



Committee for Risk Assessment
RAC

Annex 1

Background document

to the Opinion proposing harmonised classification
and labelling at Community level of

**Chlorsulfuron (ISO); 2-chloro-N-[[(4-methoxy-6-methyl-
1,3,5- triazin-2-
yl)amino]carbonyl]benzenesulphonamide**

EC number: 265-268-5
CAS number: 64902-72-3

CLH-O-0000001412-86-48/F

The background document is a compilation of information considered relevant by the dossier submitter or by RAC for the proposed classification. It includes the proposal of the dossier submitter and the conclusion of RAC. It is based on the official CLH report submitted to public consultation. RAC has not changed the text of this CLH report but inserted text which is specifically marked as 'RAC evaluation'. Only the RAC text reflects the view of RAC.

Adopted

04 December 2014

CLH report

Proposal for Harmonised Classification and Labelling

**Based on Regulation (EC) No 1272/2008 (CLP Regulation),
Annex VI, Part 2**

Substance Name: Chlorsulfuron

EC Number: 265-268-5

CAS Number: 64902-72-3

Index Number: 613-121-00-4

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CONTENTS

PART A.	6
1	PROPOSAL FOR HARMONISED CLASSIFICATION AND LABELLING..... 6
1.1	SUBSTANCE..... 6
1.2	HARMONISED CLASSIFICATION AND LABELLING PROPOSAL 6
1.3	PROPOSED HARMONISED CLASSIFICATION AND LABELLING BASED ON CLP REGULATION CRITERIA... 7
2	BACKGROUND TO THE CLH PROPOSAL 10
2.1	HISTORY OF THE PREVIOUS CLASSIFICATION AND LABELLING..... 10
2.2	SHORT SUMMARY OF THE SCIENTIFIC JUSTIFICATION FOR THE CLH PROPOSAL 10
2.3	CURRENT HARMONISED CLASSIFICATION AND LABELLING..... 10
2.3.1	CURRENT CLASSIFICATION AND LABELLING IN ANNEX VI, TABLE 3.1 IN THE CLP REGULATION 10
2.3.2	CURRENT CLASSIFICATION AND LABELLING IN ANNEX VI, TABLE 3.2 IN THE CLP REGULATION 10
2.4	CURRENT SELF-CLASSIFICATION AND LABELLING 11
2.4.1	<i>Current self-classification and labelling based on the CLP Regulation criteria</i> 11
3	JUSTIFICATION THAT ACTION IS NEEDED AT COMMUNITY LEVEL 11
	PART B. SCIENTIFIC EVALUATION OF THE DATA 12
1	IDENTITY OF THE SUBSTANCE 12
1.1	NAME AND OTHER IDENTIFIERS OF THE SUBSTANCE..... 12
1.2	COMPOSITION OF THE SUBSTANCE 12
1.2.1	<i>Composition of test material</i> 14
1.3	PHYSICO-CHEMICAL PROPERTIES 15
2	MANUFACTURE AND USES..... 19
2.1	MANUFACTURE 19
2.2	IDENTIFIED USES 19
3	CLASSIFICATION FOR PHYSICO-CHEMICAL PROPERTIES 19
4	HUMAN HEALTH HAZARD ASSESSMENT 19
4.1	<i>Toxicokinetics (absorption, metabolism, distribution and elimination)</i> 19
4.1.1	<i>Non-human information</i> 19
4.1.2	<i>Human information</i> 21
4.1.3	<i>Summary and discussion on toxicokinetics</i> 21
4.2	ACUTE TOXICITY 21
4.3	SPECIFIC TARGET ORGAN TOXICITY – SINGLE EXPOSURE (STOT SE)..... 21
4.4	IRRITATION 21
4.4.1	<i>Skin irritation</i> 21
4.4.2	<i>Eye irritation</i> 21
4.4.3	<i>Respiratory tract irritation</i> 22
4.5	CORROSIVITY 22
4.6	SENSITISATION 22

4.7	REPEATED DOSE TOXICITY	22
4.8	SPECIFIC TARGET ORGAN TOXICITY (CLP REGULATION) – REPEATED EXPOSURE (STOT RE).....	22
4.9	GERM CELL MUTAGENICITY (MUTAGENICITY).....	22
4.10	CARCINOGENICITY	22
4.11	TOXICITY FOR REPRODUCTION	22
4.12	OTHER EFFECTS.....	22
5	ENVIRONMENTAL HAZARD ASSESSMENT	22
5.1	DEGRADATION	22
5.1.1	<i>Stability</i> 25	
5.1.2	<i>Biodegradation</i>	26
5.1.2.1	Biodegradation estimation	26
5.1.2.2	Screening tests	26
5.1.2.3	Simulation tests.....	26
5.1.3	<i>Summary and discussion of degradation</i>	29
5.2	ENVIRONMENTAL DISTRIBUTION.....	30
5.2.1	<i>Adsorption/Desorption</i>	30
5.2.2	<i>Volatilisation</i>	30
5.2.3	<i>Distribution modelling</i>	30
5.3	AQUATIC BIOACCUMULATION	31
5.3.1	<i>Aquatic bioaccumulation</i>	31
5.3.1.1	Bioaccumulation estimation.....	31
5.3.1.2	Measured bioaccumulation data.....	31
5.3.2	<i>Summary and discussion of aquatic bioaccumulation</i>	31
5.4	AQUATIC TOXICITY	31
5.4.1	<i>Fish</i> 34	
5.4.1.1	Short-term toxicity to fish	34
5.4.1.2	Long-term toxicity to fish	35
5.4.2	<i>Aquatic invertebrates</i>	36
5.4.2.1	Short-term toxicity to aquatic invertebrates	36
5.4.2.2	Long-term toxicity to aquatic invertebrates	37
5.4.3	<i>Algae and aquatic plants</i>	38
5.4.4	<i>Other aquatic organisms (including sediment)</i>	42
5.5	COMPARISON WITH CRITERIA FOR ENVIRONMENTAL HAZARDS (SECTIONS 5.1 – 5.4).....	42
5.6	CONCLUSIONS ON CLASSIFICATION AND LABELLING FOR ENVIRONMENTAL HAZARDS (SECTIONS 5.1 – 5.4) 43	
6	OTHER INFORMATION.....	52
7	REFERENCES	53
8	ANNEXES.....	58

Part A.

1 PROPOSAL FOR HARMONISED CLASSIFICATION AND LABELLING

1.1 SUBSTANCE

Table 1: Substance identity.

Substance name:	Chlorsulfuron; <u>2-chloro-N-[[4-methoxy-6-methyl-1,3,5-triazin-2-yl]amino]carbonyl]benzenesulphonamide</u>
EC number:	265-268-5
CAS number:	64902-72-3
Annex VI Index number:	613-121-00-4
Degree of purity:	≥960 g/kg
Impurities:	No relevant impurities for classification ¹ .

1.2 HARMONISED CLASSIFICATION AND LABELLING PROPOSAL

Table 2: The current Annex VI entry and the proposed harmonised classification.

	CLP Regulation
Current entry in Annex VI, CLP Regulation	Aquatic Acute 1, H400 Aquatic Chronic 1, H410
Current proposal for consideration by RAC	Aquatic Acute 1, H400 Aquatic Chronic 1, H410 M-factor (acute): 1000 M-factor (chronic): 100
Resulting harmonised classification (future entry in Annex VI, CLP Regulation)	Aquatic Acute 1, H400 Aquatic Chronic 1, H410 M-factor (acute): 1000 M-factor (chronic): 100

¹ SANCO/198/08-final from 28 September 2010

1.3 PROPOSED HARMONISED CLASSIFICATION AND LABELLING BASED ON CLP REGULATION CRITERIA

Table 3: Proposed classification according to the CLP Regulation.

CLP Annex I ref	Hazard class	Proposed classification	Proposed SCLs and/or M-factors	Current classification ¹⁾	Reason for no classification ²⁾
2.1.	Explosives	No classification	Not applicable	None	Conclusive but not sufficient for classification
2.2.	Flammable gases	No classification	Not applicable	None	Data lacking
2.3.	Flammable aerosols	No classification	Not applicable	None	Data lacking
2.4.	Oxidising gases	No classification	Not applicable	None	Data lacking
2.5.	Gases under pressure	No classification	Not applicable	None	Data lacking
2.6.	Flammable liquids	No classification	Not applicable	None	Data lacking
2.7.	Flammable solids	No classification	Not applicable	None	Conclusive but not sufficient for classification
2.8.	Self-reactive substances and mixtures	No classification	Not applicable	None	Data lacking
2.9.	Pyrophoric liquids	No classification	Not applicable	None	Data lacking
2.10.	Pyrophoric solids	No classification	Not applicable	None	Conclusive but not sufficient for classification
2.11.	Self-heating substances and mixtures	No classification	Not applicable	None	Conclusive but not sufficient for classification
2.12.	Substances and mixtures which in contact with water emit flammable gases	No classification	Not applicable	None	Conclusive but not sufficient for classification
2.13.	Oxidising liquids	No classification	Not applicable	None	Data lacking
2.14.	Oxidising solids	No classification	Not applicable	None	Conclusive but not sufficient for classification
2.15.	Organic peroxides	No classification	Not applicable	None	Data lacking
2.16.	Substance and mixtures corrosive to metals	No classification	Not applicable	None	Data lacking

CLP Annex I ref	Hazard class	Proposed classification	Proposed SCLs and/or M-factors	Current classification ¹⁾	Reason for no classification ²⁾
3.1.	Acute toxicity - oral	No classification	Not applicable	None	Conclusive but not sufficient for classification
	Acute toxicity - dermal	No classification	Not applicable	None	Conclusive but not sufficient for classification
	Acute toxicity - inhalation	No classification	Not applicable	None	Conclusive but not sufficient for classification
3.2.	Skin corrosion / irritation	No classification	Not applicable	None	Conclusive but not sufficient for classification
3.3.	Serious eye damage / eye irritation	No classification	Not applicable	None	Conclusive but not sufficient for classification
3.4.	Respiratory sensitisation	No classification	Not applicable	None	Data lacking
3.4.	Skin sensitisation	No classification	Not applicable	None	Conclusive but not sufficient for classification
3.5.	Germ cell mutagenicity	No classification	Not applicable	None	Conclusive but not sufficient for classification
3.6.	Carcinogenicity	No classification	Not applicable	None	Conclusive but not sufficient for classification
3.7.	Reproductive toxicity	No classification	Not applicable	None	Conclusive but not sufficient for classification
3.8.	Specific target organ toxicity –single exposure	No classification	Not applicable	None	Conclusive but not sufficient for classification
3.9.	Specific target organ toxicity – repeated exposure	No classification	Not applicable	None	Conclusive but not sufficient for classification
3.10.	Aspiration hazard	No classification	Not applicable	None	Data lacking
4.1.	Hazardous to the aquatic environment	Aquatic Acute 1; H400 Aquatic Chronic 1; H410	M-factor (acute): 1000 M-factor (chronic): 100	Aquatic Acute 1; H400 Aquatic Chronic 1; H410	-
5.1.	Hazardous to the ozone layer	No classification	Not applicable	None	Data lacking

¹⁾ Including specific concentration limits (SCLs) and M-factors

²⁾ Data lacking, inconclusive, or conclusive but not sufficient for classification

Labelling:

Signal word: Warning

Hazard pictogram:

GHS09: Environment



Hazard statement:

H410: Very toxic to aquatic life with long lasting effects

Precautionary statements: No precautionary statements are proposed since precautionary statements are not included in Table 3.1 of Annex VI of Regulation EC no. 1272/2008.

Proposed notes assigned to an entry:

None

2 BACKGROUND TO THE CLH PROPOSAL

2.1 HISTORY OF THE PREVIOUS CLASSIFICATION AND LABELLING

Chlorsulfuron was approved in 2009 for Annex I listing under Council Directive 91/414/EEC, with Greece as Rapporteur Member State.

Chlorsulfuron is listed in Annex VI of the Regulation (EC) No 1272/2008 of the European Parliament and of the Council of 16 December 2008 on classification, labelling and packaging of substances and mixtures (CLP Regulation) with the classifications as Aquatic Acute 1; H400 and Aquatic Chronic 1; H410.

2.2 SHORT SUMMARY OF THE SCIENTIFIC JUSTIFICATION FOR THE CLH PROPOSAL

Physico-chemical properties

For chlorsulfuron, no re-evaluation of classification and labelling has been proposed regarding physical and chemical properties.

Human Health

For chlorsulfuron, no re-evaluation of classification and labelling has been proposed regarding human health.

A proposal for Car. Cat. 2; H351, limited evidence of a carcinogenic response, was made in the EFSA review based on an increase in unilateral Leydig Cell Tumours. The Dossier Notifier (DS) evaluated the information presented in the assessment of chlorsulfuron under Directive 91/414/EEC and information submitted in regard to carcinogenicity by DuPont that, to the industry knowledge, was not reviewed. Based on the fact that there was no statistically significant increase in the overall (unilateral and bilateral) tumour incidence, the DS thinks that this proposed classification is not warranted.

Environment

According to available data chlorsulfuron should be classified as Aquatic Acute 1; H400 and Aquatic Chronic 1; H410 as it is not readily biodegradable and due to its acute effect on algal/aquatic plants at a concentration ≤ 1 mg a.s./L.

The 2nd ATP to CLP Regulation (Commission Regulation (EU) No 286/2011 of 10 March 2011 amending, for the purposes of its adaptation to technical and scientific progress, Regulation (EC) No 1272/2008) brought in new criteria for classification of long-term hazards to the aquatic environment (e.g. use of chronic toxicity data in classification and separate M factors for acute and chronic toxicity). The environmental hazard assessment was performed in order to determine the acute M-factor and chronic M-factor, currently not included in Annex VI of CLP Regulation.

2.3 CURRENT HARMONISED CLASSIFICATION AND LABELLING

2.3.1 CURRENT CLASSIFICATION AND LABELLING IN ANNEX VI, TABLE 3.1 IN THE CLP REGULATION

Aquatic Acute 1; H400

Aquatic Chronic 1; H410

2.3.2 CURRENT CLASSIFICATION AND LABELLING IN ANNEX VI, TABLE 3.2 IN THE CLP REGULATION

N; R50/53

2.4 CURRENT SELF-CLASSIFICATION AND LABELLING

2.4.1 Current self-classification and labelling based on the CLP Regulation criteria

According to the information found in C&L Inventory database (database contains classification and labelling information on notified and registered substances received from manufacturers and importers; <http://www.echa.europa.eu/web/guest/information-on-chemicals/cl-inventory-database>), 64 companies notified the classification and labelling of chlorsulfuron. All companies, which notified classification and labelling according to the requirements of article 40 of CLP Regulation, classify chlorsulfuron as:

Aquatic Acute 1, H400

Aquatic Chronic 1, H410

3 JUSTIFICATION THAT ACTION IS NEEDED AT COMMUNITY LEVEL

No need for justification (cf. Article 36(3) CLP Regulation as chlorsulfuron is an active substance regulated by Regulation (EC) 1107/2009.

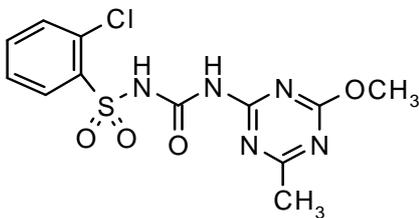
Part B.

SCIENTIFIC EVALUATION OF THE DATA

1 IDENTITY OF THE SUBSTANCE

1.1 NAME AND OTHER IDENTIFIERS OF THE SUBSTANCE

Table 4: Substance identity.

EC number:	265-268-5
EC name:	Chlorsulfuron
CAS number:	64902-72-3
CAS name:	2-chloro- <i>N</i> -[[[4-methoxy-6-methyl-1,3,5-triazin-2-yl)amino]carbonyl]benzenesulfonamide
IUPAC name:	1-(2-chlorophenylsulfonyl)-3-(4-methoxy-6-methyl-1,3,5-triazin-2-yl)urea
CLP Annex VI Index number:	613-121-00-4
Molecular formula:	C ₁₂ H ₁₂ ClN ₅ O ₄ S
Molecular weight range:	357.8 g/mol
Structural formula:	

1.2 COMPOSITION OF THE SUBSTANCE

Table 5: Constituents (non-confidential information).

Constituent	Typical concentration	Concentration range	Remarks
Chlorsulfuron	≥960 g/kg	No range, since minimal purity stated	-

Current Annex VI entry: chlorsulfuron

Table 6: Impurities (non-confidential information).

