertility gestation, parturition, neonatal viability, lactation, and post-weaning growth was normal for all test groups and did not deviate from the controls in each generation. No gross abnormalities were noted. No definitive adverse effects due to the test material were noted in the haematology and pathology.

Method: Na-LAS (chain length distribution C₁₀₋₁₄) was fed for 84 days to 4 groups of weanling rats (3 dose levels, plus control), each dose consisting of 50 animals each of both sexes (P₀-generation). When the P₀ generation was 107-112 days old, 20 females from each dose group were mated with 20 males from the same group and maintained together for 17 days. The first litters of each generation (F_{1a}- and F_{2a}-generation) were sacrificed at 21 days of age. Ten days after the final litter was sacrificed, all females were remated with different males from the same group to obtain the F_{1b} generation. From the F_{1b}-generation, 20 males and females of each group were selected at weaning to continue their respective diets and to be used for further reproduction studies. Reproduction studies on the F_{1b} and F_{2b} generations were started when the rats were 80 to 85 days old, and were continued until the F_{3b} generation was weaned.

GLP: Yes [] No [X] ? []

Test substance: Sodium salt LAS (C₁₀₋₁₄), activity: 98.1% on an anhydrous basis (41.9% active)

Reference: Buehler, E.V., Newmann, E.A., and King, W.R. 1971. Two year feeding and reproduction study in rats with linear alkylbenzene sulfonate (LAS). Toxicol. Appl. Pharmacol. 18:83-91.

Reliability: 2 Valid with restrictions

(b)

Type: Fertility []; One-generation study []; Two-generation study [] ;
Other [X] **Three generation study**

Species/strain: Charles River CD strain rats

Sex: Female []; Male []; Male/Female [X]; No data []

Administration: diet
 Exposure period: up to > 1 year
 Frequency of treatment: continuous in diet
 Premating exposure period: male: 60 days, female: 60 days
 Duration of the test: 3 generations
 Doses: 0.08, 0.4, and 2.0% continuously administered throughout the three generations (40, 200 and 1000 mg/kg bw d CLD [6.8, 3.4 and 170 mg/kg bw d LAS])
 Control group: Yes ; No ; No data ; Concurrent no treatment ; Concurrent vehicle ; Historical
 NOAEL Parental: 170 mg/kg bw d LAS (1000 mg/kg bw d CLD)
 NOAEL F1 Offspring: 170 mg/kg bw d LAS (1000 mg/kg bw d CLD)
 NOAEL F2 Offspring: 170 mg/kg bw d LAS (1000 mg/kg bw d CLD)
 Results: General parental toxicity: There were no signs of malreaction to treatment among parents and the incidence of sporadic deaths and total litter losses were unrelated to dosage. Pregnancy rate and the duration of gestation were unaffected. Food consumption and bodyweight changes showed no consistent relationship to dosage over the three generations.
 Toxicity to offspring: Examining litter parameters, statistically significant differences were occasionally observed but they showed no consistent dosage related trends over the three generations and were considered to be unrelated to treatment. The incidence of malformations was unaffected by treatment. Additional organ weight analysis, histopathology and skeletal staining of representative young from the F_{3b} generation revealed no changes that could be conclusively related to treatment.
 Method: CLD was administered in the diet of the rats and new batches of diet were prepared each week. Males and females of each generation (F₀, F_{1b}, and F_{2b}) were kept on their respective diets for 60 days. The mating period for the first litter lasted 19 days. After the weaning of the first litters, approximately 10 days, the animals were re-mated and a second litter was produced. From the second litters of the initial (F₀) and second (F_{1b}) generations, 10 males and 20 females were selected from each group at weaning in order to form the second and third (F_{2b}) generations, respectively.
 In the parent animals, observations of signs of reaction, mortalities, food consumption, bodyweight change, pregnancy rate, mating performance, and gestation period were made throughout the study. As soon as possible (< 12 hours) after birth, all young were counted, identified by toe amputation and examined for external abnormalities. Up to day 21 post partum, animals were examined daily for dead and abnormal young. Young of the first litters and surplus young of the second litters were sacrificed and examined for abnormalities internally and externally. Rats of the F_{3b} generation were killed at 3 weeks old and were also examined internally and externally for abnormalities. For the F_{3b} generation, tissue from the brain, liver, heart, pituitary, spleen, thyroid, kidneys, thymus, adrenals, lungs, gonads, pancreas, bladder, bone, bone marrow, sections of the stomach, and sections of the small and large intestines were removed and examined.
 GLP: Yes No ?
 Test substance: Commercial Light Duty liquid detergent (CLD) containing 17% LAS and 7% alkyl ether sulfate (Lion Oil and Fat Co., Ltd.)

Reference: Palmer, A.K., Cozens, D.D., Batham, P., and Cherry, C.P. 1974. Effect of CLD on reproductive function of multiple generations in the rat. Final Report. Report No. LF010/731029.

Reliability: 2 Valid with restrictions

(c)

Type: Fertility []; One-generation study []; Two-generation study [];
Other [X] : 4-generation reproduction study

Species/strain: Rat/Wistar

Sex: Female []; Male []; Male/Female [X]; No data []

Administration: drinking water

Frequency of treatment: Daily

Duration of the test: 4 generations

Doses: 0.1% (70 mg/kg bw d)

Control group: Yes [X]; No []; No data [];
Concurrent no treatment [X]; Concurrent vehicle []; Historical []

NOAEL Parental: 70 mg/kg bw d

NOAEL F1 Offspring: 70 mg/kg bw d

NOAEL F2 Offspring: 70 mg/kg bw d

Results: The administration of LAS had no adverse effects on fertility, parturition, gestation period, or lactation in any of the generations.

Method: Two groups of 20 rats of both sexes were given water containing LAS and the reproductive performance was investigated for 4 generations. Five to 10 rats of both the control and the experimental group were sacrificed at 12 weeks for pathological examinations. For successive reproduction, 15 males and 15 females produced by the first mating of rats were used.

GLP: Yes [] No [X] ? []

Test substance: LAS; mean molecular weight 348; average alkyl chain length = C_{12.0}.

Remarks: Information as cited in the IUCLID Data Set and the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Endo, T., Furuido, Y., Namie, K., Yamamoto, N., Hasunuma, H. and Ueda, K. 1980. Studies of the chronic toxicity and teratogenicity of synthetic surfactants. Ann. Rep. Tokyo Metrop. Res Inst. Environ. Prot. 236-246 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

5.9 DEVELOPMENTAL TOXICITY/ TERATOGENICITY

(a)

Species/strain: Rat (Wistar) and Rabbit (NZW)

Sex: Female [X]; Male []; Male/Female []; No data []

Administration: drinking water

Exposure period: day 6-15 of pregnancy (rat); day 6-18 of pregnancy (rabbit)

Frequency of treatment: Daily

Doses: 0.1% (70 mg/kg bw d in rat; 250 mg/kg bw d in rabbit)

Control group: Yes [X]; No []; No data [];

Concurrent no treatment []; Concurrent vehicle []; Historical []

NOAEL Maternal

Toxicity: Rat > 0.1% (383 mg/rat); rabbit = 0.1% (3030 mg/rabbit).

NOAEL teratogenicity: 0.1% for rat.

LOAEL teratogenicity: 0.1% (3030 mg/rabbit)

Results: The only effect on the dams was a slight inhibition of body weight gain in the rabbits. The litter parameters of both species did not show any significant differences from those of the controls. Delayed ossification was observed in rabbits, but there was no increase in malformations in either the rabbits or the rats.