Impurity	Typical concentration	Concentration range	Remarks
No relevant impurities for classification (according to SANCO/198/08-final from 28 September 2010)	-		The chlorsulfuron specifications were considered provisional at the time of inclusion into Annex I of 91/414 EEC. Hence, in December 2009 an application for establishment a final reference specification for chlorsulfuron technical was submitted to the evaluating MS France. France issued an addendum to the DAR reviewing the reference specifications including a proposal for removal of the impurities IN-A4097 (chlorosulfonamide) and IN-A4098 (4-methoxy-6-methyl-1,3,5-triazine-2-amine) as toxicological relevant impurities in April 2010 (Chlorsulfuron Addendum 2, Annex C after Annex I listing. Report on the finalisation of the reference specifications). The amended specifications and petition for removal of the impurities IN-A4097 (chlorosulfonamide) and IN-A4098 (4-methoxy-6-methyl-1,3,5-triazine-2-amine) as toxicological relevant were noted at the Standing Committee on the Food Chain and Animal Health on 28 September 2010

Current Annex VI entry: None

Table 7: Additives (non-confidential information).

Additive	Function	Typical concentration	Concentration range	Remarks
No additives	-	-	-	-

Current Annex VI entry: None

1.2.1 Composition of test material

The purity of the test substance is given in each test description when appropriate. Ranges are as follow:

Physico-chemical properties tests: 97.18 to 99.93%

Mammalian toxicology: 93.8 to ~100%

Ecotoxicology: 95.4 to 99.5%

The purity of the samples used for physico-chemical, toxicological and ecotoxicological testing are in the range from 93.8 to 100%.

Initial toxicological testing was done with laboratory samples of the active substance as no industrial process was in place during the early testing days of chlorsulfuron and may therefore have lower purity than the current specifications requires.

1.3 PHYSICO-CHEMICAL PROPERTIES

The following summary information was extracted from the Draft Assessment Report Vol. 3, Annex B.2 'Physical and chemical properties'.

Table 8: Summary of physico - chemical properties.

Property	Value	Reference	Comment (e.g., measured or estimated)
State of the substance at 20°C and 101.3 kPa	Solid		
Physical state, colour, odour GLP	Both technical and PAI material are white, crystalline solids. They both have a slight organic solvent (xylene) odour due to small amounts (<0.5%) of residual solvent remaining in the solid.	Moore, L.A. 2003b (DuPont-10643, Revision No. 1), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.7, B.2.1.8, B.2.1.9	chlorsulfuron purity: 98.0% - 98.5%
Melting/freezing point GLP	Average melting point range: 171.5–174°C	Moore, L.A. 2003b (DuPont-10643, Revision No. 1), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.1	chlorsulfuron purity: 98.0% - 98.5%
Boiling point	Not applicable		Chlorsulfuron is a solid at ambient temperature.
Temperature of decomposition GLP	As the melting point samples approached liquification, the white powder turned yellow throughout the melt. Thermogravimetric analysis demonstrated weight loss starting at 150°C, which continued during and after the melt. Differential Scanning Calorimetry showed an endothermic transition corresponding to decomposition of the material after the melt.	Moore, L.A. 2003b (DuPont-10643, Revision No. 1), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.3	chlorsulfuron purity: 98.5%
Relative density GLP	1.48 g/mL (at 23°C) 1.495 ± 0.001 g/mL (at 20.5 ± 0.5°C)	Moore, L.A. 2003b (DuPont-10643, Revision No. 1) Huntley, K. 2001b (DuPont-6282), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.4	chlorsulfuron purity: 97.18% - 98.5%

Property	Value	Reference	Comment (e.g., measured or estimated)
Vapour pressure	2.3 ± 10^{-11} Torr equal to 3.1 ± 10^{-9} Pa at 25°C (extrapolated from measurements between 132.2°C and 167.5°C)	Schmuckler, M.E. 1992a (G/PC-22-CA), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.5	chlorsulfuron purity: 99.1%
Volatility, Henry's law constant	Calculated values at 20°C: 5.0×10^{-10} Pa \times m ³ \times mol ⁻¹ at pH 5 3.5×10^{-11} Pa \times m ³ \times mol ⁻¹ at pH 7 3.2×10^{-12} Pa \times m ³ \times mol ⁻¹ at pH 9	Moore, L.A. 2002a (DuPont-7217), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.6	Calculation Chlorsulfuron has very low tendency of volatilization from aqueous solutions.
Surface tension GLP	73.1 ± 0.9 mN/m at $20.5 \pm 0.06^\circ\text{C}$	Huntley, K. 2001a (DuPont-6281), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.24	Chlorsulfuron is not characterized as a surface-active substance. purity: 97.18%
Water solubility GLP	Solubility at 20°C and pH 5: 0.876 g/L pH 7: 12.5 g/L pH 9: 134 g/L	Huntley, K. 2002 (DuPont-6284), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.12	chlorsulfuron purity: 99.4%
Partition coefficient n-octanol/water	The study determined the n-octanol/water partition coefficient (P_{ow}) of chlorsulfuron at two concentrations (1 ppm & 10 ppm) and at pH 5, 7 and 9. The values found at 25°C are: pH 5: $P_{ow} = 2.13$; $\log P_{ow} = 0.33$ pH 7: $P_{ow} = 0.102$; $\log P_{ow} = -0.99$ pH 9: $P_{ow} = 0.0387$; $\log P_{ow} = -1.41$	Schmuckler, M.E. 1992b (AMR 1224-88, Revision No. 1), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.14	(Values are the average of measurements at the two concentrations 1 & 10 ppm) Radiochemical purity of carbonyl ¹⁴ C-chlorsulfuron: 96% Chemical purity of chlorsulfuron: approx. 100% Specific activity: 76.8 $\mu\text{Ci}/\text{mg}$
Flash point	Not applicable		Not applicable as the melting point of chlorsulfuron is not below 40°C.
Flammability GLP	No propagation occurred. Chlorsulfuron is not considered highly flammable under the conditions of the test.	Gravell, R.L. 2001 (DuPont-4478, Revision No. 1), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.20	chlorsulfuron purity: 97.18%
Explosive properties GLP	Thermal sensitivity (effect of a flame): negative	Gravell, R.L. 2001	chlorsulfuron purity: 97.18%

Property	Value	Reference	Comment (e.g., measured or estimated)																		
	Mechanical sensitivity (shock): negative Mechanical sensitivity (friction): negative	(DuPont-4478, Revision No. 1), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.23	Chlorsulfuron is not expected to have explosive properties.																		
Self-ignition temperature GLP	Chlorsulfuron has no self-ignition temperature.	Gravell, R.L. 2001 (DuPont-4478, Revision No. 1), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.21	chlorsulfuron purity: 97.18%																		
Oxidising properties	Considering the molecular structure and the composition of chlorsulfuron, oxidizing properties are not expected.	Moore, L.A. 2004 (DuPont-14653), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.25	Structural argument																		
Granulometry	According to Regulation (EC) 1107/2009 granulometry is not required for active substances. Thus, no study or end-point has been provided.																				
Stability in organic solvents and identity of relevant degradation products	No data on stability.																				
Solubility in organic solvents GLP	Solubilities at 20°C: <table style="margin-left: auto; margin-right: auto;"> <thead> <tr> <th style="text-align: left;">Solvent</th> <th style="text-align: left;">Solubility (g/L)</th> </tr> </thead> <tbody> <tr> <td>n-hexane</td> <td>0.0024</td> </tr> <tr> <td>isopropanol</td> <td>1.6</td> </tr> <tr> <td>toluene</td> <td>2.8</td> </tr> <tr> <td>methanol</td> <td>15</td> </tr> <tr> <td>acetonitrile</td> <td>21</td> </tr> <tr> <td>ethyl acetate</td> <td>25</td> </tr> <tr> <td>acetone</td> <td>37</td> </tr> <tr> <td>dichloromethane</td> <td>140</td> </tr> </tbody> </table>	Solvent	Solubility (g/L)	n-hexane	0.0024	isopropanol	1.6	toluene	2.8	methanol	15	acetonitrile	21	ethyl acetate	25	acetone	37	dichloromethane	140	Moore, L.A. 1992 (AMR 2375-92), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.13	chlorsulfuron purity: 99.93% Chlorsulfuron is readily soluble in all organic solvents except n-hexane in which it is slightly soluble.
Solvent	Solubility (g/L)																				
n-hexane	0.0024																				
isopropanol	1.6																				
toluene	2.8																				
methanol	15																				
acetonitrile	21																				
ethyl acetate	25																				
acetone	37																				
dichloromethane	140																				
Dissociation constant GLP	$pK_a = 3.4$	Schmuckler, M.E., Moore, L.A., Wilds, J.C. 1992 (AMR 2367-92), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.18	chlorsulfuron purity: 99.93%																		
Stability in water Hydrolysis rate of purified a.s.	Hydrolysis of chlorsulfuron was significant only at pH 5 with a first order half-life of approximately 23 days at 25°C. At pH 7 and 9, chlorsulfuron	Dietrich, R.F., McAleer, N.C. 1989 (AMR 1455-89.161-1),	A 31-day test was conducted with two radiolabeled forms of chlorsulfuron at pH 5, 7 and 9 in sterilized buffered																		

Property	Value	Reference	Comment (e.g., measured or estimated)
	was essentially stable comprising 96-99% of the total applied radioactivity after 31 days of incubation at 25°C.	reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.15	aqueous solutions kept in darkness at 25°C. Chlorsulfuron is moderately hydrolyzing at pH 5 and slightly hydrolyzing at pH 7 and 9.
Stability in water Photochemical degradation of purified a.s Direct photo-transformation	At pH 7 and pH 9, chlorsulfuron was quite stable in both irradiated and non-irradiated solutions tested, with >95% of recovered radioactivity present as chlorsulfuron after 31 days. At pH 5, chlorsulfuron degraded with an average half-life of 23 days for the non-irradiated [phenyl(U)- ¹⁴ C]chlorsulfuron and 18 days for the irradiated [triazine-2- ¹⁴ C]chlorsulfuron solutions. The major contributing factor in the degradation of chlorsulfuron at pH 5 samples is hydrolysis.	Dietrich, R.F. McAleer, N.C. 1989 (AMR 1455-89.161-2), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.16	A 31-day test was conducted with two radiolabeled forms of chlorsulfuron at pH 5, 7 and 9 in sterilized buffered aqueous solutions exposed to summer sunlight. Chlorsulfuron is moderately photolytically degradable at pH 5. At pH 7 and 9 it is considered photolytically stable.
Quantum yield of direct photo-transformation in water	Quantum efficiency in water: 5.225×10^{-4} DT ₅₀ ranged from 89 days at 30° latitude (summer) to 2010 days at 60° latitude (winter)	Moore, L.A. 2003a (DuPont-7219), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.17	Data collected only at pH 5 as at pH 7 & 9 chlorsulfuron exhibits no measurable photodegradation in water when subjected to natural sunlight.
Stability in air, photochemical oxidative degradation Indirect photo-transformation	Overall OH rate constant: $k_{OH} = 2.5194 \times 10^{-12} \text{ cm}^3 \times \text{molecule}^{-1} \times \text{sec}^{-1}$ DT ₅₀ = 50.945 hours (using [OH] = 1.5×10^6 molecules/cm ³ and assuming 12-hour daylight)	Moore, L.A. 2002b (DuPont-7218), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.19	Atkinson calculation
Spectra of purified a.s. GLP	UV/VIS, ¹ H NMR, ¹³ C NMR, MS, and IR spectra are consistent with the proposed chemical structure of chlorsulfuron. UV/VIS spectra: No absorbance maxima (λ_{max}) above 290 nm were observed at any pH, for aqueous solutions of chlorsulfuron, at concentrations of 10.3 and 30.9 µg/mL.	Huntley, K. 2001c (DuPont-6283) Moore, L.A. 2003c (DuPont-11256), reviewed in DAR, Vol. 3, Annex B1-5, B.2.1.10	chlorsulfuron purity: 97.18% - 99.4%
Viscosity	Not applicable		

2 MANUFACTURE AND USES

2.1 MANUFACTURE

Not relevant for Classification and Labelling.

2.2 IDENTIFIED USES

Agriculture.

3 CLASSIFICATION FOR PHYSICO-CHEMICAL PROPERTIES

Not evaluated in this dossier

RAC general comment

During public consultation several Member States proposed the (re)evaluation of human health hazard classes. Since they were not addressed by the dossier submitter they were not subject to evaluation by RAC.

4 HUMAN HEALTH HAZARD ASSESSMENT

The following summary information was extracted, in part, from the Chlorsulfuron *Draft Assessment Report* Vol. 3, Annex B.6 'Toxicology and metabolism' and/or the *EFSA Scientific Report* (2008) 201, 1-107. In cases where the *Draft Assessment Report* (DAR) was not clear or contained the same information, the study summaries from the industry (DuPont) or the original reports were compared and used. New information or information that may not have been discussed in the EFSA review process are highlighted.

4.1 Toxicokinetics (absorption, metabolism, distribution and elimination)

4.1.1 Non-human information

Chlorsulfuron was rapidly absorbed after oral administration to rats, but to a lower extent in females (~71%) than in males (~84%). It was widely and uniformly distributed, and chlorsulfuron did not show any potential for bioaccumulation. Excretion occurred mainly via the urine, but also via the faeces (up to 27%). The main compound excreted was the parent, and only two minor metabolites were identified (IN-A4097 (2-chlorobenzenesulfonamide) and IN-A4098 (2-amino-4-methoxy-6-methyl-1,3,5-triazine)). The metabolic pathway is hydrolytic cleavage of the sulfonylurea bridge of the parent (Shrivastava, S.P. (1980); Hunt, O.R. (1981)).

Investigations were conducted using different dosing regimens in male and female Sprague-Dawley (CD) rats. These included administration of [phenyl(U)-¹⁴C]chlorsulfuron by:

- A) intravenous injection (0.11 mg/kg bw),
- B) a single oral gavage low dose (16 mg/kg bw),
- C) preconditioning for 21 days with 100 ppm (*ca.* 10 mg/kg bw/day) of unlabeled chlorsulfuron in the diet followed by a single oral gavage low dose (16 mg/kg bw) of radioactive chlorsulfuron, or,
- D) a single oral gavage high dose (3000 mg/kg bw)

Another investigation was conducted in male Sprague-Dawley (CD) rats that were preconditioned on a diet of 2500 ppm (*ca.* 125 mg/kg bw/day) of non-radiolabeled chlorsulfuron for 21 day followed by a single gavage dose of radiolabeled chlorsulfuron at 26.4 mg/kg bw.

Absorption

Absorption was based on a summation of radioactivity in the tissues, as well as the level of radioactivity appearing in the urine (as a surrogate for the plasma). At the low dose, absorption of chlorsulfuron represented 61-95% of the applied dose. With the high dose, oral absorption was 53-68% of the applied dose. Plasma levels were not monitored over time; however, absorption was rapid as indicated below by urinary elimination (see below). According to the *EFSA Scientific Report* (2008), slight differences in absorption were observed between males and females (~84% in males and ~71% in females). No data on dermal absorption was available for an assessment.

Excretion

The majority of the radioactivity was excreted *via* the urine within 24 hrs after low dose administration, and was essentially complete by 48 hrs. Urinary elimination was more prolonged at the high dose. Most radioactivity was eliminated via the urine, regardless of sex, dose level or duration of the test substance administration. Faeces represented a less significant elimination pathway (3.9-27.56% of the radioactivity) for all groups tested. No radioactivity was detected in the expired air. No significant differences were revealed with regard to dose level or mode of administration. As mentioned above, the elimination of chlorsulfuron based on the urinary excretion of radioactivity, was shown to be more rapid at the low dose (elimination $T_{1/2} = 7.6-15.3$ hrs) compared to the high dose (elimination $T_{1/2} = 20.1-33.33$ hrs).

Distribution

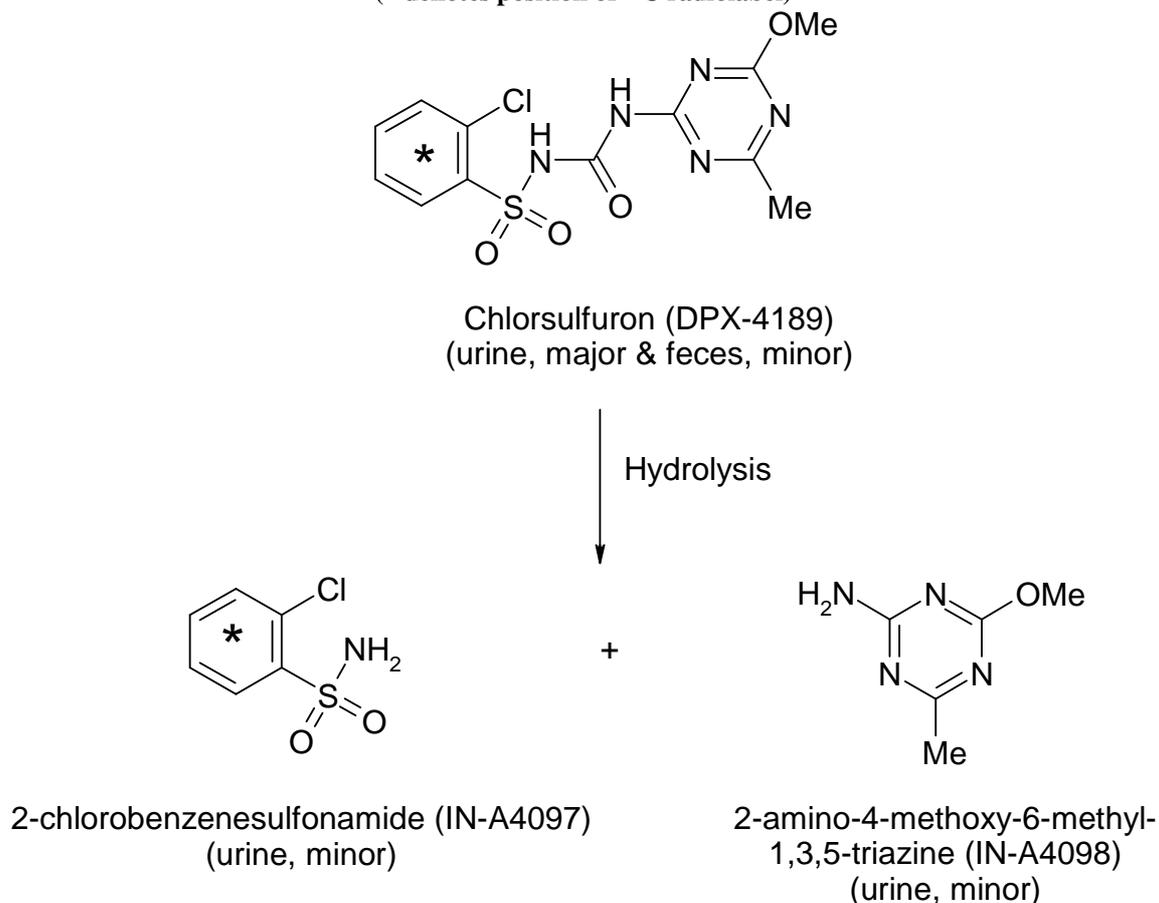
Chlorsulfuron was uniformly distributed in the rat tissues after both single oral low or high dose administration and after repeated low doses. In comparing the radioactive levels detected in the tissues and organs following single and repeated low dose administration of ^{14}C -chlorsulfuron to rats, it was concluded that there is no potential for chlorsulfuron accumulation in the rat.

Metabolism

The major portion of radioactivity excreted was found to be intact chlorsulfuron (49-76% of the radioactivity in the urine, higher values reported in the preliminary test). Only two metabolite were definitively identified: IN-A4097 (2-chlorobenzenesulfonamide) and IN-A4098 (2-amino-4-methoxy-6-methyl-1,3,5-triazine). Thus, the main metabolic pathway observed for chlorsulfuron in rats was hydrolytic cleavage of the sulfonyleurea bridge to form IN-A4097 and IN-A4098.

Total radioactive recovery in these metabolism studies ranged between 76.64 to 99.84% of the applied radioactivity (after 72 or 168 hrs).

Figure 1
Proposed metabolic pathway of chlorsulfuron in the rat
(* denotes position of ¹⁴C radiolabel)



4.1.2 Human information

No data available.

4.1.3 Summary and discussion on toxicokinetics

See Section 4.1.1.

4.2 ACUTE TOXICITY

Not evaluated in this dossier.

4.3 SPECIFIC TARGET ORGAN TOXICITY – SINGLE EXPOSURE (STOT SE)

Not evaluated in this dossier.

4.4 IRRITATION

4.4.1 Skin irritation

Not evaluated in this dossier.

4.4.2 Eye irritation

Not evaluated in this dossier.

4.4.3 Respiratory tract irritation

Not evaluated in this dossier.

4.5 CORROSIVITY

Not evaluated in this dossier.

4.6 SENSITISATION

Not evaluated in this dossier.

4.7 REPEATED DOSE TOXICITY

Not evaluated in this dossier.

4.8 SPECIFIC TARGET ORGAN TOXICITY (CLP REGULATION) – REPEATED EXPOSURE (STOT RE)

Not evaluated in this dossier.

4.9 GERM CELL MUTAGENICITY (MUTAGENICITY)

Not evaluated in this dossier.

4.10 CARCINOGENICITY

Not evaluated in this dossier.

4.11 TOXICITY FOR REPRODUCTION

Not evaluated in this dossier.

4.12 OTHER EFFECTS

Not evaluated in this dossier.

5 ENVIRONMENTAL HAZARD ASSESSMENT**5.1 DEGRADATION**

The environmental fate properties assessment for chlorsulfuron is based on the Draft Assessment Report, the Addendum to the Draft Assessment Report and the EFSA Scientific Report on the peer review of chlorsulfuron.

All the studies on the fate and behaviour of chlorsulfuron in the environment were performed under GLP and according to EPA, OECD or equivalent guidelines.

Table 9: Summary of relevant information on degradation

Property	Method	Results	Remarks	Reference
Stability Hydrolysis	U.S. EPA Pesticide Assessment Guidelines Subdivision N, 161-1	Hydrolysis of chlorsulfuron is significant only at pH 5. Chlorsulfuron is essentially stable at pH 7 and pH 9. At pH 5, first-order half-life is ~23 days at 25°C.	[Phenyl(U)- ¹⁴ C] chlorsulfuron, 11.5 µCi/mg, radiochemical purity >95% [Triazine-2- ¹⁴ C] chlorsulfuron, 15.2 µCi/mg, radiochemical	Dietrich, R.F., McAleer, N.C. (1989) AMR 1455-89.161-1 reviewed in DAR, Vol. 3,

Property	Method	Results	Remarks	Reference
			purity >95%.	Annex B8, B.8.4.1
Aqueous photolysis	U.S. EPA Pesticide Assessment Guidelines Subdivision N, 161-1	<p>Photolysis is not a major degradation process for chlorsulfuron at pH 5, pH 7, or pH 9 at 25°C.</p> <p>At pH 7 and pH 9, chlorsulfuron is quite stable in irradiated and non-irradiated solutions tested, with >95% of recovered radioactivity present as chlorsulfuron after 31 days.</p> <p>At pH 5, chlorsulfuron degrades with an average half-life of 23 days for the non-irradiated [phenyl(U)-¹⁴C]chlorsulfuron and 18 days for the irradiated [triazine-2-¹⁴C]chlorsulfuron solutions. The major contributing factor in the degradation of chlorsulfuron at pH 5 samples is hydrolysis.</p>	<p>[Phenyl(U)-¹⁴C] chlorsulfuron, 11.5 µCi/mg, Radiochemical purity >95%.</p> <p>[Triazine-2-¹⁴C] chlorsulfuron, 15.2 µCi/mg, Radiochemical purity >95%.</p>	<p>Dietrich, R.F., McAleer, N.C. (1989) AMR 1455-89.161-2 reviewed in DAR, Vol. 3, Annex B8, B.8.4.2</p>

Property	Method	Results	Remarks	Reference
Soil photolysis	U.S. EPA Pesticide Assessment Guidelines Subdivision N, 161-3 GLP	Chlorsulfuron degrades in a dry irradiated alkaline soil with DT ₅₀ and DT ₉₀ values of 62.2 and 207 days and is relatively stable in non-irradiated systems.	[Phenyl(U)- ¹⁴ C] chlorsulfuron, 11.5 µCi/mg, Radiochemical purity >95%. [Triazine-2- ¹⁴ C] chlorsulfuron, 15.2 µCi/mg, Radiochemical purity >95%.	Hawkins, D.R., Kirkpatrick, D., Dean, G.M., Mellor, S. (1990) AMR 1563-89 reviewed in DAR, Vol. 3, Annex B8, B.8.1.3.3
Biodegradation Ready biodegradability	Procedure C.4-C (Commission Regulation (EC) No 440/2008) and OECD 301B . GLP	Not readily biodegradable according to criteria of OECD 301B.	chlorsulfuron purity: 97.18%	Barnes, S.P. (2001) DuPont-6705 reviewed in DAR, Vol. 3, Annex B8, B.8.4.3.1
Aerobic water/sediment	SETAC Guideline Procedures for Assessing the Environmental Fate & Ecotoxicity of Pesticides Part 1.8.2 (1995); OECD Draft Guidelines for the Testing of Chemical under Aerobic and Anaerobic Transformation in Water-Sediment Systems, Section 3, N. 308 (April 2002). GLP SETAC Europe (1995), OECD 308 (2002), U.S. EPA 162-4 (1995), OPPTS 835.4300 (2008) GLP	Chlorsulfuron degraded in an alkaline aerobic sediment system with DT ₅₀ and DT ₉₀ values of 21 and 69 days in the water phase and 26 and 87 days in the total system. Chlorsulfuron degraded in an acidic aerobic water/sediment system with DT ₅₀ and DT ₉₀ values of 54.6 and 181.4 days in the water phase and 66.7 and 221.5 days in the total system.	Phenyl(U)- ¹⁴ C] chlorsulfuron, 61.70 µCi/mg, Radiochemical purity >95%. [Triazine-2- ¹⁴ C] chlorsulfuron, 15.2 µCi/mg, Radiochemical purity >95%. [Phenyl(U)- ¹⁴ C] chlorsulfuron: 60.39 µCi/mg Radiochemical purity 97.8% [Triazine-2- ¹⁴ C] chlorsulfuron, 51.05 µCi/mg, Radiochemical purity 98.7%.	Van-Nguyen, A., Theilacker, W. (2003) DuPont-4593 reviewed in DAR, Vol. 3, Annex B8, B.8.4.3.2 Tunink, A. (2010 b) DuPont-27915, Revision No. 2 reviewed in DAR Addendum, Vol. 3, Annex B8, B.8.5.3.2

Property	Method	Results	Remarks	Reference
Aerobic soil degradation (laboratory conditions)	SETAC (1995), OECD Guideline for Testing of Chemicals: Aerobic and Anaerobic transformation in Soil (Draft 1999) GLP	Chlorsulfuron degraded in an alkaline aerobic soil with DT ₅₀ and DT ₉₀ values of 232 and 771 days.	[Phenyl(U)- ¹⁴ C] chlorsulfuron: 61.7 µCi/mg Radiochemical purity >95% [Triazine-2- ¹⁴ C] chlorsulfuron, 66.0 µCi/mg, Radiochemical purity >95%.	Ryan, D.L., McMillan, J.A. (2002) DuPont-4498 reviewed in DAR, Vol. 3, Annex B8, B.8.1.1.1
	OECD 307 (2002), SETAC Europe (1995), OPPTS 835.4100 (2008), U.S. EPA 162-1 (1982) GLP	Chlorsulfuron degraded in five aerobic soils of varying pH and organic matter content with DT ₅₀ values ranging from 13.5 to 72.0 days and DT ₉₀ values from 44.8 to 239.1 days.	[Phenyl(U)- ¹⁴ C] chlorsulfuron: 60.39 µCi/mg Radiochemical purity 97.8% [Triazine-2- ¹⁴ C] chlorsulfuron, 51.05 µCi/mg, Radiochemical purity 98.7%.	Tunink, A. (2010 a) DuPont-27603 reviewed in DAR Addendum, Vol. 3, Annex B8, B.8.2.2.1.1
Terrestrial Field Dissipation	U.S. EPA Pesticide Assessment Guidelines Subdivision N, 164-1. GLP	Chlorsulfuron DT ₅₀ = 2.5–70.1 days; DT ₉₀ = 8.3–232.9 days	Phenyl(U)- ¹⁴ C] chlorsulfuron: 23.8 µCi/mg Radiochemical purity >95% [Triazine-2- ¹⁴ C] chlorsulfuron, 15.2 µCi/mg, Radiochemical purity >95%.	Dietrich, R.F., Taylor, G.T. (1990) AMR 1417-89 Rhodes, B.C. (1994) AMR 2266-91

5.1.1 Stability

Hydrolysis

Hydrolysis of chlorsulfuron is significant only at pH 5. The first order half-life of chlorsulfuron was ~23 days at 25°C. Chlorsulfuron was essentially stable at pH 7 and pH 9. No hydrolysis product was found at significant concentrations under the conditions of this test; 96-99% of the total applied radioactivity remained as chlorsulfuron after 31 days of incubation at ~25°C. Hydrolytic processes are not expected to be major contributing factors in the environmental degradation of chlorsulfuron at pH 7 and pH 9. The major hydrolysis products for [phenyl(U)-¹⁴C]chlorsulfuron were IN-A4097 (2-chlorobenzenesulfonamide), IN-JJ998 (N-[(N-carbamoylcarbamiidoyl)carbamiidoyl]-2-chlorobenzenesulfonamide), IN-M6957 (2-chloro-N-[(4-hydroxy-6-methyl-1,3,5-triazin-2-yl)carbamiidoyl]benzenesulfonamide) and a product identified as “ring-opened acetyl triuret chlorsulfuron”. The major hydrolysis products observed for [triazine-2-¹⁴C]chlorsulfuron were IN-A4098 (4-methoxy-6-methyl-1,3,5-triazine-2-amine), IN-F5475 (6-methyl-1,3,5-triazine-2,4-diol) (maximum 9%) and a product identified as “ring-opened acetyltriuret chlorsulfuron”.

Dissociation constant

Chlorsulfuron is a weak acid herbicide with a pKa of 3.4.

Aqueous photolysis

Photolysis is not a major degradation process for chlorsulfuron at pH 5, pH 7 or pH 9 at 25°C. At pH 7 and pH 9, chlorsulfuron was quite stable in both irradiated and non-irradiated solutions tested, with >95% of recovered radioactivity present as chlorsulfuron after 31 days. At pH 5, chlorsulfuron degraded with an average half-life of 23 days for the non-irradiated [phenyl(U)-¹⁴C]chlorsulfuron and 18 days for the irradiated [triazine-2-¹⁴C]chlorsulfuron solutions. The major contributing factor in the degradation of chlorsulfuron at pH 5 samples is hydrolysis. Direct phototransformation is not expected to be a major contributing factor in the environmental dissipation of chlorsulfuron. The quantum efficiency in water for photolysis of chlorsulfuron was estimated to be 5.225×10^{-4} . The half-life of chlorsulfuron ranged from 89 days at 30° latitude (summer) to 2010 days at 60° latitude (winter). Data was calculated for chlorsulfuron in solution at pH 5.

Soil photolysis

Chlorsulfuron degraded in a dry irradiated alkaline soil with DT₅₀ and DT₉₀ values of 62.2 and 207 days and was relatively stable in non-irradiated systems with only insignificant evolution of ¹⁴CO₂. Mineralisation for the phenyl-labeled chlorsulfuron was significant with 8.0% evolving as ¹⁴CO₂ by Day 31 in the irradiated system. Formation of IN-V7160 (1-(4-methoxy-6-methyl-1,3,5-triazine-2-yl)urea) (maximum 7%) occurred only in irradiated systems treated with triazine labeled chlorsulfuron, indicating that sunlight may contribute to the formation of this degradation product. Other principal degradation products of chlorsulfuron in the photolytic soil study (none >10%) were observed included IN-A4097 (2-chlorobenzenesulfonamide), IN-A4098 (4-methoxy-6-methyl-1,3,5-triazine-2-amine), and IN-M6957 (2-chloro-N-[(4-hydroxy-6-methyl-1,3,5-triazin-2-yl)carbamoyl]benzenesulfonamide).

5.1.2 Biodegradation

5.1.2.1 Biodegradation estimation

5.1.2.2 Screening tests

Ready biodegradability

The ready biodegradability of chlorsulfuron was assessed in the CO₂ evolution test (Modified Sturm test). Mean cumulative CO₂ production by mixtures containing chlorsulfuron was at most 4% of the theoretical value by the end of the test on Day 29. Substances are considered to be readily biodegradable in this test if CO₂ production is equal to or greater than 60% of the theoretical value within 10 days of achieving the 10% level. Chlorsulfuron does not meet this criterion and is therefore not considered to be readily biodegradable.

5.1.2.3 Simulation tests

Water/sediment systems

Two studies describing the biodegradation of chlorsulfuron in aerobic water/sediment systems have been conducted.