Method: LAS was given to 40 rats (20 controls) and 22 rabbits (11 controls) from day 6 to 15 (rats) and day 6 to 18 (rabbits) of pregnancy, respectively.

GLP: Yes [] No [X] ? []

Test substance: LAS; activity: 38.74%; average alkyl chain length = C_{12.0}; (mean molecular weight 348)

Remarks: Information as cited in the IUCLID Data Set and the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Endo, T., Furuido, Y., Namie, K., Yamamoto, N., Hasunuma, H. and Ueda, K. 1980. Studies of the chronic toxicity and teratogenicity of synthetic surfactants. Ann. Rep. Tokyo Metrop. Res. Inst. Environ. Prot. 236-246 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(b)

Species/strain: Rat: CD

Sex: Female [X]; Male []; Male/Female []; No data []

Administration: gavage

Duration of the test: sacrifice at day 20 of gestation

Exposure period: day 6 - 15 of pregnancy

Frequency of treatment: daily

Doses: 0.2, 2, 300, 600 mg/kg bw d

Control group: Yes [X]; No []; No data [];
Concurrent no treatment [X]; Concurrent vehicle []; Historical []

NOAEL Maternal

Toxicity: 300 mg/kg bw

NOAEL teratogenicity: 600 mg/kg bw

Results: Maternal toxicity:
Body weight gain was retarded in the highest dose group from the start of dosing and showed partial recovery toward the end of the dosing period. One animal died in this group but it could not be conclusively related to treatment. The toxic effects were associated with disturbance of the gastrointestinal tract. Pregnancy rate was comparable at all dosages.
Teratogenicity:
No differences were observed among dose groups and the control group with respect to number of litters, viable young, litter weight, foetal weight, embryonic deaths, implantations, corpora lutea, pre- and post implantation embryonic loss, major malformations, minor visceral or embryonic loss,

major malformations, minor visceral or skeletal anomalies or incidence of pups with extra ribs.

Method: Animals received doses by gavage daily from days 6-15 of gestation. Twenty animals per dose group were used.

GLP: Yes No ?

Test substance: LAS

Reference: Palmer, A.K., Readshaw, M.A. and Neuff, A.M. 1975a. Assessment of the teratogenic potential of surfactants (Part I) – LAS, AS and CLD. Toxicology 3:91-106.

Reliability: 2 Valid with restrictions

(c)

Species/strain: Charles River CD strain rat

Sex: Female ; Male ; Male/Female ; No data

Administration: Oral in distilled water

Duration of the test: 20 days

Exposure period: day 6 to day 15 of pregnancy

Frequency of treatment: daily

Doses: 0.2, 2.0, 300 and 600 mg/kg

Control group: Yes ; No ; No data ; Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL Maternal: 300 mg/kg

NOAEL teratogenicity: 300 mg/kg

Results: Maternal toxicity: Parent animals were observed daily. Change in bodyweight was not affected by treatment at 0.2, 2.0, and 300 mg/kg, but treatment at 600 mg/kg was associated with retarded weight gain and a transient diarrhea following initiation of treatment. The pregnancy rate was comparable at all dosages.

Pregnancy/litter data: The litter parameters assessed included litter size, fetal loss and litter weight. These parameters were not significantly affected by any dosage. Mean pup weights were statistically higher at 0.2, 2.0 and 300 mg/kg.

Teratogenicity: Embryonic and fetal development were assessed by the incidence of major malformations. The incidence of minor visceral anomalies was unaffected by treatment at any dosage. The distribution of skeletal variants were not statistically significant with the exception of a marginal retardation of sternebral ossification at 600 mg/kg.

Method: After overnight mating, the rats were randomly allocated to five groups which included one control group and four different treatment groups. LAS was prepared daily as a series of graded aqueous solutions. Animals in all groups were dosed orally at the standard volume of 1.0 mL/100 g. Control animals were dosed in a similar manner with distilled water used as the vehicle. The dams were observed daily for signs of toxicity and weighed on days 1, 3, 6, 10, 14, 17 and 20 of pregnancy. On day 20, the rats were killed by CO₂ euthanasia. Their ovaries and uterine contents were examined immediately for number of corpora lutea, number of viable young, number of resorption sites, litter weight, and fetal abnormalities.

GLP: Yes No ?

Test substance: LAS (Na salt) as a slurry containing 64.0% w/v of active ingredient (Lion Oil and Fat Co., Ltd.); average alkyl chain length (based on LAS SIDS Consortium Survey, 2002) = C_{11.7-12.3}.

Reference: Palmer, A.K. and Lovell, M.R. 1971a. Effect of LAS detergent on pregnancy of the rat. Report No. 4331/71/487.
Reliability: 2 Valid with restrictions

(d)

Species/strain: Charles River Specific Pathogen Free mice of the CD-1 strain
Sex: Female ; Male ; Male/Female ; No data
Administration: Oral in distilled water
Duration of the test: 17 days
Exposure period: day 6 to day 15 of gestation
Frequency of treatment: daily
Doses: 0.2, 2.0, 300, 600 mg/kg
Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical
NOAEL Maternal: 2.0 mg/kg
NOAEL teratogenicity: 600 mg/kg
Results:

Maternal toxicity: After examining parent animals, treatment at 300 and 600 mg/kg was linked with increased mortality (35% and 90% respectively) and weight loss. At 600 mg/kg no dams bearing feasible young survived to termination. Autopsy revealed the consistent occurrence of tympanites sometimes associated with gastritis.

Pregnancy/litter data: The litter parameters were assessed by litter size, fetal loss and litter weight, none of which were significantly affected by treatment at any dosage. Mean pup weight was increased at 0.2 and 2.0 mg/kg.

Teratogenicity: Embryonic and fetal development was assessed by the incidence of major malformations and minor visceral anomalies and the distribution of skeletal variants. Any malformations and anomalies observed were not dose related. Development was not significantly affected at any dosage.

Method: After overnight mating, the mice were randomly allocated to five groups which included one control group and four different treatment groups. LAS was prepared daily as a series of graded aqueous solutions. Animals in all groups were dosed orally at the standard volume of 0.06 mL/10 g. Control animals were dosed in a similar manner with distilled water used as the vehicle. The dams were observed daily for signs of toxicity and weighed on days 1, 3, 6, 10, 14, and 17 of pregnancy. On day 17, the mice were killed by cervical dislocation. Their uterine contents were examined immediately for number of viable young, number of resorption sites, litter weight, and fetal abnormalities.

GLP: Yes No ?

Test substance: LAS (Na salt), as a slurry containing 64.0% w/v of active ingredient (Lion Oil and Fat Co., Ltd.); average alkyl chain length (based on LAS SIDS Consortium Survey, 2002) = C_{11.7-12.3}.

Reference: Palmer, A.K. and Lovell, M.R. 1971b. Effect of LAS detergent on pregnancy of the mouse. Report No. 4330/71/486.

Reliability: 2 Valid with restrictions

(e)

Species/strain: Mouse/CD-1
Sex: Female ; Male ; Male/Female ; No data
Administration: gavage

Duration of the test: sacrifice at day 17 of pregnancy
 Exposure period: days 6 - 15 of pregnancy
 Frequency of treatment: daily
 Doses: 0.2, 2, 300, 600 mg/kg bw d
 Control group: Yes ; No ; No data ;
 Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL Maternal
 Toxicity: 2 mg/kg bw d
 NOAEL teratogenicity: 300 mg/kg bw d
 Results: Maternal toxicity:
 Among parent animals treatment at 300 and 600 mg/kg bw d was associated with increased mortality (35% and 90% respectively). At 300 mg/kg bw d weight gain was retarded only during the first four days. No assessment could be made at 600 mg/kg bw d, due to the high mortality rate. Necropsy revealed a ubiquitous occurrence of tympanites, sometimes associated with gastritis. Pregnancy rate was essentially comparable for all groups.
 Teratogenicity:
 At doses with no maternal toxicity, no differences were observed among the dose group and the control group with respect to number of litters, viable young, litter weight, foetal weight, embryonic deaths, implantations and post implantation embryonic loss. At these doses the incidences of major malformations and minor abnormalities were not affected. At doses with maternal toxicity there was an increased foetal loss and reduced litter size due almost entirely to total litter loss, which was considered to be a secondary effect due to the maternal toxicity. The incidences of major malformations was not affected; minor skeletal or visceral anomalies were increased at 300 mg/kg.

Method: Twenty female mice were administered 0.2, 2.0, 300, or 600 mg/kg bw of LAS by gavage at days 6-15 of gestation. All animals were sacrificed at day 17 of pregnancy.

GLP: Yes No ?

Test substance: LAS

Remarks: The maternal NOAEL of 2 mg/kg bw d is considered very conservative because the range (2-300 mg/kg bw d) was too wide.

Reference: Palmer, A.K., Readshaw, M.A. and Neuff, A.M. 1975a. Assessment of the teratogenic potential of surfactants (Part I) – LAS, AS and CLD. Toxicology 3:91-106.

Reliability: 2 Valid with restrictions

(f)

Species/strain: New Zealand white rabbit
 Sex: Female ; Male ; Male/Female ; No data

Administration: Gavage
 Exposure period: day 6 to 18 of pregnancy
 Frequency of treatment: Daily
 Duration of the test: sacrifice at day 29 of pregnancy
 Doses: 0.2, 2, 300, 600 mg/kg
 Control Group: Yes; concurrent
 NOAEL maternal toxicity: 2 mg/kg bw
 NOAEL teratogenicity: 2 mg/kg bw

GLP: Yes No ?

Test substance: LAS

Results: Maternal toxicity:
At 300 and 600 mg/kg severe maternal toxicity was observed resulting in body weight loss and associated with diarrhoea, anorexia, and cachexia prior to death.

Teratogenicity:
At doses with no maternal toxicity, no differences were observed among the dose groups and the control group with respect to number of litters, viable young, litter weight, foetal weight, embryonic deaths, implantations and post implantation embryonic loss. At these doses the incidences of major malformations and minor abnormalities were not affected. Higher doses resulted in total litter which was considered to be a secondary effect due to the maternal toxicity. Since there were no survivors, malformations and anomalies were not assessed at these doses.

Remarks: Information as cited in the IUCLID Data Sheet and the IPCS document.

Reference: Palmer, A.K., Readshaw, M.A. and Neuff, A.M. 1975a. Assessment of the teratogenic potential of surfactants (Part I) – LAS, AS and CLD. Toxicology 3:91-106.

Reliability: 2 Valid with restrictions

(g)

Species/strain: New Zealand White rabbit

Sex: Female ; Male ; Male/Female ; No data

Administration: intragastric intubation

Duration of the test: 29 days

Exposure period: day 6 to day 18 of pregnancy

Frequency of treatment: daily

Doses: 0.2, 2.0, 300, 600 mg/kg

Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL Maternal

Toxicity: 2.0 mg/kg

NOAEL teratogenicity: 2.0 mg/kg

Results: Maternal toxicity: Daily assessment of bodyweight change and pregnancy rate determined that at 0.2 and 2.0 mg/kg the treatment did not adversely affect parent animals. At 300 and 600 mg/kg parent animals showed signs of severe anorexia, diarrhoea, weight loss and death. Respective mortality rates were 85 and 100% and autopsy consistently revealed changes in the gastrointestinal tract.

Pregnancy/litter data: The influence of maternal toxicity restricted assessment of effect on litter parameters to animals treated at 0.2 and 2.0 mg/kg. At these two dosages there were no adverse effects on litter parameters, as assessed by litter size and fetal loss, litter and mean pup weights.

Teratogenicity: Also at these two dosages there were no adverse effects on embryonic and fetal development, as assessed by the incidence of major and minor malformations, minor anomalies and skeletal variants.

Method: Thirteen rabbits were mated on a one-to-one basis with males of proven fertility. The does were then injected intravenously with 10 i.u. luteinizing hormone to ensure that ovulation occurred. The rabbits were identified by an ear tag and allocated to one control group and four

treatment groups. LAS was prepared daily and administered by intragastric intubation by a series of graded solutions in distilled water so that all animals were dosed at the standard volume of 4 mL/kg. Control animals were dosed at the same rate with distilled water as the vehicle. The parent animals were observed daily for signs of toxicity and weighed on days 1, 6, 10, 14, 21, and 28. On day 29, the animals were killed by cervical dislocation and immediately examined to determine the numbers and uterine disposition of young and resorption sites. The number of corpora lutea were also counted. Any rabbit containing abnormal fetus and/or resorption sites was thoroughly examined for signs of natural disease. Viable young were weighed, sexed and examined internally and externally for abnormalities. All young were preserved in alcohol for subsequent clearing, staining and skeletal examination. Resorption sites were classified as early or late. Abnormalities were classified as major or variant.

GLP: Yes No ?
 Test substance: LAS (Na salt), as a slurry containing 64% active ingredient (Lion Oil and Fat Co., Ltd.); average alkyl chain length (based on LAS SIDS Consortium Survey, 2002) = C_{11.7-12.3}.
 Remarks: The maternal NOAEL of 2 mg/kg bw d is considered very conservative because the range (2-300 mg/kg bw d) was too wide.
 Reference: Palmer, A.K. and Neuff, A.M. 1971. Effect of LAS detergent on pregnancy of the New Zealand white rabbit. Report No. 4387/71/543.
 Reliability: 2 Valid with restrictions

(h)

Species/strain: Mouse/ICR
 Sex: Female ; Male ; Male/Female ; No data
 Administration: gavage
 Duration of the test: See method.
 Exposure period: See method.
 Frequency of treatment: daily
 Doses: 0.4, 4.0% (40, 400 mg/kg bw d)
 Control group: Yes ; No ; No data ;
 Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL Maternal

Toxicity: 40 mg/kg bw d
 NOAEL teratogenicity 400 mg/kg bw d
 Results: In mice given 400 mg/kg from day 0 to 6, the pregnancy rate was 46.2% compared to 92.9% in the controls. There was no increase in malformations. Although no information on maternal toxicity is available, it appears likely that maternal toxicity was present at the high dose group.

Method: LAS was administered from day 0 to day 6 of pregnancy or from day 7 to 13 of pregnancy. Thirteen to fourteen mice were used in each dose group.