The first study (DuPont-4593; Van-Nguyen, A., Theilacker, W. 2003; DAR, Vol. 3, Annex B8, B.8.4.3.2) was conducted in two unique water/sediments. The first system was from Mill Stream Pond (UK) with water and sediment pH values of 8.1 and 7.8, respectively. The second system was from Cavrini Farm Pond (Italy) with water and sediment pH values of 7.5 and 8.3, respectively. The majority of the radioactivity remained initially in the water phase, but by study termination was almost evenly divided between the water and sediment phases in the Mill Stream system. In the Cavrini system the majority of the radioactivity (>80%) remained in the water phase throughout the

duration of the study. Carbon dioxide evolution from [¹⁴C]chlorsulfuron was measurable ranging from 0.7%-1.9% AR in the Mill Stream system and from 2.0-2.9% AR in the Cavrini system. From the combined data for the phenyl- and triazine labeled material for the Mill Stream system, the DT₅₀ and DT₉₀ values for chlorsulfuron were 21 and 69 days in the water phase and 26 and 87 days in the total system, respectively. For the Cavrini system, the calculated DT₅₀ and DT₉₀ values for chlorsulfuron were 282 and 973 days in the water phase and 375 and 1250 days in the total system, respectively. These values were extrapolated beyond 120 days and should be treated with caution due to the anaerobic nature of the system.

In the Mill Stream system treated with phenyl-labeled chlorsulfuron, IN-A4097 (2-chlorobenzenesulfonamide) reached a maximum of 1.1% in the water phase at Day 71 and declined slightly to 0.9% on Day 100 and reached a maximum of 2.5% in the sediment phase at study termination. IN-JJ998 (N-[(N-carbamoylcarbamimidoyl)carbamoyl]-2-chlorobenzenesulfonamide) reached a maximum of 11.4% in the water phase and 2.5%, both at study termination. IN-M6957 (2-chloro-N-[(4-hydroxy-6-methyl-1,3,5-triazin-2-yl)carbamoyl]benzenesulfonamide) reached a maximum of 22.2% at Day 70 on the water phase and 18.2% at Day 50 in the sediment phase. In the Mill Stream system treated with triazine-labeled chlorsulfuron, IN-A4098 (4-methoxy-6-methyl-1,3,5-triazine-2-amine) remained <1% in the water and sediment phases. IN-F5475 (6-methyl-1,3,5-triazine-2,4-diol), a degradation product was found at similar levels. IN-JJ998 (N-[(N-carbamoylcarbamimidoyl)carbamoyl]-2-chlorobenzenesulfonamide) reached maxima of 10.7% and 16.5% in the water and sediment phases, respectively, at study termination. IN-M6957 (2-chloro-N-[(4-hydroxy-6-methyl-1,3,5-triazin-2-yl)carbamoyl]benzenesulfonamide) reached a maximum 22.2% in the water phase at Day 71 and 21.6 at Day 50 in the sediment phase. IN-V7160 (1-(4-methoxy-6-methyl-1,3,5-triazine-2-yl)urea) remained at 0.5% or less throughout the study.

In the Cavrini system overall lower levels of degradation products were found than in the Mill Stream system, possibly a result of the generally more anaerobic conditions and lower organic content, which resulted in most of the radioactivity (>80%) remaining in the water phase. Only IN-JJ998 (N-[(N-carbamoylcarbamimidoyl)carbamoyl]-2-chlorobenzenesulfonamide) and IN-M6957 (2-chloro-N-[(4-hydroxy-6-methyl-1,3,5-triazin-2-yl)carbamoyl]benzenesulfonamide) were found at 5% or greater in the total system (only at study termination), with the majority of the compound in the water phase but at <5%.

The second study (DuPont-27915, Revision No. 2, Tunink, A., 2010; DAR Addendum, Vol. 3, Annex B8, B.8.5.3.2) was conducted in one acidic water/sediment system under aerobic conditions. This water had a measured pH of 6.0, and the sediment a pH of 4. The water phase of the test system was treated with duplicate labels of the test substance, [phenyl(U)-¹⁴C] and [triazine-2-¹⁴C]chlorsulfuron at a concentration of 0.3 µg/g water. The test systems were incubated in darkness at 20 ± 2°C. Aerobic conditions were maintained by passing a steady stream of humidified air through the test apparatus. The flow through systems was designed to trap evolved carbon dioxide (CO₂) and volatile organic compounds. The amount of radioactivity in the water layer decreased over the course of the study from a mean of 100.3% AR at Day 0 to a mean of 54.0% AR at Day 102. The amount of radioactivity in the sediment layer increased over the course of the study from a mean of 3.6% AR at Day 0 to a mean of 30.0% AR at Day 102. Non-extractable residues increased during the study, reaching a mean maximum of 9.7% AR on Day 102. The amount of ¹⁴CO₂ evolved was minimal (<3.7% AR). Since mass accountability was maintained, volatile organic traps were not sampled. The DT₅₀ and DT₉₀ values for chlorsulfuron in the acidic water phase were calculated to be 54.6 and 181.4 days, respectively. In the total system the DT₅₀ and DT₉₀ values for chlorsulfuron were 66.7 and 221.5 days, respectively.

In the phenyl system, transformation products were IN-M6957 (2-chloro-N-[(4-hydroxy-6-methyl-1,3,5-triazin-2-yl)carbamoyl]benzenesulfonamide), IN-JJ998 (N-[(N-carbamoylcarbamimidoyl)carbamoyl]-2-chlorobenzenesulfonamide), and IN-A4097 (2-chlorobenzenesulfonamide), and Triuret, reaching maximum concentrations of 18.9% AR, 2.0% AR, 10.1% AR, and 4.4% AR on Day 102, respectively. In the triazine system, transformation products

were IN-M6957 (2-chloro-N-[(4-hydroxy-6-methyl-1,3,5-triazin-2-yl)carbamoyl]benzenesulfonamide), IN-B5528 (4-amino-6-methyl-1,3,5-triazin-2-ol), IN-JJ998 (N-[(N-carbamoylcarbamimidoyl)carbamoyl]-2-chlorobenzenesulfonamide), IN-V7160 (1-(4-methoxy-6-methyl-1,3,5-triazine-2-yl)urea), IN-A4098 (4-methoxy-6-methyl-1,3,5-triazine-2-amine), and Triuret, reaching maximum concentrations of 22.4% AR (Day 102), 7.8% AR (Day 102), 2.0% AR (Day 102), 0.8% AR (Day 62), 17.8% AR (Day 102), and 13.4% AR (Day 102), respectively.

Aerobic soil metabolism

The biodegradation of ^{14}C -chlorsulfuron in soil was investigated in two studies conducted under aerobic conditions in the laboratory.

In the first study (DuPont-4498, Ryan, D.L., McMillan, J.A., 2002; DAR, Vol. 3, Annex B8, B.8.1.1.1), ^{14}C -chlorsulfuron was applied to a loam soil from Alfamen, Spain with pH of 8.3 and OC content of 2.1%. The ^{14}C -chlorsulfuron was applied at a nominal rate of 2 μg a.s./g dry soil which is equivalent to a field use rate of 300 g a.s./ha. The treated soils were maintained under aerobic conditions and removed at intervals over 120 days; the soils were extracted and analysed to determine the concentration of chlorsulfuron and metabolites remaining over time. Chlorsulfuron degraded in the aerobic soil with DT_{50} and DT_{90} values of 232 and 771 days, respectively. Mass balance was maintained throughout the study. At 120 days after treatment, chlorsulfuron accounted for approximately 66% of the recovered radioactivity. Mineralization to $^{14}\text{CO}_2$ was significant with 1.9-5.5% recovered at the end of the study. Non-extractable residues were approximately 9% at the end of the study.

In the second study (DuPont-27603, Tunink, A., 2010; DAR addendum, Vol. 3, Annex B8, B.8.2.2.1.1) chlorsulfuron was applied to five different soils. The five soils were a sandy loam from Chesapeake Farms, Maryland, U.S.A. (Mattapex #25), a heavy clay from the Lleida region of Spain, a sandy clay loam from Nambenheim, France, a sandy loam from Goch, Germany, and a sandy loam from Suchozobry, Poland. The organic matter content (Walkley-Black method) for these soils was 1.5, 3.1, 2.7, 3.1, and 1.3%, while the pH (1:1; soil: 0.01 M CaCl_2) was 4.35, 7.50, 7.01, 5.13, and 5.04, respectively. The test soils were treated with ^{14}C -chlorsulfuron at a concentration of 0.5 μg a.s./g dry weight soil and incubated under aerobic condition and darkness at $20 \pm 2^\circ\text{C}$. Soils were removed at intervals over 120 days and extracted and to determine the concentration of chlorsulfuron and metabolites remaining over time. The DT_{50} and DT_{90} values for chlorsulfuron in the five soils range from 12.4 to 71.2 d and 51.0 to 255.9 days, respectively. Mass balance was maintained (92.2% to 106.6% AR) throughout the study. Over the duration of the study, extractability generally decreased while the non-extractable residues (NER) increased. Mineralization to $^{14}\text{CO}_2$ was significant with as much as 10.7% AR measured at the end of the study.

Field soil dissipation

The dissipation behaviour of chlorsulfuron was investigated at field sites.

The first field study (AMR 1417-89, Dietrich, R.F., Taylor, G.T., 1990, DAR, Vol. 3, Annex B8, B.8.1.2.2.1) treated with ^{14}C -chlorsulfuron was conducted in Madera, California, USA on a sandy loam soil of pH 6.3 and OC (organic carbon) content of 0.2%. The ^{14}C -chlorsulfuron was applied at a nominal rate of 158 g a.s./ha. The application was made to the bare soil surface contained in steel cylinders. Chlorsulfuron degraded in a field soil with DT_{50} and DT_{90} values of 29.0 and 96.3 days, respectively. Quantifiable levels were not detected after 59 days. No radioactivity was detected below the 45-60 cm segment, with quantifiable radioactivity detected only in the 0-15 and 15-30 cm segments, showing that movement into the soil by chlorsulfuron or its degradation products was relatively insignificant.

The second field study (AMR 2266-91, Rhodes, B.C., 1994; DAR, Vol. 3, Annex B8, B.8.1.2.2.1) treated with ^{14}C -chlorsulfuron was conducted at a field site in Moscow, Idaho, USA on a silt loam soil of pH 6.1 and OC content of 1.3%. The ^{14}C -chlorsulfuron was applied at a nominal rate of

158 g a.s./ha. The application was made to the bare soil surface contained in steel cylinders. Chlorsulfuron degraded in the field soil with DT_{50} and DT_{90} values of 11.2 and 37.1 days, respectively. Quantifiable levels were not detected after 272 days. No radioactivity was detected below the 20 cm segment with one exception showing that movement into the soil by chlorsulfuron or its degradation products was relatively insignificant.

5.1.3 Summary and discussion of degradation

Stability

Chlorsulfuron is a weak acid herbicide with a pK_a of 3.4. Aqueous hydrolysis of chlorsulfuron was significant only in pH 5 buffered solutions with a calculated half-life of approximately 23 days at 25°C and essentially stable in buffered solutions at pH 7 and 9. Photolysis in buffered solution of pH 5 to pH 9 is not a major dissipation process for chlorsulfuron in the environment.

Aerobic soil degradation

Chlorsulfuron degraded in aerobic soil with DT_{50} values ranging from 13.5 to 232 days and DT_{90} values from 44.8 to 771 days. Mineralisation was significant ranging from 1.9-10.7% evolving as $^{14}CO_2$ over the study duration. *O*-demethylation of the triazine ring of chlorsulfuron and cleavage of the sulfonylurea bridge were the predominant initial mechanisms in the degradation of chlorsulfuron in aerobic soil. The principal degradation products observed were IN-A4097 (2-chlorobenzenesulfonamide), IN-A4098 (4-methoxy-6-methyl-1,3,5-triazine-2-amine), IN-JJ998 (N-[(N-carbamoylcarbamimidoyl)carbamoyl]-2-chlorobenzenesulfonamide) and IN-M6957 (2-chloro-N-[(4-hydroxy-6-methyl-1,3,5-triazin-2-yl)carbamoyl]benzenesulfonamide).

Surface water/sediment

The behaviour of chlorsulfuron was studied in aerobic water/sediments that were alkaline and acidic in nature. In alkaline water/sediment systems, IN-JJ998 (N-[(N-carbamoylcarbamimidoyl)carbamoyl]-2-chlorobenzenesulfonamide) and IN-M6957 (2-chloro-N-[(4-hydroxy-6-methyl-1,3,5-triazin-2-yl)carbamoyl]benzenesulfonamide) were the major products observed at 27.4% and 42.5%, respectively in the total system. In the acidic water/sediment system, IN-A4097 (2-chlorobenzenesulfonamide), IN-A4098 (4-methoxy-6-methyl-1,3,5-triazine-2-amine) and IN M6957 (2-chloro-N-[(4-hydroxy-6-methyl-1,3,5-triazin-2-yl)carbamoyl]benzenesulfonamide) were the major products observed at 10.5, 17.8% and 18.9%, respectively in the total system. The DT_{50} values for chlorsulfuron in the aerobic alkaline system (Mill Stream system only; Cavrini system considered more anaerobic in nature) were 21 and 26 days in the water phase and total system, respectively. In the aerobic acidic system the DT_{50} values for chlorsulfuron were 54.6 and 66.7 in the water phase and total system, respectively. Approximately 37% and 67% of the applied chlorsulfuron remained in the water phase of the alkaline and acidic water/sediment systems at Day-30 and Day-29, respectively.

The results from the CO_2 evolution test (Modified Sturm test) demonstrated that chlorsulfuron is not readily biodegradable. The CO_2 production for chlorsulfuron was at most 4% of the theoretical value by the end of the test on Day 29 not achieving the level to be considered readily biodegradable.

The behaviour in aerobic water/sediment systems and the limited production of CO_2 in the ready biodegradability test suggests that chlorsulfuron is not rapidly degradable according to the CLP criteria (CLP Regulation Annex I, point 4.1.2.9).

5.2 ENVIRONMENTAL DISTRIBUTION

5.2.1 Adsorption/Desorption

The adsorption/desorption behaviour of chlorsulfuron was studied on four soils using a batch equilibrium procedure at 20°C. The average K_{oc} value for chlorsulfuron in the four tested soils was 33.6 mL/g. The average K_d value was 0.38 mL/g. As soil pH decreased, the K_d and K_f values generally increased. The average K_f value was 0.42 mL/g and the average K_{foc} was 36.3 mL/g. The average $1/n$ value was 0.89, which indicates that non-linearity of adsorption with decreasing concentration, was significant. Soil pH and organic carbon content were significant factors in the sorption behaviour of chlorsulfuron with greater adsorption occurring at lower pH and higher OC content.

5.2.2 Volatilisation

The low vapour pressure of chlorsulfuron of 3.1×10^{-9} Pascals (2.3×10^{-11} mm Hg) at 25°C indicates little potential for volatilization and thus it would not be found in significant concentrations in air. The Henry's Law constant value of 3.5×10^{-11} Pa \times m³ \times mol⁻¹ calculated from the measured water solubility in pH 7 buffer of 12.5 g/L and vapour pressure indicates chlorsulfuron has a negligible vapour pressure above dilute aqueous solutions.

5.2.3 Distribution modelling

The low vapour pressure and Henry's law constant of chlorsulfuron indicate a low potential for volatilisation of chlorsulfuron under practical conditions of use. The calculation of the second-order rate constant and associated half-life for the reaction of chlorsulfuron in the gas phase in the troposphere was made using the method of Atkinson. In addition, the Predicted Environmental Concentrations in air (PEC_a) were also estimated for chlorsulfuron.

Currently, no models have been recommended for the estimation of the predicted environmental concentrations of chemicals in air. In the absence of a recommended model, volatilisation losses and predicted concentrations in air were estimated using the volatilisation estimation method developed by Lyman *et al.* (1982) as follows:

$$k_v = 4.4 \times 10^7 \left(\frac{vp}{K_{oc} \cdot S} \right)$$

Where

k_v = rate constant for volatilisation, day⁻¹

vp = vapour pressure of chemical in air, mm Hg

K_{oc} = organic carbon partition coefficient, mL/g

S = solubility, mg/L

The initial daily loss from a treated field are estimated as:

$$loss = (1 - \exp(-k_v)) \cdot 100$$

Where

$loss$ = percent of applied compound volatilised within 24 hours after application

k_v = rate constant for volatilisation, day⁻¹

Parameter	Value	Units	Reference
Water solubility at pH 7	12500	mg/L	DAR, Vol. 3, Annex B8, B.8.2.1a
Average soil sorption coefficient	36	mL/g	
Vapour pressure at 25°C	2.3×10^{-11}	Pa	

	1.7×10^{-13}	mm Hg	
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Based on this data and the equations in the table above it can be predicted that < 0.01% of applied chlorsulfuron would be lost to the atmosphere within 24 hours. This rate of loss is likely to result in negligible concentrations in air (PEC_{air}) due to the combination of a low rate of loss as well as the dilution effects of moving air.