GLP: Yes No ?
 Test substance: Japan LAS; average alkyl chain length (based on LAS SIDS Consortium Survey, 2002) = C_{11.7-12.3}; activity: 99.5%

Remarks: Information as cited in the IUCLID Data Sheet and the IPCS document.
 Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
 2) Takahashi, M., Sato, K., Ando, H., Kubo, Y. and Hiraga, K. 1975. Teratogenicity of some synthetic detergents and linear alkylbenzene

sulfonate (LAS). Ann. Rep. Tokyo Metrop. Res. Lab. Public Health 26: 67-78 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(i)

Species/strain: Mouse/ICR.
Sex: Female ; Male ; Male/Female ; No data
Administration: gavage
Duration of the test: see text.
Exposure period: day 6 through day 15 of pregnancy.
Frequency of treatment: daily.
Doses: 10, 100, 300 mg/kg bw d
Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical

LOAEL Maternal

Toxicity: 10 mg/kg bw d

NOAEL teratogenicity: 300 mg/kg bw d

Results: The dams showed inhibition of body weight gains in all groups, especially in the high dose group. In this group, two dams died, and there was one case of premature delivery and death of all fetuses. There were findings such as decreased body weight and delayed ossification among the living fetuses, but there was no increase in malformations.

Method: LAS was administered by gavage to 25 to 33 mice per dose on days 6 through 15 of gestation.

GLP: Yes No ?

Test substance: Japan LAS; average alkyl chain length (based on LAS SIDS Consortium Survey, 2002) = C_{11.7-12.3}; activity: 48.6%

Remarks: Information as cited in the IUCLID Data Sheet and the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Shiobara, S. and Imahori, A. 1976. Effects of linear alkylbenzene sulfonate orally administered to pregnant mice and their fetuses. J. Food Hyg. Soc. Jpn. 17:295-301 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(j)

Species/strain: Rat: SD-JCL
Sex: Female ; Male ; Male/Female ; No data
Administration: oral feed
Exposure period: from day 0 to 20 of gestation
Frequency of treatment: Daily
Doses: 0.1%, 1.0% (80, 780 mg/kg bw d)
Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL Maternal: 780 mg/kg bw d
 NOAEL teratogenicity: 780 mg/kg bw d
 Results: The LAS intake was about 780 mg/kg with the 1% diet, but there were no abnormalities in the body weight gains of the dams, or in the occurrence and maintenance of pregnancy. The values of the litter parameters did not differ from those of the controls and there was no evidence of teratogenicity. The numbers of offspring were rather low in the 1% group, and the weaning rate of 78.3% was lower than the 100% rate observed in the controls. However, there were no abnormalities in body weight gain, organ weights or functions in the offspring.
 Method: LAS was fed in the diet to 16 pregnant female rats/dose from day 0 to 20 of gestation.
 GLP: Yes No ?
 Test substance: Japan LAS; average alkyl chain length (based on LAS SIDS Consortium Survey, 2002) = C_{11.7-12.3}.
 Remarks: Information as cited in IUCLID Data Sheet and IPCS document.
 Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
 2) Tiba, S., Shiobara, S., Imahori, A. and Kitagawa, T. 1976. Effects of linear alkylbenzene sulfonate on dam, fetus and newborn rat. J. Food Hyg. Soc. Jpn. 17:66-71. (In Japanese); cited in IPCS. 1996. Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates and Related Compounds. World Health Organization, Geneva, Switzerland. Original article in Japanese.
 Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(k)
 Species/strain: Weanling Charles River CD Sprague-Dawley albino rats and New Zealand rabbits
 Sex: Female ; Male ; Male/Female ; No data
 Administration: oral in feed
 Duration of the test: two generation (rats); one generation (rabbits)
 Exposure period: Rat: continuously or during organogenesis period of six pregnancies
 Rabbit: day 2-16 of gestation during a single pregnancy
 Frequency of treatment: continuous
 Doses: Rat: 0.1, 0.5 or 1.0% TAE₃S/LAS (equivalent to 50, 250 or 500 mg/kg/day in female rats corresponding to LAS doses of 22.5, 112.5 and 225 mg/kg bw d.)
 Rabbit: 50, 100, or 300 mg/kg TAE₃S/LAS, corresponding to LAS doses of 22.5, 45, and 135 mg/kg bw d
 Control group: Yes ; No ; No data ;
 Concurrent no treatment ; Concurrent vehicle ; Historical
 NOAEL Maternal: 225 mg/kg bw d LAS (rat)
 135 mg/kg bw d LAS (rabbit)
 NOAEL teratogenicity: 225 mg/kg bw d LAS (rat)
 135 mg/kg bw d LAS (rabbit)
 Results: Maternal toxicity: No treatment related adverse effects were observed in the maternal generation of either rats or rabbits. Fetal toxicity: No treatment-related adverse effects were observed on conception, fetal viability or post-natal survival in either generation of rats. Some

statistically significant differences were observed in live-born and surviving pup numbers, but there were no consistent trend or patterns. Combined with the high lactation index in all groups, indicating a very low mortality among the suckling rats, these differences were considered due to causes not related to treatment with the test material. There were no statistical differences among the groups of rat fetuses taken by Caesarian section and examined for birth defects. Some minor soft-tissue and skeletal anomalies were observed in rat fetuses from both generations. Of the 1210 rat fetuses, the overall incidence of abnormal young was 9.0% and did not vary significantly between treatment groups or the controls. Similarly, no treatment-related adverse effects were seen in rabbits treated with the surfactant mixture. Of the 855 rabbit fetuses, 5.7% were abnormal, but the incidence of defective fetuses in the test groups were not significantly different from those in controls. Therefore, no test related effects were seen on reproduction or embryonic development in either animal species.

Method:

(A) Rat studies

The rats were divided into seven groups consisting of 25 males and 25 females after a five day acclimation period in the laboratory. The tallow alkyl ethoxy sulfate (55%)-LAS(45%) mixture (TAE₃S/LAS) was mixed into the ground commercial feed at levels of 0.1, 0.5 or 1.0% and fed to two generations of male and female rats continuously or only to females during each period of organogenesis (days 6-15) of pregnancy. A control group was fed the commercial feed with no additive. The parent animals body weights were recorded weekly for the first eight weeks in each generation and afterwards recorded only at each mating phase.

Once sexually mature, five rats of each sex per group were sacrificed for histology during each generation. The remaining rats were mated on a one-to-one basis three successive times during each generation. The first two pregnancies (F_{1a}, 1b, 2a and 2b) in each generation were allowed to proceed to natural births. These pups were counted and inspected for abnormalities at birth. The third pregnancies in each generation (F_{1c} and F_{2c}) were used for teratology purposes. At weaning all pups except the F_{1b} litters, which became the second generation parents, were discarded. Animals for the second generation were selected on the same basis as previously described for the first generation. During the third pregnancies of both generations (F_{1c} and F_{2c}), one-half of each group of females was sacrificed on day 13 of gestation. A laparotomy was performed and the number of corpora lutea of pregnancy and the number of implantation and resorption sites were observed and recorded. On day 21 of gestation, the remaining dams were examined in a similar manner.

One-third of the fetuses in each of the third litters were examined for skeletal development and defects. The others were examined for soft tissue defects. During the teratology period, tissues were collected from five parent females of each group and from five parent males of the control and continuously treated groups. The heart, liver, kidneys and gonads were weighed, blood was taken for routine hemograms, and tissues were set in 10% formalin, paraffin-sectioned and stained with haemotoxylin-eosin for histopathy.

(B) Rabbit study

Five groups of 25 sexually mature does were distributed on the based on body weights and litter mates. The does were artificially inseminated with

0.25 mL of undiluted semen, collected from sperm-tested untreated males. Ovulation was induced by a 1 mg/kg injection of PLH immediately prior to insemination. The day of insemination was considered day 0 of gestation. The TAE₃S/LAS mixture was administered by gavage from day 2 through day 16 of gestation at daily doses of 50, 100, or 300 mg/kg of body weight. Distilled water was the vehicle and each doe received 2 mL of solution per kg of bodyweight. For the control groups, one received no treatment and the other received a treatment with water. In order to monitor the dose level, the females were weighed every three days. The dams were sacrificed on day 28 of gestation and the number of corpora lutea, resorptions and live or dead fetuses were observed and recorded. The fetuses were removed and treated and examined for abnormalities.

GLP: Yes No ?

Test substance: A mixture of 55% tallow alkyl ethoxylate sulfate (TAE₃S) and 45% LAS (assumed Procter and Gamble products)

Remarks: The authors indicate that rats received up to 6000 times the estimated “worst-case” human exposure without causing any deleterious effects on the development or variability of the embryo or fetus. Rabbits also received doses many times the “worst-case” human dose without causing significant effects.

Reference: Nolen, G.A., Klusman, L.W., Patrick, L.F. and Geil, R.G. 1975. Teratology studies of a mixture of tallow alkyl ethoxylate and linear alkylbenzene sulfonate in rats and rabbits. Toxicology. 4:231-243.

Reliability: 2 Valid with restrictions

(1)

Species/strain: Rat: CD

Sex: Female ; Male ; Male/Female ; No data

Administration: Dermal

Exposure period: days 2 through 15 of gestation

Frequency of treatment: daily

Doses: 0.03, 0.3, or 3% on the shaved skin as 0.5 ml aqueous solution.

Control group: Yes ; No ; No data ; Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL Maternal: 0.3% (6 mg/kg bw d)

NOAEL teratogenicity: 3% (60 mg/kg bw d)

Results: Maternal toxicity: At the high dose, local irritation was observed resulting in a slightly lower body weight gain and hypersensitivity (i.e., animals were increasingly irritable).
Teratogenicity: No differences were observed among the dose groups and the control group with respect to number of litters, viable young, litter weight, foetal weight, embryonic deaths, implantations, corpora lutea, or pre- and post implantation embryonic loss. The incidences of major malformations, minor visceral or skeletal anomalies, and skeletal variants were not different between controls and dose groups even at maternal by toxic doses.

Method: The dosage volume was 0.5 mL which was applied to an area of skin (4x4 cm) from which the fur was removed. The nominal doses were 0.6, 6.0, and 60 mg/kg bw d.

GLP: Yes No ?

Test substance: Japan LAS; average alkyl chain length (based on LAS SIDS Consortium Survey, 2002) = C_{11.7}.

Reference: Palmer, A.K., Readshaw, M.A. and Neuff, A.M. 1975b. Assessment of the teratogenic potential of surfactants (Part III) – Dermal application of LAS and soap. Toxicology 4:171-181.

Reliability: 2 Valid with restrictions

(m)

Species/strain: Rat/Wistar

Sex: Female ; Male ; Male/Female ; No data

Administration: Dermal

Duration of the test: sacrifice at day 21 of gestation

Exposure period: 21 days (days 0 through 21 of gestation)

Frequency of treatment: Daily

Doses: 0.05, 0.1, and 0.5% (0.1, 2 and 10 mg/kg bw d) or 1.0, 5.0, and 20% (20, 100, and 400 mg/kg bw d)

Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL Maternal: 1% (20 mg/kg bw d)

NOAEL teratogenicity: 20% (400 mg/kg bw d)

Results: Maternal toxicity:
The dams treated with 20% and 5% showed inhibition of body weight gain and local skin effects.
Teratogenicity:
There were no indications of teratogenic or embryotoxic effects at any level in either group tested.

Method: LAS was applied to depilated areas on the chests and backs of female rats 12-18 weeks of age. Five to six hours prior to treatment an exposure site (roughly 24 cm²) in the dorsothoracic region of each animal from group II through IX was clipped to a length of 1 mm. The animals were reclipped every 48 hr throughout the study. Group I animals were unclipped, group II animals were clipped but not treated and group III animals were clipped and treated with tap water. The mated female rats were treated daily from day 0 through day 20 of gestation. A 0.5-ml sample of the appropriate concentration of LAS and/or tap water was applied once daily to the clipped area and spread with a gloved finger over as much of the exposure site as possible. Each application was carried out slowly over a 3-min period. In the 1, 5 and 20% LAS groups (groups VII, VIII and IX, respectively corresponding to 20, 100 and 400 mg/kg/day) the test material was allowed to remain on the backs of the animals for 30 min. after which it was removed with warm tap water. The test material was not removed from the backs of the animals in the 0.05, 0.1 and 0.5% LAS groups (groups IV, V and VI corresponding to 1, 2 and 10 mg/kg/day). Animal body weight and food consumption were determined during the treatment period. Daily observations were also made for toxicological effects.

GLP: Yes No ?

Test substance: LAS; mean chain length: 11.7; mean molecular weight: 344, activity: 20.5%

Reference: Daly, I.W., Schroeder, R.E. and Killeen, J.C. 1980. A teratology study of topically applied linear alkylbenzene sulphonate in rats. Fd. Cosmet. Toxicol. 18:55-58.

Reliability: 2 Valid with restrictions

(n)

Species/strain: Mouse/CD-1
Sex: Female ; Male ; Male/Female ; No data
Administration: Dermal
Exposure period: days 2 through 13 of gestation
Frequency of treatment: daily
Doses: 0.03, 0.3, or 3% LAS in aqueous solution onto the shaved skin (5, 50 and 500 mg/kg bw d)
Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical
NOAEL Maternal: 0.03% (5 mg/kg bw d)
NOAEL teratogenicity: 0.3% (50 mg/kg bw d)
Results: Maternal toxicity:
At the high dose, severe local irritation was observed resulting in body weight loss and hypersensitivity (i.e., animals were increasingly irritable), which was also observed at the medium dose. The conclusion of the authors was that LAS caused marked toxicity at the high dose and moderate or mild toxicity at the medium dose.
Teratogenicity:
At the lowest dose, the dose with no maternal toxicity, no differences were observed among the LAS group and the control group with respect to number of litters, viable young, litter weight, foetal weight, embryonic deaths, implantations, corpora lutea, pre- and post implantation embryonic loss. The incidences of major malformations, minor visceral or skeletal anomalies, and skeletal variants were not different between controls and the LAS group.
Maternally toxic dosages were associated with a significantly increased foetal loss and consequent reduction of litter size. This was due almost entirely to total litter losses as values, for the one surviving litter at 3% was similar to the control litters. At the medium dose, the moderate degree of maternal toxicity correlated with a moderate effect on litter values in that, whilst the higher incidence of embryonic deaths differed significantly from control values, the consequent reduction in litter size was not statistically significant. With regard to major malformations and minor skeletal or visceral anomalies, the assessment of litters was not possible in the highest dose group due to the low survival. At the low doses, no treatment related increase of the incidences of major malformations and minor skeletal and visceral anomalies were observed.
Method: The dosage volume was 0.5 mL which was applied to an area of skin (2 x 3 cm) from which the fur was removed. The nominal doses were 5, 50, and 500 mg/kg bw/day.
GLP: Yes No ?
Test substance: Japan LAS; average alkyl chain length (based on LAS SIDS Consortium Survey, 2002) = C_{11.7-12.3}.
Reference: Palmer, A.K., Readshaw, M.A. and Neuff, A.M. 1975b. Assessment of the teratogenic potential of surfactants (Part III) Dermal application of LAS and soap. Toxicology. 4:171-181.
Reliability: 2 Valid with restrictions

(o)

Species/strain: mouse: ddy.