5.3 AQUATIC BIOACCUMULATION

5.3.1 Aquatic bioaccumulation

The purpose of the fish accumulation study is to determine if pesticide residues accumulate in fish as human food sources and to determine the magnitude of residue in the edible fish portions. A fish-bioconcentration study is not required, due to the low $\log K_{ow}$, which is below the trigger value of 4 (pH = 7: $\log K_{ow} = 0.102$).

5.3.1.1 Bioaccumulation estimation

Since, the $\log K_{ow}$ of chlorsulfuron is lower than the threshold values (CLP Regulation ≥ 4), the potential risk for bioaccumulation in tissues of aquatic organisms is low.

5.3.1.2 Measured bioaccumulation data

No data required (see 5.3.1.1).

5.3.2 Summary and discussion of aquatic bioaccumulation

The measured $\log K_{ow}$ values for chlorsulfuron were all below the threshold value for bioaccumulation, *i.e.* threshold CLP Regulation ≥ 4 . Therefore, no experimental bioaccumulation data are required. The potential risk for bioaccumulation of chlorsulfuron in tissues of aquatic organisms is considered low.

5.4 AQUATIC TOXICITY

The ecotoxicological properties assessment for chlorsulfuron is based on the Draft Assessment Report, Addendum to the Draft Assessment Report and the EFSA Scientific Report on the peer review of chlorsulfuron.

All ecotoxicological studies on chlorsulfuron were performed under GLP and according to EPA, OECD or equivalent guidelines.

Table 10: Summary of relevant information on aquatic toxicity

Test/Method	Test substance purity; Test concentrations	Test system	Endpoints	Reference
Fish acute <i>Oncorhynchus mykiss</i> OECD 203; Commission Regulation (EC) No 440/2008 - Method C.1 Deviations: None	chlorsulfuron purity: 97.18%; mean measured concentration of 122 mg a.s./L	96-hour static juvenile, 10 fish/replicate 3 replicates /treatment	LC ₅₀ >122 mg a.s./L, measured (limit test)	Ward, T.J., Wyskeil, D.C., Boeri, R.L. (2001b) DuPont-5276 reviewed in DAR, Vol. 3, Annex B9, B.9.2.1
Fish acute <i>Lepomis macrochirus</i> OECD 203; Commission Regulation (EC) No 440/2008 - Method C.1 Deviations: None	chlorsulfuron purity: 97.18%; mean measured concentration of 128 mg a.s./L	96-hour static fingerling, 10 fish/replicate 3 replicates /treatment	LC ₅₀ >128 mg a.s./L, measured (limit test)	Ward, T.J., Wyskeil, D.C., Boeri, R.L. (2001a) DuPont-5136 reviewed in DAR, Vol. 3, Annex B9, B.9.2.1
Fish early life stage toxicity test, <i>Oncorhynchus mykiss</i> U.S. EPA 72-4 Deviations: None	chlorsulfuron purity: 97.9%; Mean, measured concentrations of 18, 32, 66, 120, 250, 470, and 900 mg a.s./L	Flow-through 77-day chronic test 40 embryos/replicate 2 replicates/treatment	NOEC = 32 mg a.s./L, measured	Pierson, K.B. (1991) HLR 494-91, Revision No. 1 reviewed in DAR, Vol. 3, Annex B9, B.9.2.2
Daphnia acute <i>Daphnia magna</i> OECD 202; Commission Regulation (EC) No 440/2008 - Method C.2 Deviations: None	chlorsulfuron purity: 97.18%; Mean, measured concentrations of 14.8, 24.4, 41.2, 68.4, and 112 mg a.s./L	48-hour static, un aerated 5 daphnids/replicate 4 replicates/treatment	EC ₅₀ >112 mg a.s./L, measured	Ward, T.J., Wyskeil, D.C., Boeri, R.L. (2001 c) DuPont-5275 reviewed in DAR, Vol. 3, Annex B9, B.9.2.1
Daphnia chronic <i>Daphnia magna</i> OECD 202 Deviations: None	chlorsulfuron purity: 95.4%; Mean, measured concentrations of 12, 28, 58, 120, 250, and 480 mg a.s./L	21-day static, un aerated 4 daphnids/replicate 10 replicates/ treatment	NOEC = 12 mg a.s./L, measured, growth	Hutton, D.G. (1989) HLR 35-89 reviewed in DAR, Vol. 3, Annex B9, B.9.2.2
Algae acute, <i>Selenastrum capricornutum</i>	chlorsulfuron purity: 98.2%; Nominal	72-hour algae 3 replicates/treatment	EC ₅₀ = 0.050 mg a.s./L, nominal,	Blasberg, J., Hicks, S. L., Stratton, J. L. (1991)

Test/Method	Test substance purity; Test concentrations	Test system	Endpoints	Reference
U.S. EPA 122-2, U.S. EPA 123-2 Deviations: None	concentrations of 0.01, 0.018, 0.032, 0.058, and 0.103 mg a.s./L	Cell counts at were made at approximately 24, 48, 72, 96, and 120 hours	cell count 120 hr NOEC = 0.010	AMR 2081-91 reviewed in DAR, Vol. 3, Annex B9, B.9.2.1
Algae acute, <i>Anabaena flos-aquae</i> OECD 201; U.S. EPA 123-2 Deviations: None	chlorsulfuron purity: 97.79%; Mean, measured concentrations of 0.236, 0.485, 0.961, 1.92, and 3.95 mg a.s./L	120-hour algae 3 replicates/treatment Cell counts were taken at 24 hour intervals	EC ₅₀ = 0.609 mg a.s./L, measured, area under the curve	Boeri, R.L., Wyskiel, D.C., Ward, T.J. (2000) DuPont-4466 reviewed in DAR, Vol. 3, Annex B9, B.9.2.1
Aquatic plants <i>Lemna gibba</i> , U.S. EPA 122-2 and 123-2 Deviations: None	chlorsulfuron purity: 97.79%; Nominal concentrations of 0.00006, 0.00012, 0.00024, 0.00048, and 0.00096 mg a.s.L	14-day growth and reproduction 5 plants with 3 fronds per plant were used/replicate 3 replicates/test concentration	EC ₅₀ = 0.00035 mg a.s./L, biomass 0.00069 mg/L, calculated average specific growth rate	Boeri, R.L., Wyskiel, D.C., Ward, T.J. (2002) DuPont-4468 reviewed in DAR, Vol. 3, Annex B9, B.9.2.1 McKelvey, R.A. (2011) DuPont-33183 reviewed in DAR Addendum, Vol. 3, Annex B9, B.9.2.16
Aquatic plants <i>Lemna gibba</i> , U.S. EPA 850.4400 (1996), OECD 221 (2006) Deviations: None	chlorsulfuron purity: 99.5% Test levels for the 4-hour exposure were 1.2, 4.1, 14, 45, 150, and 500 µg a.s./L; 8-hour exposure interval were 0.36, 1.2, 4.1, 14, 45, and 150 µg a.s./L; 24- and 48-hour intervals were 0.033, 0.11, 0.36, 1.2, 4.1, and 14 µg a.s./L	7-day variable duration Exposure intervals were 4, 8, 24, and 48 hours in duration, and there were six test levels for each exposure interval. Test concentrations varied with the exposure level and ranged from 0.033 to 500 µg a.s./L along with an untreated control.	EC ₅₀ (frond count yield) 0.1 mg/L (4 h exposure) 0.034 mg/L (8 h exposure) 0.0087 mg/L (24 h exposure) 0.001 mg/L (48 h exposure) EC ₅₀ (frond count growth rate) 0.307 mg/L (4 h exposure) >0.15 mg/L (8 h exposure) >0.014 mg/L (24 h exposure) >0.014 mg/L (48 h exposure) The 48 hour NOEC, based on frond count, biomass, yield based on frond count and biomass, and growth rate based on frond count and	Porch, J.R., Kendall, T.Z., Krueger, H.O. (2010 a) DuPont-28843 reviewed in DAR Addendum, Vol. 3, Annex B9, B.9.2.11.2

Test/Method	Test substance purity; Test concentrations	Test system	Endpoints	Reference
			biomass = 0.00036 mg a.s./L	
Aquatic plants <i>Lemma gibba</i> , OPPTS 850.4400 (1996), OECD 221 (2006)	chlorsulfuron purity: 99.5% Nominal concentrations of 1.6, 3.1, 6.3, 13, 25, and 50 µg a.s./L	21-day dormancy Frond counts were made on Days 0, 14, 17, 19, and 21. The pre-exposure and exposure periods were conducted at 8 ± 1°C in order to maintain dormancy of the plants. At the end of 14 days, the temperature was increased to 24 ± 2°C in order to stimulate resumption of rapid growth. Biomass was determined at the completion of the 21-day test. Four plants containing a total of 12 fronds were used/replicate. 6 replicates/test concentration.	EC ₅₀ (dry weight yield) 0.0072 mg/L (no clearance period) >0.051 mg/L (clearance period)	Porch, J.R., Kendall, T.Z., Krueger, H.O (2010 b) DuPont-30213 reviewed in DAR Addendum, Vol. 3, Annex B9, B.9.2.11.1

5.4.1 Fish

5.4.1.1 Short-term toxicity to fish

The acute toxicity of chlorsulfuron to unfed juvenile rainbow trout, *Oncorhynchus mykiss*, was determined in an unaerated, static, 96-hour limit test (Ward, T.J., Wyseil, D.C., Boeri, R.L. (2001b). Treatments consisted of a HEPES-buffered dilution water control and a single nominal concentration of 120 mg chlorsulfuron/L. Three replicates containing 10 fish each were exposed to the treatment concentration. Test solutions were maintained between 11.5 and 12.4°C. Summaries of cumulative mortality and sublethal effects are presented in the table below. The mean, measured concentration of chlorsulfuron was 122 mg a.s./L, which was 102% of the nominal concentration. There were no mortality or sublethal effects at 122 mg a.s./L. The highest concentration causing no mortality was 122 mg a.s./L and the lowest concentration causing 100% mortality was >122 mg a.s./L.

Table 11. Observed mortality and sublethal effects of rainbow trout, *Oncorhynchus mykiss*, exposed to chlorsulfuron for 96 hours in an unaerated, static, acute test.

Mean, measured test item concentration (mg a.s./L)	Cumulative mortality ^{a, b} (No. dead / No. at test start)			
	24 hour	48 hour	72 hour	96 hour
Water control	0/30	0/30	0/30	0/30
Chlorsulfuron: 122	0/30	0/30	0/30	0/30

^a Total of three replicates. Each replicate contained 10 fish (total 30 fish per test concentration) at test start.

^b There were no sublethal effects.

Conclusion: The 96-hour LC₅₀ in the rainbow trout was >122 mg chlorsulfuron/L, based on mean, measured concentrations.

The acute toxicity of chlorsulfuron to unfed fingerling bluegill sunfish, *Lepomis macrochirus*, was determined in an aerated, static, 96-hour test (Ward, T.J., Wyskeil, D.C., Boeri, R.L. (2001a)). The definitive study was conducted with a single nominal concentration of chlorsulfuron of 120 mg/L and a HEPES-buffered solution water control. Three replicates containing 10 fish were exposed to a 128 mg/L mean, measured concentration of chlorsulfuron. Test solutions were maintained between 22.1 and 23.0°C. Summaries of cumulative mortality and sublethal effects are presented in Table 12. The mean, measured concentration of chlorsulfuron was 128 mg a.s./L, which was 107% of the nominal concentration. There were no mortality or sublethal effects at 128 mg a.s./L. The highest concentration causing no mortality was 128 mg a.s./L and the lowest concentration causing 100% mortality was >128 mg a.s./L.

Table 12. Observed mortality and sublethal effects of bluegill sunfish, *Lepomis macrochirus*, exposed to chlorsulfuron for 96 hours in an unaerated, static, acute test.

Mean, measured test item concentration (mg a.s./L)	Cumulative mortality ^a (No. dead / No. at test start ^b)			
	24 hour	48 hour	72 hour	96 hour
Water control ^c	0/30	0/30	0/30	0/30
Chlorsulfuron: 128	0/30	0/30	0/30	0/30

^a There were no sublethal effects.

^b Total of three replicates. Each replicate contained 10 fish (total 30 fish per test concentration) at test start.

^c HEPES-buffered dilution water control.

Conclusion: The 96-hour LC₅₀ in the bluegill sunfish was >128 mg chlorsulfuron/L, based on mean, measured concentrations.

5.4.1.2 Long-term toxicity to fish

The effects of chlorsulfuron on the early life stages of rainbow trout (*Oncorhynchus mykiss*) were determined in a flow-through 77-day chronic test (Pierson, K.B. (1991)). A dilution water control and nominal test substance concentrations of 15.6, 31.3, 62.5, 125, 250, 500, and 1000 mg a.s./L were

used during the study. A total of 80 embryos (2 replicates of 40 embryos each) were exposed per concentration level. Test solutions were maintained at $10 \pm 2^\circ\text{C}$. Low light intensity (2.5 lux; 16-hour light:8-hour dark) was maintained during hatching and through swim-up (Day-44). On Day-44 fingerlings were thinned to 15 per replicate (30 fingerlings/treatment level). Light was then increased to 86-140 lux (16-hour light:8-hour dark) during the remainder of the test. Analytical verification of chlorsulfuron concentrations was made prior to the beginning of the test. Data on embryo and larval survival, and length were taken at 44 days. Fingerling survival, length, and wet weight were determined at 77 days. Mean, measured concentrations of chlorsulfuron were 18, 32, 66, 120, 250, 470, and 900 mg a.s./L and ranged from 90 to 115% of nominal concentrations. All chemical and physical parameters for the 77-day study were within acceptable ranges. The $\text{LC}_{50\text{s}}$ could not be calculated since 50% mortality was not observed at the highest test concentration. The MATC was 46 mg a.s./L and the NOEC for rainbow trout exposed to chlorsulfuron for 77 days in a flow-through test was 32 mg a.s./L based on standard length. The LOEC was 66 mg a.s./L (based on standard length). Behaviour that could be correlated with toxicity was only noted at 470 and 900 mg a.s./L

A summary of embryo/larval survival (44 days) and fingerling survival, length, and wet weight is presented in Table 13. .

Table 13. Summary of length, weight, and survival of chlorsulfuron in an early life stage test with rainbow trout.

Mean, measured test item concentration (mg a.s./L)	Mean standard length (Day-77) ^{a,b}	Mean wet weight (Day-77) ^{a,b}	Embryo/larval survival (Day-44) ^c		Fingerling survival (Day-77) ^{a,c}	
	(cm)	(g)	No. alive/total	(%)	No. alive/total	(%)
Water control	3.5	0.66	59/80	74	28/30	93
Chlorsulfuron:						
18	3.6	0.70	60/80	75	28/30	93
32	3.4	0.74	55/80	69	30/30	100
66	3.2*	0.67	58/80	73	29/30	97
120	3.2*	0.69	52/80	65	30/30	100
250	3.2*	0.61	61/80	76	30/30	100
470	3.2*	0.47*	50/80	63	29/30	97
900	2.6*	0.16*	52/80	65	25/30	83*

* Statistically significant relative to the control ($\alpha = 0.05$)

^a Based on 77-day data (fingerlings from original 80 embryos were thinned to 30 fingerlings/treatment level on Day 44).

^b Kruskal-Wallis test and Mann-Whitney test with a Bonferroni correction for multiple comparisons.

^c Cochran-Armitage test.

Conclusion: The LC_{50} for rainbow trout exposed to chlorsulfuron for 77 days in a flow-through test was >900 mg a.s./L mean measured concentration (the highest rate tested) and the NOEC based on standard length was 32 mg a.s./L mean measured concentration.

5.4.2 Aquatic invertebrates

5.4.2.1 Short-term toxicity to aquatic invertebrates

The acute toxicity of chlorsulfuron to unfed *Daphnia magna* (<24-hour old) was determined in an unaerated, static, 48-hour test (Ward, T.J., Wyseil, D.C., Boeri, R.L. (2001c). Treatments consisted of a HEPES-buffered dilution water control and nominal concentrations of 16, 26, 43, 72, and 120 mg chlorsulfuron/L. Five daphnids were used per replicate with four replicates per test concentration and

control. A summary of the findings is presented in Table 14. The mean, measured concentrations of chlorsulfuron during the test were 14.8, 24.4, 41.2, 68.4, and 112 mg/L. Mean measured concentrations chlorsulfuron ranged from 93 to 96% of the nominal concentrations. The highest concentration causing no immobility was 68.4 mg/L. None of the tested concentrations caused 100% immobility.