Sex: Female ; Male ; Male/Female ; No data

Administration: Dermal

Exposure period: day 0 through day 13 of pregnancy

Frequency of treatment: Daily

Doses: 2.2% (110 mg/kg bw d)

Control group: Yes ; No ; No data ; Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL Maternal: 2.2% (110 mg/kg bw d)

NOAEL teratogenicity: 2.2% (110 mg/kg bw d)

Results: No abnormalities were seen in the dam or fetuses.

Method: An area of 4 x 4 cm on the backs of mice was depilated and LAS was applied at a dose of 0.5 ml/mouse/day. Sixteen animals were used per group.

GLP: Yes No ?

Test substance: LAS; molecular wt = 346; average alkyl chain length = 11.8; activity: 99.5%

Remarks: Information as cited in the IUCLID Data Sheet and the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Sato, K., Ando, H., Yuzawa, K. and Hiraga, K. 1972. Studies on toxicity of synthetic detergents (III) Examination of teratogenic effects of alkyl benzene sulfonates spread on the skin of mice. Ann. Rep. Tokyo Metrop. Res. Lab. Public Health. Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(p)

Species/strain: mouse/ICR

Sex: Female ; Male ; Male/Female ; No data

Administration: dermal

Exposure period: from day 6 through day 15 of pregnancy

Frequency of treatment: daily

Doses: 0.03, 0.3, 3% (15, 150, and 1500 mg/kg bw d)

Control group: Yes ; No ; No data ; Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL Maternal: 0.3% (150 mg/kg bw d)

NOAEL teratogenicity: 3% (1500 mg/kg bw d)

Results: The 3% group showed a clear decrease in the pregnancy rate (67.9%) when compared with a rate of 96.3% in the controls. However, there were no decreases in the litter size, and no changes in the litter parameters with the exception of a slight decrease in fetal body weight. There were no significant increases in the incidence of malformations in the fetuses.

Method: Areas of 4 x 4 cm on the backs of the mice were depilated and aqueous solutions of LAS were applied.

GLP: Yes No ?

Test substance: C₁₀₋₁₄ LAS (CAS #69669-44-9); average alkyl chain length (based on LAS SIDS Consortium Survey, 2002) = C_{11.7}; activity: 46.6%

Remarks: Information as cited in the IUCLID Data Sheet and the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Imahori, A., Kinagawa, T. and Shiobara, S. 1976. Effects of linear alkyl benzene sulfonate (LAS) applied dermally to pregnant mice and their fetuses. Jpn. J. Public Health. 23:68-72 (in Japanese); cited in IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(q)

Species/strain: mouse/ICR
Sex: Female ; Male ; Male/Female ; No data
Administration: Subcutaneous injection
Exposure period: Day 0 to 3 or day 8 to 11 of pregnancy
Frequency of treatment: daily
Doses: 0.35, 1% in water (20, 200 mg/kg bw d)
Control group: Yes ; No ; No data ;
Concurrent no treatment ; Concurrent vehicle ; Historical
NOAEL Maternal: 0.35% (20 mg/kg bw d)
NOAEL teratogenicity : 1% (200 mg/kg bw d)
Results: When dams were administered the 1% solution from day 0 to 3 of pregnancy, there was an initial decrease in body weight and necrosis at the injection sites. The number of pregnancies decreased in the mice given the 1% solution compared to the controls (61.5% vs. 93.3%) There were no significant changes with respect to litter parameters, major malformations or minor abnormalities.

Method: LAS was injected at doses of 20 mL/kg/day from day 0 to 3 or day 8 to 11 of pregnancy. There were 12 - 19 mice in each treatment group.

GLP: Yes No ?
Test substance: LAS, activity: 99.5%
Remarks: Information as cited in the IUCLID Data Sheet and the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Takahashi, M., Sato, K., Ando, H., Kubo, Y. and Hiraga, K. 1975. Teratogenicity of some synthetic detergents and linear alkylbenzene sulfonate (LAS) Ann. Rep. Tokyo Metrop. Res. Lab. Public Health 26:67-78 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document

(r)

Species/strain: rabbit/New Zealand white
Sex: Female ; Male ; Male/Female ; No data
Administration: Dermal
Exposure period: 17 days (days 1 through 16 of gestation)
Frequency of treatment: Daily
Doses: 0.03, 0.3, or 3% (0.9, 9, or 90 mg/kg bw d)

Control group: Yes ; No ; No data ;
 Concurrent no treatment ; Concurrent vehicle ; Historical

NOAEL Maternal: 0.03% (0.9 mg/kg bw d)

NOAEL teratogenicity: 3% (90 mg/kg bw d)

Results: Maternal toxicity:
 At the high dose, local irritation was observed resulting in body weight loss and hypersensitivity (i.e., animals were increasingly irritable). The medium dose caused retarded body weight gain and hypersensitivity.

Teratogenicity:
 At the medium and low dose, no differences were observed among the dose groups and the control group with respect to number of litters, viable young, litter weight, foetal weight, embryonic deaths, implantations, corpora lutea, pre and post implantation embryonic loss. The high dose was associated with slightly, but not significantly, higher foetal loss and lower litter size. The incidences of major malformations, minor visceral or skeletal anomalies, and skeletal variants were not different between controls and dose groups even at maternal toxic doses.

Method: The dosage volume was 10 mL which was applied to an area of skin (12 x 20 cm) from which the fur was removed. The nominal doses were 0.9, 9.0, and 90 mg/kg bw/day. Thirteen rabbits per dose were used.

GLP: Yes No ?

Test substance: LAS

Reference: Palmer, A.K., Readshaw, M.A. and Neuff, A.M. 1975b. Assessment of the teratogenic potential of surfactants (Part III) – Dermal application of LAS and soap. Toxicology 4:171-181.

Reliability: 2 Valid with restrictions

5.10 OTHER RELEVANT INFORMATION

(a)

Type: Toxicokinetics

Results: ³⁵S-LAS (15 x 10⁸ cpm) was administered topically, once, onto the back skin of rats and guinea pigs. Absorption and distribution in major organs and blood were studied. Urine was collected 24 hours after topical application of the test substance. In the guinea pig, the amount of ³⁵S excreted in the urine was about 0.1% of the total administered dose. Organ distribution in the rat was about 5 times greater than in the guinea pig, and "relatively large amounts" of ³⁵S were noted in the liver and kidneys. Conclusion states that: "when 0.2 to 0.5% LAS was topically applied once, approximately 0.1 to 0.6% was absorbed"; there was no accumulation in specific organs; the "test chemical was quickly excreted in the urine after being metabolized".

Reference: Debane, C. 1978. National Hygiene Laboratory; in: "Report on Studies on Synthetic Detergents", October 1978, Japan's Science and Technology Agency [in Japanese].

Reliability: 4 Not assignable

(b)

Type: Toxicokinetics

Method: The absorption, distribution, metabolism and elimination of LAS (radioactively labelled with ³⁵S) were studied in male Charles River rats. LAS was administered as an aqueous solution.

Results: The compound was readily absorbed from the gastrointestinal tract (80-90% of the dose). Most of the absorbed ³⁵S was eliminated within 72 hours and 60-65% of the absorbed dose was eliminated in the urine, with sulfophenyl butanoic and sulfophenyl pentanoic acid as metabolites. These metabolites were not reabsorbed from the kidney tubules. 35% of the absorbed ³⁵S was excreted in the bile and were reabsorbed completely from the gastrointestinal tract. Although the metabolites in the bile were not identified, it was shown that no unchanged LAS was eliminated via this pathway.

Test substance: C₁₀₋₁₃, LAS (CAS #68411-30-3); alkyl chain length predominately C₁₁, C₁₂ and C₁₃.

Remarks: The authors suggested that metabolism proceeded via omega oxidation with subsequent beta-oxidation. Retention of radioactivity was not observed in any organ.

Reference: Michael, W.R. 1968. Metabolism of linear alkylate sulfonate and alkyl benzene sulfonate in albino rats. *Toxicol. Appl. Pharmacol.* 12:473-485.

Reliability: 2 Valid with restrictions

(c)

Type: Toxicokinetics

Results: LAS is well absorbed by via the gastrointestinal tract of pigs treated with 3.3 mmol/animal ³⁵S-Na-dodecylbenzene sulfonate. At 200 hours after oral administration, the radioactivity was relatively high in bristles and bones, while low in liver, kidney and spleen. After 10 weeks only traces of radioactivity were still in the body. 40 hours after the administration, 40% of the dose was excreted into the urine and 60% of the dose via the faeces.

Remarks: Information as cited in the IUCLID Data Sheet and the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Havermann, H. and Menke, K.H. 1959. Biological study of the water-soluble surface-active substances. *Fette. Seifen. Anstrichmittel* 61:429-434. (in German); cited in IPCS. 1996. Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates and Related Compounds. World Health Organization, Geneva, Switzerland. Original article in Japanese.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(d)

Type: Toxicokinetics

Results: Four (2 male, 2 female; 5 kg average body weight) adult rhesus monkeys (*Macaca mulatta*) were given single or repeated oral (30, 150 or 300 mg/kg) or subcutaneous (0.1, 0.5 or 1 mg/kg) doses of ¹⁴C-LAS. After single 30 mg/kg oral doses the radioactivity was rapidly excreted, mostly during the first 24 hours. Means of 71.2% and 23.1% of the dose were excreted in the urine and feces, respectively, during 5 days. Similarly, after single 1 mg/kg subcutaneous doses, means of 64.1% and 10.9% were excreted in urine and feces, respectively, during 5 days, mostly during the

first 24 hours. After single oral doses of 30, 150 and 300 mg/kg, peak plasma concentrations (at 4 hours in all cases) were very similar, with levels of 34, 41 and 36 µg/mL, respectively. Concentrations declined during the period of 6-24 hours, with a biological half life of about 6.5 hours. After single subcutaneous doses of 0.1, 0.5 and 1 mg/kg, peak plasma concentrations increased almost proportionately, with levels of 0.16, 0.72 and 1.13 µg/mL, respectively. During the 120 hours after single oral (30 mg/kg) or subcutaneous doses (1 mg/kg) the average rate of excretion was between 63 and 74% in the urine and between 9 and 26% in the feces.

During seven consecutive daily oral (30 mg/kg/day) or subcutaneous (1 mg/kg/day) doses, there was no accumulation of radioactivity in plasma. Mean peak concentrations and biological half-lives were similar after the first and seventh doses. Two hours after the last dose, the highest radioactivity was observed in the stomach. Radioactivity was also observed in the intestinal tract, kidneys, liver, lung, pancreas, adrenals and pituitary. At 24 hours, concentrations were highest in the intestinal tract, probably indicating biliary excretion. Since the concentrations in the tissues in general were lower than in plasma, no specific accumulation of LAS occurred. When ¹⁴C-LAS was injected into the skin, most of the radioactivity remained at the site of injection. No localization of radioactivity in any tissue occurred. No unchanged LAS was detected in urine samples after oral or subcutaneous doses (either single or repeated). Five metabolites were excreted but they were not identified. Incubations with beta-glucuronidase/sulfatase did not affect the metabolites, indicating that the metabolites were probably not present as the corresponding conjugates.

Test substance: Alkyl benzene sulfonate, sodium salt; mean molecular weight 349 (supplied by the Japan Soap and Detergent Association)

Reference: Cresswell, D.G., Baldock, G.A., Chasseaud, L.F. and Hawkins, D.R. 1978. Toxicological studies of linear alkylbenzene sulfonate (LAS) in rhesus monkeys: (II) the disposition of [¹⁴C] LAS after oral or subcutaneous administration. Toxicology 11:5-17.

Reliability: 2 Valid with restrictions

(e)

Type: Toxicokinetics

Results: Rats were dosed orally with ¹⁴C-Na-LAS and radioactivity was detected 0.25 hr after administration, reaching a maximum at 2 hrs. The biologically half lives were calculated to be 10.9 hrs. The distribution was high in the digestive tract and in the bladder at 4 hours after administration. Concentrations were also high in the liver, kidney, testis, spleen and lung. 168 hours after the administration, the rates of excreted radioactivity were 47% in the urine and 50% in the faeces

Remarks: Information as cited in the IUCLID Data Sheet and the IPCS document.

Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.

2) Sunakawa, T., Ikida, Y. and Okamoto, K. 1979. Absorption, distribution, metabolism, and excretion of linear alkylbenzene sulfonate in rats. J. Jpn. Oil Chem. Soc. 39:59-68 (in Japanese); cited in: IPCS (1996); Environmental Health Criteria 169: Linear Alkylbenzene Sulfonates (LAS) and Related Compounds. WHO, Geneva, Switzerland.

Reliability: 4 This study is assigned a reliability score of 4 because the original report was not available for review. However, the study was evaluated by IPCS prior to inclusion in their criteria document.

(f)

Type: Toxicokinetics
Method: Studies were conducted with isolated human skin preparations as well as *in vivo* investigations of percutaneous administration of LAS to rats. Two C₁₂ LAS solutions were tested: a 3 mM solution in 25% v/v polyethylene glycol 400 in water, and a 3 mM suspension in water prepared by homogenizing and equilibration in an all-glass homogenizer.
Results: No radioactivity was detected in urine or faeces.
Test substance: LAS (CAS #25155-30-0); activity: >99%
Remarks: These studies demonstrated that penetration through skin and subsequent systemic absorption of this surfactant does not occur to any significant extent at 24 to 48 hrs.
Reference: Howes, D. 1975. The percutaneous absorption of some anionic surfactants. J. Soc. Cosmet. Chem. 26:47-63.
Reliability: 2 Valid with restrictions

(g)

Type: *in vitro* studies with fertilised eggs
Method: Eggs from B6 x C3F₁ female mice, which were fertilised *in vitro* with sperm from C3 x 101F₁ male mice, were treated with LAS for 1 hour at the pronucleus stage and then cultivated for 5 days.
Results: Eggs treated with LAS at concentrations of less than 0.025% developed to the blastocyst stage as well as the untreated ones. At higher concentrations no egg developed beyond the 1-cell stage. The group that was treated with natural soap had no effect up to a concentration of 0.05%.
Test substance: Commercial LAS detergent (Japan)
Remarks: The authors suggest that LAS interrupts mouse pregnancy by killing fertilized eggs, however, the relevance of the results obtained in this assay for the *in vivo* situation has not been proven.
Reference: Ishii, Y., Samejima, Y., Saji, F. and Nomura, T. 1990. Effect of alcohol sulfate, linear alkylbenzene sulfonate, and natural soap on the development of fertilized eggs of the mouse *in vitro*. Mutat. Res. 242:151-155.
Reliability: 2 Valid with restrictions

5.11 EXPERIENCE WITH HUMAN EXPOSURE

(a)

Type: Human Repeat Insult Patch Test
Number of Subjects: 95 (at completion)
Methods: LAS was applied at 0.10% (w/v) on the upper arms of volunteers, under occlusive patch conditions. Test material was applied for 24 hours, 3 times a week, for 3 weeks during the induction period. After a 14-17-day rest, a 24-hour challenge patch was applied on the original and alternate arm sites.
Results: There was no evidence of skin sensitization on the 95 subjects who completed the test.