Table 14 Summary of observed immobility and sublethal effects of unfed, <24 hour old *Daphnia magna* exposed to chlorsulfuron for 48 hours in an unaerated, static, acute test.

Mean, measured test item concentration (mg/L)	Immobility (No. immobile/No. at test start)							
	24 hours				48 hours			
	A ^a	B ^a	C ^a	D ^a	A ^a	B ^a	C ^a	D ^a
Dilution water control	0/5	0/5	0/5	0/5	0/5	0/5	0/5	0/5
Chlorsulfuron:								
14.8	0/5	0/5	0/5	0/5	0/5	0/5	0/5	0/5
24.4	0/5	0/5	1/5	0/5	0/5	0/5	1/5	0/5
41.2	0/5	0/5	0/5	0/5	0/5	0/5	0/5	0/5
68.4	0/5	0/5	0/5	0/5	0/5	0 ^{1-b} /5	0/5	0/5
112	0/5	0/5	0/5	0/5	0/5	0/5	0/5	1/5

^a Replicate test chamber containing 5 daphnids at test start.

^b Daphnids at surface; the superscript numbers indicate the number of daphnids with this sublethal effect.

Conclusion: The 48-hour EC₅₀ in *Daphnia magna* was >112 mg/L, based on mean, measured concentrations.

5.4.2.2 Long-term toxicity to aquatic invertebrates

The effects of chlorsulfuron on the growth and reproduction of *Daphnia magna* (<24-hour old) were assessed in an unaerated, static, 21-day test (Hutton, D.G. (1989). Treatments consisted of a dilution water control, and nominal concentrations of 16, 31, 62, 125, 250, and 500 mg chlorsulfuron/L. Ten replicates were used per treatment, with four daphnid per replicate. Test concentrations were renewed every Monday, Wednesday and Friday or 48 hours. Statistical procedures employed were: Probit analysis (calculation of the EC₅₀ values), Fisher's Exact test (survival), one-way analysis of variance followed by means comparison with Dunnett's test (growth and reproduction parameters). Analytical verification of chlorsulfuron concentrations was made on test solutions sampled on Day 0 and at regular intervals during the study. Mean, measured concentrations of chlorsulfuron were 12, 28, 58, 120, 250, and 480 mg/L and ranged from 75 to 96% of nominal concentrations. All chemical and physical parameters for the 21-day study were within acceptable ranges. A summary of percent adult survival, first day of reproduction, total live young produced per surviving female, total immobile young produced per surviving female, and length of surviving adults is presented in Table 15.

Table 15. Summary of test endpoints following exposure of *Daphnia magna* to chlorsulfuron for 21 days.

Mean, measured test item concentration (mg/L)	Mean % adult survival ^a	Mean first day of reproduction ^b	Mean total live young ^c	Mean total immobile young ^d	Mean adult length (mm)
Water control	100	10	397	0	4.2
Chlorsulfuron:					
12 ^e	98	10	390	0	4.1
28	95	10	376	0	4.0*
58 ^f	90	10	359	0	4.0*
120 ^g	100	10	332*	0	4.0*
250	98	10	270*	0	3.6*
480	92	10	76*	0	3.2*

* = Statistically significant relative to the control group ($\alpha = 0.05$)

^a Percent of adult daphnids alive at the end of the test (immobility was synonymous with death)

^b First day that reproduction was observed in the replicates

^c Mean of live young produced per surviving female

^d Mean of immobile young produced per surviving female

^e NOEC for growth based on mean adult length, (Dunnett's test, $\alpha = 0.05$)

^f NOEC based on mean total live young, (Dunnett's test, $\alpha = 0.05$)

^g LOEC based on mean total live young

Table 16. Chronic toxicity, reproduction and growth in *Daphnia magna* exposed to chlorsulfuron - Summary of endpoints.

NOEC (Highest tested dose without toxic effect) based on survival	>480 mg/L
LOEC (Lowest observed effect concentration) for survival	>480 mg/L
MATC (Maximum Acceptable Toxicant Concentration) for survival	>480 mg/L
21-day NOEC for reproduction	58 mg/L
21-day LOEC for reproduction	120 mg/L
MATC for reproduction (range from highest NOEC to LOEC)	Range: 58 to 120 mg/L
21-day NOEC for growth	12 mg/L
21-day LOEC for growth	28 mg/L
MATC for growth	Range: 12 to 28 mg/L
EC ₅₀ (immobilisation) and	>480 mg/L
EC ₅₀ (reproduction)	>480 mg/L

Conclusion: The 21-day NOEC for the most sensitive parameter, growth, was 12 mg/L. The 21-day LOEC for growth was 28 mg/L, based on mean measured concentration.

The EC₅₀ (immobilisation) and EC₅₀ (reproduction), for *Daphnia magna* neonates exposed for 21 days under unaerated, static conditions were each >480 mg/L, the highest concentration tested.

5.4.3 Algae and aquatic plants

The effect of chlorsulfuron on the growth and growth rate of green algae, *Selenastrum capricornutum*, was determined using algal cultures with AAP nutrient medium, incubated at $24 \pm 2^\circ\text{C}$ for 120 hours (Blasberg, J., Hicks, S. L., Stratton, J. L. (1991). The algae were exposed to total nominal formulation

concentrations of 0.010, 0.030, 0.060, 0.120, and 0.240 mg chlorsulfuron/L of nutrient medium. For the dose response test, the organisms were exposed for 120 hours (5 days) without test medium renewal. Each blank control, test concentration, and vehicle blank (acetone) was tested as 3 replicates. Cell counts were made approximately 24, 48, 72, 96, and 120 hours after test initiation. The effect was expressed as healthy cell count (cell density) for the 120-hour (Day 5) interval of the test.

Chlorsulfuron was determined to be stable over the course of the definitive test as evidenced by the analytical recoveries obtained from the Day 0 and Day 5 (120 hour) test solutions. The 120-hour mean-measured concentrations ranged from 94 to 106% of the nominal concentrations.

The effects of chlorsulfuron on the growth and growth rate of *Selenastrum capricornutum* are shown in Table 17. The 72-hour EC₅₀ was determined to be 0.068 mg chlorsulfuron/L (confidence interval: 0.045 to 0.091 mg/L) and the 96-hour EC₅₀ was determined to be 0.090 mg chlorsulfuron/L (confidence interval: 0.056 to 0.120 mg/L). The 120-hour EC₅₀ was determined to be 0.050 mg chlorsulfuron/L (confidence interval: 0.040 to 0.060 mg/L) and the 120-hour NOEC was determined to be 0.010 mg chlorsulfuron/L, each based on nominal concentrations.

Table 17. Summary of algal growth following exposure of green algae, *Selenastrum capricornutum* to chlorsulfuron.

Nominal test item concentration (mg/L)	Cell counts (cells × 10 ⁴ /mL)		
	72-hour	96-hour	120 hour
Blank control	14	29	110
Vehicle blank	9.7	27	94
Chlorsulfuron:			
0.010	13	36	100
0.018	7.9	30	72*
0.032	7.7	30	59*
0.058	5.6*	17*	40*
0.103	3.1*	12*	28*

* Significantly different from the control by the Dunnett's test criteria, $\alpha = 0.05$.

Conclusion: Growth data (cell counts) obtained with chlorsulfuron on *Selenastrum capricornutum* were as follows:

Cell count: 72-hour EC₅₀ = 0.068 mg chlorsulfuron/L
72-hour NOEC = 0.032 mg chlorsulfuron/L

120-hour EC₅₀ = 0.050 mg chlorsulfuron/L
120-hour NOEC = 0.010 mg chlorsulfuron/L

The effect of chlorsulfuron on *Anabaena flos-aquae* was determined using algal cultures with AAP nutrient medium. Three replicates at nominal concentrations of 0.25, 0.50, 1.0, 2.0, and 4.0 mg chlorsulfuron/L, were incubated at 24 ± 2°C for 120 hours and cell counts were taken at 24-hour intervals (Boeri, R.L., Wyskiel, D.C., Ward, T.J. (2000)). To assess recovery after the initial 120 hours exposure period, algae from a nominal concentration of 40 mg/L was placed in nutrient medium without chlorsulfuron. (A determination of whether effects were algistatic was conducted with the maximally inhibited test concentration (4.0 mg/L, the highest rate tested) rather than with all test concentrations with inhibited growth). Recovery cell counts were taken every 3 days for up to 14 days.

The mean measured concentrations of chlorsulfuron ranged from 94 to 99% of the targeted nominal concentrations corrected for test substance purity of 97.79%. Control solutions showed no detectable concentrations of chlorsulfuron.

The effects of chlorsulfuron on the growth and growth rate of *Anabaena flos-aquae* are shown in Table 18. The 72-hour EC₅₀ values determined with the number of cells per mL, growth rate, and the area under the growth curve are 0.585 mg/L, 3.46 mg/L, and 0.371 mg/L chlorsulfuron, respectively. The 120-hour EC₅₀ values determined with the number of cells per mL, growth rate, and the area under the growth curve are 0.807 mg/L, 1.77 mg/L, and 0.609 mg/L chlorsulfuron, respectively.

Table 18. Summary of algal growth inhibition following exposure of *Anabaena flos-aquae* to chlorsulfuron for 120 hours.

Mean. measured test item concentration (mg/L)	Mean cell density (cells/mL)	% Inhibition relative to control		
		Cell density	Area under the growth curve	Growth rate
Water control	1.41 × 10 ⁶	—	—	—
Chlorsulfuron:				
0.236	1.16 × 10 ⁶	102	81	100
0.485	8.93 × 10 ⁵	78	67	94
0.961	4.85 × 10 ⁵	43	30	84
1.92	3.30 × 10 ⁴	3	10	40
3.95	<1.0 × 10 ⁴	<1	5	20

During the algistatic/algicidal test set up for 120 hours at the termination of the final definitive test, algae collected from each 4.0 mg/L (nominal) test chamber and combined into fresh dilution media increased from a calculated cell concentration of <150 cells per mL to 348000 cells/mL. These data indicates that the effect of chlorsulfuron at this concentration was algistatic rather than algicidal.

Conclusion: Growth inhibition values obtained with chlorsulfuron on *Anabaena flos-aquae* were as follows:

Cell density: 120-hour EC₅₀ = 0.807 mg chlorsulfuron/L

Area under the growth curve: 120-hour EC₅₀ = 0.609 mg chlorsulfuron/L

Growth rate: 120-hour EC₅₀ = 1.77 mg chlorsulfuron/L

The effects of chlorsulfuron on growth and growth rate of *Anabaena flos-aquae* were found to be algistatic at concentrations less than or equal to 4.0 mg/L.

The effect of chlorsulfuron on *Lemna gibba* G3 was determined in 20X AAP nutrient medium (Boeri, R.L., Wyskiel, D.C., Ward, T.J. (2002)). Treatments consisted of nominal concentrations of 0.00006, 0.00012, 0.00024, 0.00048, and 0.00096 mg chlorsulfuron/L and a test media control. Five plants with 3 fronds per plant were used per replicate with three replicates per test concentration and control. Replicates were incubated for 14 days at a mean temperature of approximately 24.3°C (range 24.0 to 24.6°C). Frond count and biomass were determined after 14 days of exposure. To assess recovery after the initial 14-day exposure period, fronds from nominal concentrations 0.00048, and 0.00096 mg chlorsulfuron/L exhibiting >50% growth inhibition were placed in fresh nutrient medium without chlorsulfuron. Fronds were counted after an additional 14 days. Statistical methods employed were: Bruce-Versteeg method (calculation of EC values), and one-way analysis of variance followed by means comparison with Dunnett's test ($\alpha = 0.05$) to determine the NOEC. In the recovery test, normal growth and reproduction resumed in test concentrations equal to or lower than 0.00048 mg/L.

The effects of chlorsulfuron on the growth and reproduction of *Lemna gibba* G3 are shown in Table 19.

Table 19. Summary of growth inhibition following exposure of *Lemna gibba* G3 to chlorsulfuron for 14 days.

Test item nominal concentration (mg/L)	FronD number ^a		Biomass	
	14-Day mean frond number	% Inhibition relative to control	14-Day mean biomass (mg)	% Inhibition relative to control
Blank control	837	—	78.7	—
Chlorsulfuron:				
0.00006	624	25	48.7	38
0.00012	727	13	70.8	10
0.00024 ^b	598	29	45.9	42
0.00048	308	63	23.5	70
0.00096	37	96	11.9	85

^a Based on total number of non-chlorotic fronds.

^b NOEC value for both parameters as determined by Dunnett's test ($\alpha = 0.05$)

Conclusion: Growth inhibition values based on nominal concentrations obtained with chlorsulfuron on *Lemna gibba* G3 were as follows:

**FronD number: 14-day EC₅₀ = 0.00042 mg chlorsulfuron/L
14-day NOEC = 0.00024 mg chlorsulfuron/L**

**FronD biomass: 14-day EC₅₀ = 0.00035 mg chlorsulfuron/L
14-day NOEC = 0.00024 mg chlorsulfuron/L**

The effects of chlorsulfuron on *Lemna gibba* G3 are expected to be reversible at concentrations <0.00048 mg chlorsulfuron/L.

For *Lemna gibba*, the EC₅₀ based on average specific growth rate was not provided in the original study report DuPont-4468. Therefore, this endpoint was not available in the DAR (July 2007). The detailed calculation of average specific growth rates based on frond counts has been provided in the document position paper DuPont-33183 (McKelvey, 2011): EC₅₀ = 0.00069 mg/L (which was reviewed in the DAR addendum).

Toxicity after various lengths of exposure of chlorsulfuron to the floating freshwater vascular plant *Lemna gibba* G3 was determined in a 7-day test (Porch, J.R., Kendall, T.Z., Krueger, H.O. (2010a)). The test was based on U.S. Environmental Protection Agency (EPA) Series 850 – Ecological Effects Test Guidelines, OPPTS Number 850.4400 and OECD Guideline 221. Treatments consisted of four exposure intervals (4, 8, 24, and 48 hours), each with six nominal concentrations. Test concentrations varied with the exposure interval, and ranged from 0.033 to 500 µg a.s./L, along with an untreated control. The 7-day EC₅₀ values, based on nominal chlorsulfuron concentrations were as follows: 4-hour exposure - frond count 119 µg a.s./L, frond count yield 100 µg a.s./L, biomass 128 µg a.s./L, biomass yield 112 µg a.s./L, growth rate (based on frond count) 307 µg a.s./L, and growth rate (based on frond biomass) >500 µg a.s./L; 8-hour exposure - frond count 40 µg a.s./L, frond count yield 34 µg a.s./L, biomass 42 µg a.s./L, biomass yield 36 µg a.s./L, growth rate (based on frond count) >150 µg a.s./L, and growth rate (based on frond biomass) >150 µg a.s./L; 24-hour exposure - frond count 12 µg a.s./L, frond count yield 8.7 µg a.s./L, biomass >14 µg a.s./L, biomass yield 12 µg a.s./L, growth

rate (based on frond count) >14 µg a.s./L, and growth rate (based on frond biomass) >14 µg a.s./L; 48-hour exposure - frond count 1.1 µg a.s./L, frond count yield 1.0 µg a.s./L, biomass 1.7 µg a.s./L, biomass yield 1.2 µg a.s./L, growth rate (based on frond count) >14 µg a.s./L, and growth rate (based on frond biomass) >14 µg a.s./L. The 7-day NOEC based on nominal chlorsulfuron concentration was 0.36 µg a.s./L for biomass after 8 and 48 hours of exposure, and 1.2 µg a.s./L for biomass after 4 and 48 hours of exposure.

Toxicity of chlorsulfuron to the floating freshwater vascular plant *Lemna gibba* G3 was determined in a 21-day test with exposure during a period of induced dormancy to evaluate potential effects from chlorsulfuron exposure during the late fall, winter, and early spring (Porch, J.R., Kendall, T.Z., Krueger, H.O. (2010b)). The test was conducted using methods based on modifications to U.S. Environmental Protection Agency (EPA) Series 850 – Ecological Effects Test Guidelines, OPPTS Number 850.4400 and OECD Guideline 221. Treatments consisted of six nominal concentrations of 1.6, 3.1, 6.3, 13, 25, and 50 µg a.s./L and an untreated control. Two sets of plants were tested at each nominal concentration. One set of plants was exposed to the test concentrations followed by a seven-day clearance period in untreated nutrient medium; the other set of plants had no clearance period following exposure. The pre-exposure and exposure periods were conducted at $8 \pm 1^\circ\text{C}$ in order to induce and maintain dormancy of test plants. At the end of 14 days, the temperature of the test was increased to $24 \pm 2^\circ\text{C}$ in order to stimulate the resumption of rapid growth.