Test Material: LAS; activity: 30.0%
Reference: The Procter & Gamble Company, unpublished data, Report No. ISC-124-0470.
Reliability: 4 Not assignable

(b)
Type: Human Repeat Insult Patch Tests.
Number of subjects: 2,294 (exposed to LAS as a raw material)
17,887 (exposed to LAS in formulations)
Results: No evidence of skin sensitization.
Reference: Nusair, T.L., Danneman, P.J., Stotte, J., and Bay, P.H.S. 1988. Consumer products: Risk assessment process for contact sensitization. Toxicologist 8:258. (abstract).
Reliability: 4 Not assignable

(c)
Type: Occlusive epicutaneous
Results: LAS was applied at 1% once to middle Europeans for 24 hours. Test duration was 6 days. The authors concluded that LAS was sufficiently compatible to the skin.
Remarks: Information as cited in the IUCLID Data Sheet.
Reference: 1) European Commission. 2000a. Benzenesulfonic acid, C₁₀₋₁₃-alkyl derivs., sodium salts. Year 2000 CD-ROM edition.
2) Matthies, W., Henkel KgaA, unpublished data, Report No. 890356 (1989).
Reliability: 4 Not assignable

(d)
Type: Comparison of human experience to eye exposure to surfactants with animal eye irritation studies
Methods: Summaries of human manufacturing accident and consumer accident eye irritation incidents over several years were collected for laundry, household and personal cleaning products. These summaries included the date the incident occurred, the exact product or formulation involved, the estimated time for the eyes to return to normal, and a brief description of the eye response. A total of 231 manufacturing employee incidents and 284 consumer incidents were usable, covering 24 and 23 different products, respectively. The results of these human contact incidents were compared to the results of studies conducted using two rabbit eye irritation procedures commonly used to assess eye irritation. These two methods are briefly summarized below:

1. The FHSA (modified Draize) test utilized albino rabbits, which were dosed into the conjunctival sac with 0.1 mL of liquid product or the weight of the solid product equivalent to 0.1 cc. The eyelids were held shut for one second after instillation. The animals were observed at 1, 2, 3, 4, 7, 14 and 21 days or longer.
2. The Griffith low-volume eye irritation test utilized albino rabbits, with the test substances dosed directly on the cornea with 0.01 mL of liquid product or the weight of solids equivalent to 0.01 cc. The eyelid was released immediately after dosing without forced closing. The animals were observed for the same time periods as above.

Results:	Median days-to-clear for human accident eye exposure are minimal. Only one product was as high as 7 days and the rest were 2 days or less. A total of 88.1% of the eyes cleared in 4 days or less. There was no reported permanent eye damage. Both of the animal methods produced more severe eye responses than were reported from human eye accidents with the same consumer products (Freeberg et al. 1984).
Remarks:	Animal studies consistently overestimated the human response to accidental exposure. Of the two animal methods, the low-volume rabbit test gave a closer correlation, while the FHSA test gave the least correlation. A follow-up study published in 1986 confirmed this conclusion. Finally, an additional paper published in 1995 compared consumer eye irritation comments from 1985 to 1992 with the results of low volume eye tests (LVET). The clinical data and consumer experience consistently showed less eye irritation in humans from exposure to products than was observed in animal studies. Recovery in humans was similar to that reported previously, supporting milder irritation response and faster healing in humans than in rabbits.
Reference:	Freeberg, F.E., Griffith, J.F., Bruce, R.D., and Bay, P.H.S. 1984. Correlation of animal test methods with human experience for household products. <i>J. Toxicol.- Cut. & Ocular Toxicol.</i> 1:53-64. Freeberg, F.E., Hooker, D.T., and Griffith, J.F. 1986. Correlation of animal eye test data with human experience for household products: an update. <i>J. Toxicol.- Cut. & Ocular Toxicol.</i> 5:115-123. Cormier, E.M., Hunter, J. E., Billhimer, W., May, J., and Farage, M.A. 1995. Use of clinical and consumer eye irritation data to evaluate the low-volume eye test. <i>J. Toxicol.- Cut. & Ocular Toxicol.</i> 14:197-205.
Reliability:	2 Valid with restrictions
(e) Type: Methods:	Characterization of aerosols generated from a consumer spray product The study was designed to evaluate size distribution of aerosols suspended in air after normal use of consumer spray products. Size distribution of aerosols generated from six different consumer trigger spray product nozzles was measured using a laser diffraction particle sizer (Mastersizer Model X, Malvern Instruments Ltd). A 300 mm receiving lens was used, which covers a particle size range of 1.2-600 microns. The exit of the trigger sprayer was positioned at 20 mm from the lens to the center of the device to avoid vignetting, and 120 mm from the laser beam axis to the tip of the trigger sprayer to avoid its interference with the laser beam. Measurements were repeated 5 times for each sprayer.
Results:	The overall mean (n=30) particle size is 0.11% particles under 10 microns, with a standard deviation of 0.21. The very highest observation was 0.80%. Under normal use conditions, the peak breathing zone concentration under 10 microns ranged from 0.13 to 0.72 mg/m ³ .
Remarks:	This testing only captured the spray particles that are under 600 microns, so the actual percentage of total volume sprayed is less than 0.1%.
Reference:	Battelle. 1999. Measurement and Characterization of Aerosols Generated from a Consumer Spray Product – Pilot Study. Prepared for The Soap and Detergent Association. Battelle Study No. N003043A, January 18, 1999.
Reliability:	1 Valid without restriction

(f)

Type:

Modeling of dose observed from inhalation of aerosols

Methods:

The worst case air concentration of LAS resulting from use in surface cleaning spray products was modeled using methods recommended by the HERA Guidance Document (06/2001). In this modeling, HERA reports the results of experimental measurements of the concentration of aerosol particles from a 2001 Procter & Gamble study. The following algorithm was used to model the absorbed dose:

$$\text{Exp}_{\text{sys}} = F_1 \cdot C' \cdot Q_{\text{inh}} \cdot t \cdot n \cdot F_7 \cdot F_8 / \text{bw} \text{ (mg/kg bw/day)}$$

Where:

Exp_{sys} = dose absorbed via inhalation

F_1 = weight fraction of substance in product = 6% (worst case assumption)

C' = product concentration = 0.35 mg/m³

Q_{inh} = ventilation rate of user = 0.8 m³/hr

t = duration of exposure = 0.17 hr (10 minutes)

n = product use frequency, in number of events per day = 1

F_7 = weight fraction respirable = 100%

F_8 = weight fraction absorbed or bioavailable = 75%

bw = body weight = 60 kg

Results:

The modeling resulted in an Exp_{sys} (inhalation of aerosols) = 0.04 µg/kg bw/day. Measured aerosol particles under 6.4 microns in size were generated upon spraying with typical surface cleaning spray products, resulting in a product concentration of 0.35 mg/m³.

Reference:

HERA. 2002. HERA-LAS Human and Environmental Risk Assessment: Linear Alkylbenzene Sulphonates, LAS. CAS No. 68411-30-3, Draft, July 2002. <http://www.heraproject.com/riskassessment.cfm>

Reliability:

4 This score was assigned because the original Procter & Gamble study and the HERA model inputs were not available for review. However, the study and all assumptions were evaluated by HERA.

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APPENDIX A

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