For exposure to chlorsulfuron without a clearance period, the 21-day EC₅₀ values, based on mean, measured concentrations of chlorsulfuron for frond count, frond count growth rate, and biomass growth rate were greater than 51 µg a.s./L, for frond count yield was 19.3 µg a.s./L, for biomass was 11.8 µg a.s./L, and for biomass yield was 7.2 µg a.s./L. For exposure to chlorsulfuron followed by a clearance period, the 21-day EC₅₀ values, based on mean, measured concentrations of chlorsulfuron for all variables was greater than 51 µg a.s./L.

5.4.4 Other aquatic organisms (including sediment)

To prevent unnecessary testing with substances of low toxicity to aquatic invertebrates, the NOEC in the chronic *Daphnia magna* test must be <0.1 mg/L for testing on sediment dwelling organisms to be warranted (SANCO/3268/2001). For chlorsulfuron, the chronic NOEC for *Daphnia magna* is 12 mg a.s./L.

5.5 COMPARISON WITH CRITERIA FOR ENVIRONMENTAL HAZARDS (SECTIONS 5.1 – 5.4)

Aquatic Toxicity

Both acute and chronic toxicity tests were conducted for the three trophic levels.

The 96 hour acute LC₅₀ for two species of fish (*Oncorhynchus mykiss* and *Lepomis macrochirus*) are both greater than values, with LC₅₀ values of >122 mg a.s./L and >128 mg a.s./L, respectively. The flow-through 77 day chronic fish test resulted in a NOEC of 32 mg a.s./L.

The 48 hour EC₅₀ for aquatic invertebrates is greater than 112 mg a.s./L, with a chronic 21 day NOEC = 12 mg a.s./L.

Two species of algae were tested, with the most sensitive endpoint belonging to *Selenastrum capricornutum*. The EC₅₀ for cell count is 0.068 mg a.s./L.

The most sensitive species are *Lemna gibba*, with a 14 day study giving an EC₅₀ (biomass) of 0.00035 mg a.s./L and an average specific growth rate calculated to be 0.00069 mg a.s./L.

5.6 CONCLUSIONS ON CLASSIFICATION AND LABELLING FOR ENVIRONMENTAL HAZARDS (SECTIONS 5.1 – 5.4)

Chlorsulfuron shows low acute toxicity to fish, as well as invertebrates, with acute values >100 mg a.s./L in accordance with GLP testing.

A fish-bioconcentration study is not required, due to the low log K_{ow} , which is below the trigger value of 4 (pH = 7: log K_{ow} = 0.102).

In toxicity studies for algal and aquatic plants, EC_{50} s at concentrations ≤ 1 mg a.s./L were observed. In addition, chlorsulfuron is not readily biodegradable, and is unlikely to bioaccumulate in aquatic organisms (log K_{ow} <4). As a consequence, and according to the CLP Regulation, due to its acute effect on algal/aquatic plants at a concentration ≤ 1 mg a.s./L and due to its low degradability, chlorsulfuron should be classified as Aquatic Acute 1 and Aquatic Chronic 1.

In accordance with article 10 of the CLP Regulation (EC) No. 1272/2008, if an M-Factor is not yet given in Part 3 of Annex VI to the CLP Regulation, an M-Factor should be determined and a scientific justification provided, when classifying substances for Acute Category 1 or Chronic Category 1.

For this substance, an acute M-Factor has been set at “1000” based on the following criteria:

- The lowest reported effects in a 14 day *Lemna gibba* study, with an EC_{50} value of 0.00035 mg a.s./L. This GLP study was conducted according to U.S. EPA 122-2 and 123-2, and is summarized in DuPont-4468 (reviewed in DAR, Vol. 3, Annex B9, B.9.2.1).
- The CLP Regulation (Table 4.1.3) M-Factor of “1000” for Acute Toxicity is in the range of “0.0001 < EC_{50} \leq 0.001”

For this substance, a chronic M-Factor has been set at “100” based on the following criteria:

- The 48 hour NOEC for *Lemna gibba*, based on frond count, biomass, yield based on frond count and biomass, and growth rate based on frond count and biomass = 0.00036 mg a.s./L. This GLP study was conducted according to U.S. EPA 850.4400 (1996), OECD 221 (2006) and is summarized in DuPont-28843 (reviewed in DAR Addendum, Vol. 3, Annex B9, B.9.2.11.2).
- The test substance is not readily biodegradable, determined from the results of a modified Sturm Test, according to the criteria of OECD 301B, and summarized in DuPont-6705 (reviewed in DAR, Vol. 3, Annex B8, B.8.4.3.1).
- The CLP Regulation (Table 4.1.3) M-Factor of “100” for Chronic Toxicity for not readily biodegradable substances is in the range of “0.0001 < NOEC \leq 0.001”

Proposed classification of chlorsulfuron based on CLP criteria:

Aquatic Acute 1

Aquatic Chronic 1

M-factor (acute): 1000

M-factor (chronic): 100

RAC evaluation of environmental hazards

Summary of the Dossier submitter’s proposal

Chlorosulfuron is a herbicide and is listed in Annex VI of CLP Regulation since 2008. The

dossier submitter (DS) reviewed classification for environmental hazards to include the 2nd ATP changes and proposed that the current classification (Aquatic Acute 1 - H400 and Aquatic Chronic 1 - H410) should be kept, and an acute M-factor of 1000 and a chronic M-factor of 100 should be added to the entry.

The DS concluded that chlorsulfuron is not readily biodegradable. It was also considered as very toxic to algae and aquatic plants, the latter being the most sensitive species in both acute and chronic tests as was confirmed by two studies on *Lemna gibba*. In the first GLP experiment (Boeri *et al.*, 2002), the inhibition values on frond count after 14 days of exposure were the following: EC₅₀ = 0.00035 mg/L and NOEC = 0.00024 mg a.s./L. In the second GLP experiment (Porch *et al.*, 2010a), chlorsulfuron toxicity was tested after four periods of exposure (4, 8, 24, and 48 hours), each with six nominal concentrations ranging from 0.033 to 500 µg a.s./L. The lowest NOEC based on frond count was equal to 0.00036 mg a.s./L.

In conclusion, the DS proposed to add an acute M-factor of 1000 and a chronic M-factor of 100 to the current Annex VI entry.

Comments received during public consultation

Two Member States agreed with the proposed M-factors, but requested further information on the studies or had minor comments on the data presented. Two member states, suggested to recalculate data from the Boeri *et al.* (2002) study and for classification purposes to use the 7-day E_rC₅₀ and NOEC and the 14-day E_rC₅₀ and corresponding NOEC.

Assessment and comparison with the classification criteria

All available studies on fate and behaviour of chlorsulfuron in the environment were performed under GLP and according to US EPA, OECD or equivalent guidelines.

Degradability

Hydrolysis

Chlorsulfuron is essentially stable at pH 7 and pH 9. At pH 5 chlorsulfuron hydrolyses significantly with a calculated first-order half-life of ~23 days at 25 °C (Dietrich, 1989).

Aqueous photolysis

Photolysis is not considered a major degradation process for chlorsulfuron at pH 5, pH 7, or pH 9 at 25 °C (Dietrich, 1989).

Soil photolysis

Chlorsulfuron degrades in dry irradiated alkaline soil with DT₅₀ and DT₉₀ values of 62.2 and 207 days and is relatively stable in non irradiated systems (Hawkins, 1990).

Biodegradation

Not readily biodegradable according to the criteria of OECD 301B (Barnes, 2001).

Aerobic water/sediment

Chlorsulfuron degrades in an alkaline aerobic sediment system with DT₅₀ and DT₉₀ values of 21 and 69 days in the water phase and 26 and 87 days in the total system.

In conclusion, RAC agrees with the DS that chlorsulfuron should be considered not rapidly degradable according to CLP.

Aquatic bioaccumulation

The only available information on bioaccumulation potential was the measured log K_{ow}, which is below the trigger value of ≥ 4 (pH = 7: log K_{ow} = 0.102). RAC agrees with the

DS that chlorsulfuron has a low potential for bioaccumulation.

Aquatic Toxicity

Both acute and chronic toxicity tests were conducted for three trophic levels. The 96 hour acute LC₅₀ values for two species of fish (*Oncorhynchus mykiss* and *Lepomis macrochirus*) are greater than 122 mg a.s./L and 128 mg a.s./L, respectively. The flow-through 77 day chronic fish test resulted in a NOEC of 32 mg a.s./L.

The 48 hour EC₅₀ for aquatic invertebrates (*Daphnia magna*) is greater than 112 mg a.s./L with a chronic 21 day NOEC = 12 mg a.s./L.

Two species of algae were tested with the most sensitive endpoint belonging to *Selenastrum capricornutum*. The E_rC₅₀ for cell count is 0.068 mg a.s./L.

The most sensitive species is *Lemna gibba* (Boeri *et al.*, 2002) with a 7 day E_rC₅₀, a 7 day NOE_rC, a 14 day E_rC₅₀, and a 14 day NOE_rC for average specific growth rate, based on nominal and geometric mean concentrations, which are presented in Table 1.

Table 1. 7 and 14 day E_rC₅₀ and NOE_rC values based on growth rate

Response	Day	NOE _r C (µg/L)	Concentrations	E _r C ₅₀ (µg/L)	95% Confidence Intervals
Growth rate	7	0.24	Nominal	0.5967	(0.4760, 0.7250)
	14	0.24	Nominal	0.715 ^a	(0.6438, 0.7857)

^a Previously calculated to be 0.69 µg/L (McKelvey, 2011)

In toxicity studies for algal and aquatic plants, E_rC₅₀ and NOE_rC values at concentrations ≤ 1 mg a.s./L were observed. In addition, chlorsulfuron is not readily biodegradable, and is unlikely to bio-accumulate in aquatic organisms (log Kow < 4). As a consequence, and according to the CLP Regulation, due to its acute effects on algae and aquatic plants at concentrations < 1 mg a.s./L and its low degradability, RAC confirms the current chlorsulfuron classification, i.e. Aquatic Acute 1 and Aquatic Chronic 1.

RAC agrees with the DS proposal of an **acute M-Factor of 1000** based on the following criteria:

- A 14 day static study conducted on *Lemna gibba*, with a calculated 7 day E_rC₅₀ of 0.60 µg a.s./L (0.0006 mg/L) (7 day calculation based on frond count data collected on day 6 and day 8, (Boeri *et al.*, 2002). Calculations were conducted outside of study report, see also Supplemental Information – In depth analysis by RAC).
- The CLP Regulation states that an M-factor of 1000 is to be used if the acute toxicity is in the range of 0.0001 < EC₅₀ ≤ 0.001 (mg/L).

RAC agrees with the DS proposal of a **chronic M-Factor of 100** based on the following criteria:

- A 14 day static study conducted on *Lemna gibba*, with a calculated 7 and 14 day NOEC value, based on growth rate, of 0.24 µg a.s./L (0.00024 mg/L) (Boeri *et al.*, 2002, additional calculations conducted outside of study report, see also Supplemental Information – In depth analysis by RAC).
- Chlorsulfuron is not readily biodegradable, determined from the results of a modified Sturm Test, according to the criteria of OECD 310B, and summarized in Barnes (2001).

- The CLP Regulation states that an M-factor of 100 is to be used if the chronic toxicity for non-readily biodegradable substances is in the range of $0.0001 < \text{NOEC} \leq 0.001$ (mg/L).

In conclusion in agreement with DS proposal, RAC recommends that Chlorsulfuron should be classified as:

**Aquatic Acute 1; H400, M-factor = 1000,
Aquatic Chronic 1; H410, M-factor = 100**

according to CLP (Regulation (EC) No. 1272/2008).

Supplemental information - In depth analyses by RAC

Analyses

L. GIBBA FROND COUNT DATA

Original *L. gibba* frond count data for Chlorsulfuron

The frond count data from day 0 to day 14 by treatment group and replicate, as presented in Boeri *et al.* (2002), are provided in Table 2.

Table 2. *L. gibba* frond count data from Boeri *et al.* (2002)

Rep.	Exposure Initiated: Day 0:		Exposure Ended: Day 14:						Nominal Chlorsulfuron Concentration $\mu\text{g/L}$	
	Count (Fronds) by Test Day									
	0	1	4	6	8	11	13	14		
1	15	22	86	134	240	415	482	529	Blank Control	
2	15	22	80	143	322	710	812	1018		
3	15	20	80	152	359	584	716	965		
	15	21	82	143	307	570	670	837	Mean	
	0	1	3	9	61	148	170	268	Std. Dev.	
	0.0	5.4	4.2	6.3	19.8	26.0	25.3	32.0	Coeff. of Variation	
1	15	22	66	119	214	344	432	493	0.06	
2	15	23	100	167	333	607	756	784		
3	15	20	81	125	235	395	520	594		
	15	22	82	137	261	449	569	624	Mean	
	0	2	17	26	64	139	168	148	Std. Dev.	
	0.0	7.1	20.7	19.1	24.4	31.1	29.4	23.7	Coeff. of Variation	
	0	-2	0	4	15	21	15	26	% Inhibition	
1	15	21	81	150	320	597	712	1059	0.12	
2	15	25	80	151	268	434	548	576		
3	15	21	72	129	231	407	516	546		
	15	22	78	143	273	479	592	727	Mean	
	0	2	5	12	45	103	105	288	Std. Dev.	
	0.0	10.3	6.4	8.7	16.4	21.4	17.8	39.6	Coeff. of	

									Variation
	0	-5	5	0	11	16	12	13	% Inhibition
1	15	26	88	144	284	445	576	625	0.24
2	15	19	75	134	244	433	592	608	
3	15	21	79	145	259	446	536	562	
	15	22	81	141	262	441	568	598	Mean
	0	4	7	6	20	7	29	33	Std. Dev.
	0.0	16.4	8.3	4.3	7.7	1.6	5.1	5.4	Coeff. of Variation
	0	-3	2	1	15	23	15	29	% Inhibition
1	15	19	31	44	62	109	206	273	0.48
2	15	23	32	50	61	116	196	238	
3	15	21	36	49	84	206	350	414	
	15	21	33	48	69	144	251	308	Mean
	0	2	3	3	13	54	86	93	Std. Dev.
	0.0	9.5	8.0	6.7	18.8	37.7	34.4	30.2	Coeff. of Variation
	0	2	60	67	78	75	63	63	% Inhibition
1	15	18	22	34	29	31	31	30	0.96
2	15	18	24	26	37	40	37	38	
3	15	17	28	31	39	41	40	43	
	15	18	25	30	35	37	36	37	Mean
	0	1	3	4	5	6	5	7	Std. Dev.
	0.0	3.3	12.4	13.3	15.1	14.8	12.7	17.7	Coeff. of Variation
	0	17	70	79	89	93	95	96	% Inhibition

Calculated growth rate by test interval

The mean frond count is determined using the number of fronds observed in a test beaker on a given observation day.

Growth rate is calculated for each treatment group and control group based on frond count (or biomass). Growth rate is calculated in this analysis using frond count data and the following formula:

$$\mu = \frac{\ln N_n - \ln N_0}{t_n}$$

where:

μ = Average specific growth rate

N_0 = Number of fronds (or biomass) at the beginning of the test

N_n = Number of fronds (or biomass) at t_n

t_n = Time of nth measurement after beginning of test (days).

Inhibition is calculated for each treatment group as the percent reduction in mean frond count and mean growth rates relative to the respective control means. The following formula was used:

$$\% I = \frac{C - T}{C} \times 100$$

where:

C = Control mean frond count or growth rate

T = Treatment group mean frond count or growth rate

The results are presented in Table 3.

TABLE 3. CALCULATED GROWTH RATE BY TEST INTERVAL BASED ON FROND COUNTS

Nominal Chlorsulfuron Concentration µg/L	Rep	Exposure Initiated:Day 0: Exposure Ended:Day 14:						
		Growth Rate Based on Count (Fronds) by Test Day						
		Day 0- 1	Day 0- 4	Day 0- 6	Day 0- 8	Day 0- 11	Day 0- 13	Day 0- 14
Blank Control	1	0.3830	0.4366	0.3650	0.3466	0.3018	0.2669	0.2545
	2	0.3830	0.4185	0.3758	0.3833	0.3507	0.3070	0.3013
	3	0.2877	0.4185	0.3860	0.3969	0.3329	0.2974	0.2974
Mean		0.3512	0.4245	0.3756	0.3756	0.3285	0.2904	0.2844
Std. Dev.		0.0550	0.0105	0.0105	0.0260	0.0247	0.0209	0.0260
Coeff. of Variation		15.7	2.5	2.8	6.9	7.5	7.2	9.1
0.06	1	0.3830	0.3704	0.3452	0.3322	0.2848	0.2585	0.2495
	2	0.4274	0.4743	0.4017	0.3875	0.3364	0.3015	0.2826
	3	0.2877	0.4216	0.3534	0.3439	0.2973	0.2728	0.2628
Mean		0.3660	0.4221	0.3668	0.3545	0.3062	0.2776	0.2650
Std. Dev.		0.0714	0.0520	0.0305	0.0291	0.0269	0.0219	0.0167
Coeff. of Variation		19.5	12.3	8.3	8.2	8.8	7.9	6.3
% Inhibition		-4	1	2	6	7	4	7
0.12	1	0.3365	0.4216	0.3838	0.3825	0.3349	0.2969	0.3041
	2	0.5108	0.4185	0.3849	0.3604	0.3059	0.2768	0.2606
	3	0.3365	0.3922	0.3586	0.3418	0.3001	0.2722	0.2568
Mean		0.3946	0.4108	0.3758	0.3616	0.3136	0.2820	0.2738
Std. Dev.		0.1006	0.0162	0.0149	0.0204	0.0186	0.0131	0.0263
Coeff. of Variation		25.5	3.9	4.0	5.6	5.9	4.7	9.6
% Inhibition		-12	3	0	4	5	3	4
0.24	1	0.5500	0.4423	0.3770	0.3676	0.3082	0.2806	0.2664
	2	0.2364	0.4024	0.3650	0.3486	0.3057	0.2827	0.2644
	3	0.3365	0.4153	0.3781	0.3561	0.3084	0.2751	0.2588
Mean		0.3743	0.4200	0.3734	0.3574	0.3074	0.2795	0.2632
Std. Dev.		0.1602	0.0204	0.0073	0.0096	0.0015	0.0039	0.0039
Coeff. of Variation		42.8	4.8	1.9	2.7	0.5	1.4	1.5
% Inhibition		-7	1	1	5	6	4	7
0.48	1	0.2364	0.1815	0.1794	0.1774	0.1803	0.2015	0.2072
	2	0.4274	0.1894	0.2007	0.1754	0.186	0.1977	0.1974
	3	0.3365	0.2189	0.1973	0.2153	0.2382	0.2423	0.2370
Mean		0.3334	0.1966	0.1925	0.1894	0.2015	0.2138	0.2139
Std. Dev.		0.0955	0.0197	0.0114	0.0225	0.0319	0.0247	0.0206
Coeff. of Variation		28.7	10.0	5.9	11.9	15.8	11.6	9.6
% Inhibition		5	54	49	50	39	26	25

0.96	1	0.1823	0.0957	0.1364	0.0824	0.0660	0.0558	0.0495
	2	0.1823	0.1175	0.0917	0.1129	0.0892	0.0695	0.0664
	3	0.1252	0.1560	0.1210	0.1194	0.0914	0.0754	0.0752
Mean		0.1633	0.1231	0.1164	0.1049	0.0822	0.0669	0.0637
Std. Dev.		0.0330	0.0305	0.0227	0.0198	0.0141	0.0101	0.0131
Coeff. of Variation		20.2	24.8	19.5	18.8	17.1	15.0	20.5
% Inhibition		54	71	69	72	75	77	78

Results

Statistical analyses are reported based on nominal concentrations and were conducted using SAS Version 9.4. The 7 and 14 day E_rC_{50} values (and 95% confidence intervals) for growth rate based on frond count and nominal concentrations were obtained with the 3-parameter exponential model (1 of 5 models used for toxicity experiments advocated by Slob (2002)).

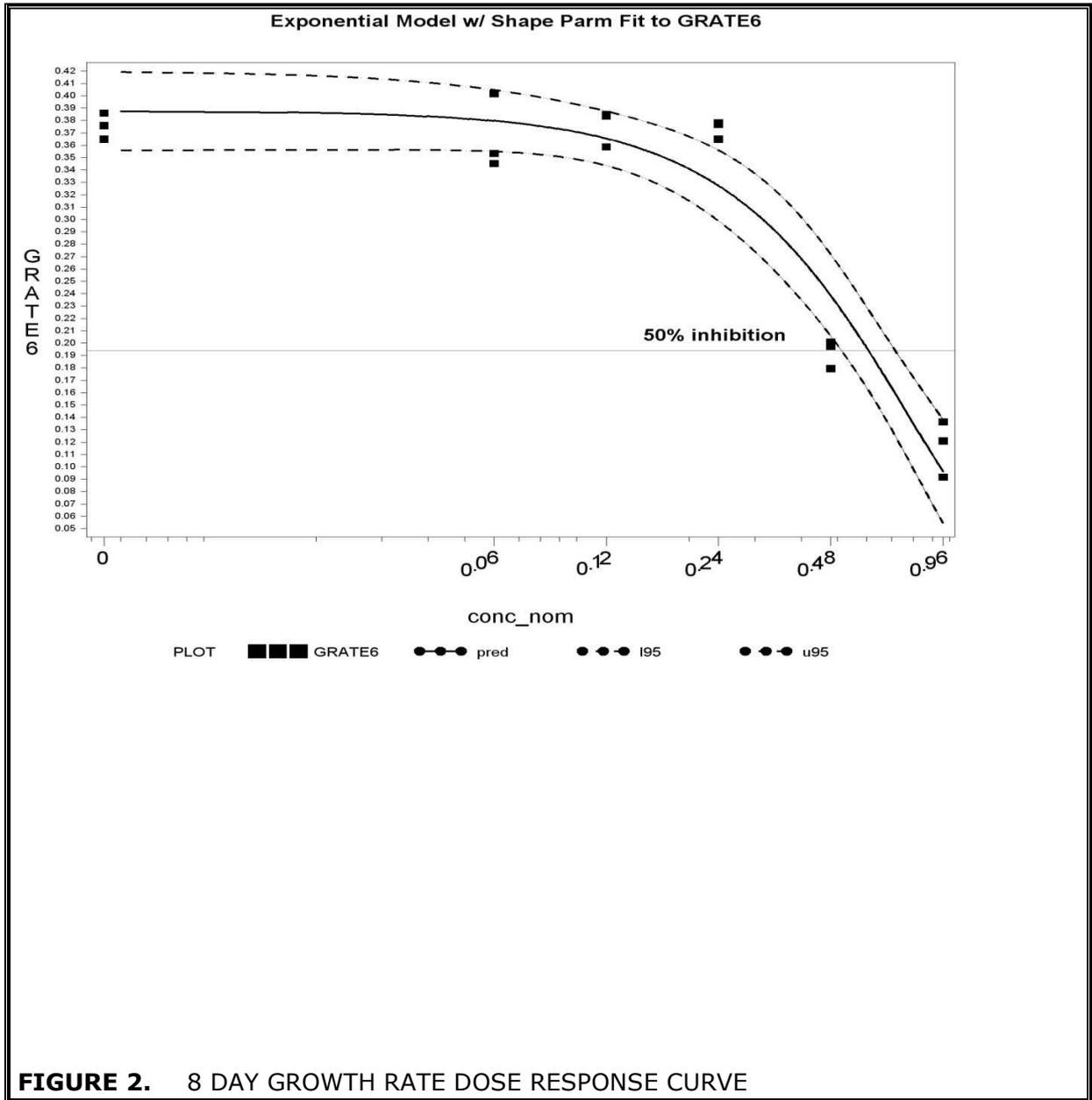
Determination of the 7 and 14 day E_rC_{50} and NOEC values

A complication in this analysis was that no observations of effects were measured at day 7. Instead, measures were made at 6 and 8 days of exposure. However, since there was consistency in results from 6 and 8 days exposure, it was possible to obtain meaningful NOEC determinations and E_rC_{50} estimates for 7 days of exposure from the data available.

The E_rC_{50} estimates for day 6 and day 8 were 0.6067 and 0.5868 $\mu\text{g a.s./L}$, with 95% confidence intervals of (0.4884, 0.7250) and (0.4760, 0.6975), respectively. Consequently, the E_rC_{50} estimate for 7 days of exposure is the geometric mean of the day 6 and day 8 estimates. The calculated 7 day E_rC_{50} = 0.5967 $\mu\text{g a.s./L}$, with approximate 95% confidence intervals of (0.4760, 0.7250).

The 14 day E_rC_{50} based on average specific growth rate has been recalculated previously (McKelvey, 2011) and resulted in an E_rC_{50} = 0.69 $\mu\text{g/L}$. The 14 day E_rC_{50} calculated in this analysis was determined to be 0.71 $\mu\text{g/L}$. The difference observed in these two values is not significant, and is most likely due to differences in rounding of the raw data.

FIGURE 1. 6 DAY GROWTH RATE DOSE RESPONSE CURVE



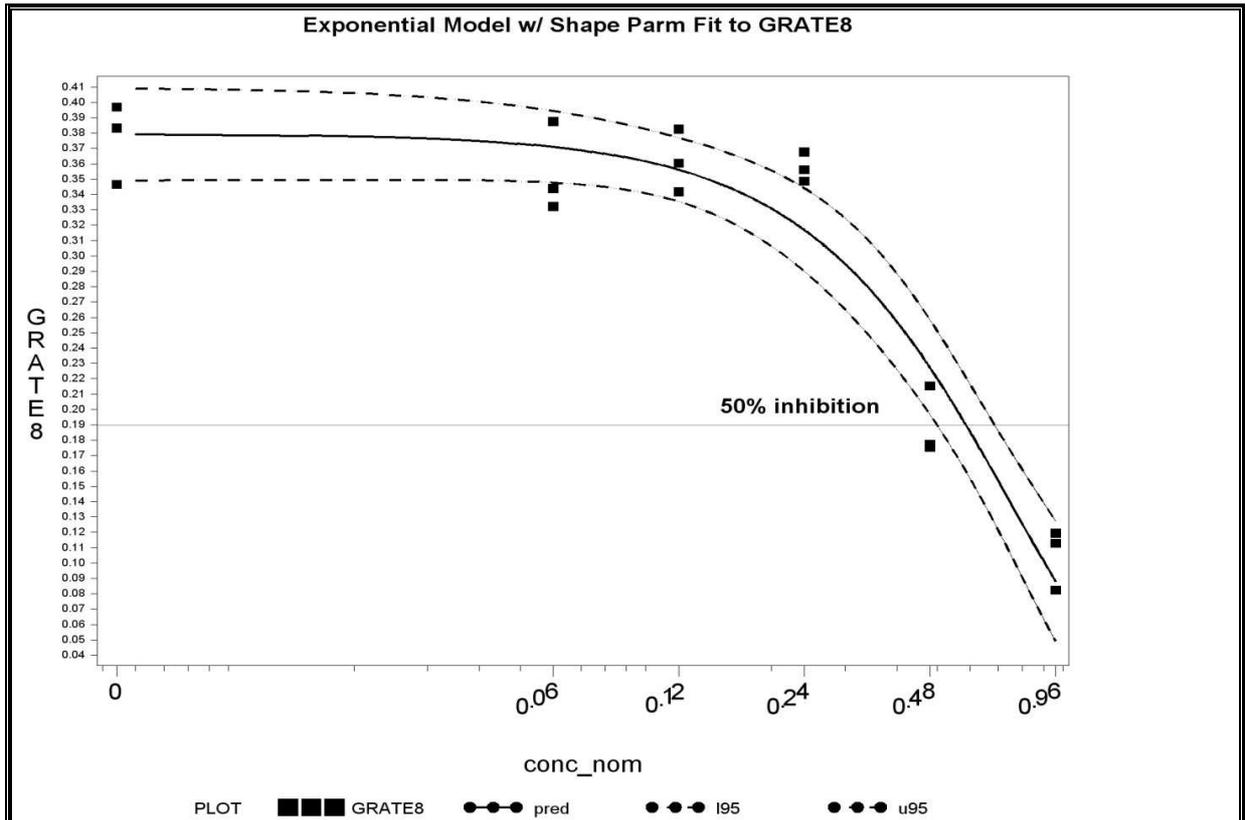
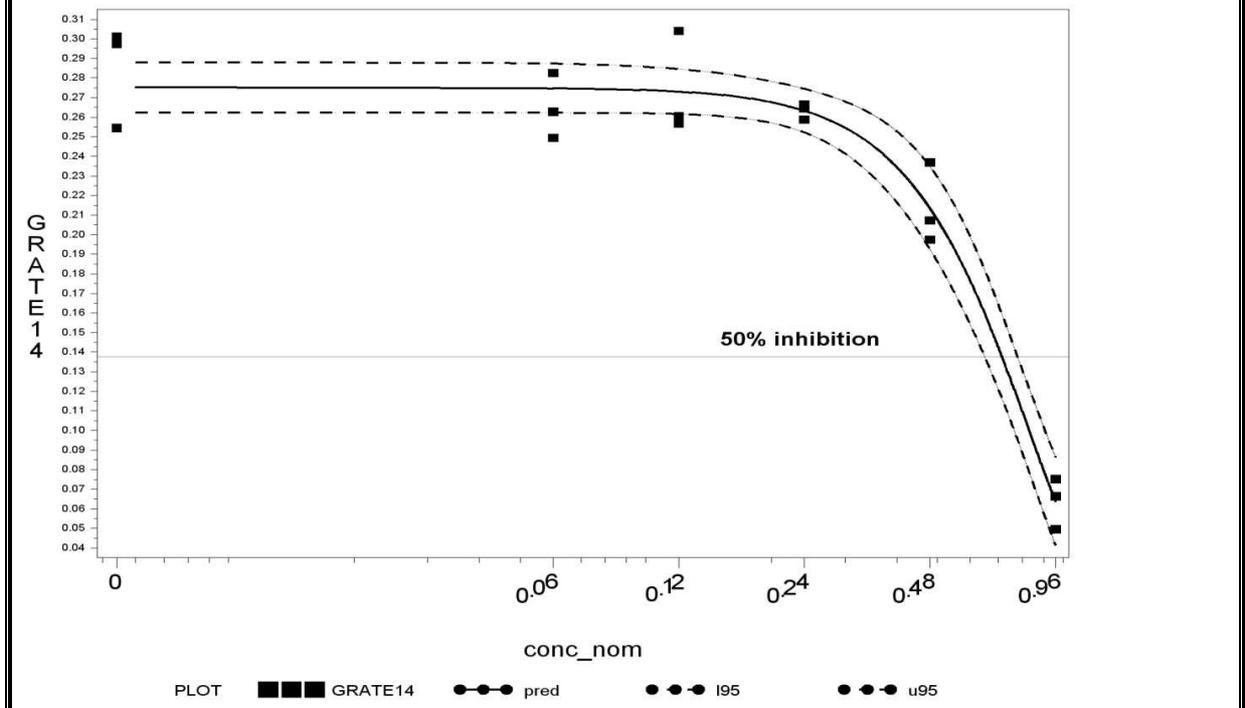


FIGURE 3. 14 DAY GROWTH RATE DOSE RESPONSE CURVE
Exponential Model w/ Shape Parm Fit to GRATE14



Recalculation of 7 and 14 day E_rC_{50} and NOEC values based on geometric mean

concentration

The E_rC_{50} and NOEC for *Lemna Giba* growth rate endpoint based on the geometric mean of initially measured concentrations and one half of LOQ (LOQ for chlorsulfuron is equal to 0.0132 µg/L) were recalculated (Table 4). The results for 14 days test are the following: The E_rC_{50} is 0.064 µg/L and the NOEC is 0.04 µg/L. There are no available measured test concentrations on the days 6 and 8, the recalculation of E_rC_{50} and NOEC based on geometric mean of initial concentrations and half of LOQ is rather speculative. Despite the recalculation was performed and the resulting values for 7 days test were the following: E_rC_{50} equal to 0.0595 µg/L and the NOEC to 0.04 µg/L.

Table 4. 7 and 14 day E_rC_{50} and NOEC Values Based on Growth Rate

Response	Day	NOEC (µg/L)	Concentrations	E_rC_{50} (µg/L)	95% Confidence Intervals
Growth rate	7	0.24	Nominal	0.5967	(0.4760, 0.7250)
	7	0.04	Geometric mean	0.0595	(0.0563, 0.0627)
	14	0.24	Nominal	0.715 ^a	(0.6438, 0.7857)
	14	0.04	Geometric mean	0.064	(0.0618, 0.0662)

Details of Douglas *et al.* (1988) study

(DAR 07, Vol 3, Annex B, part 5, B.9)

Test Substance: DPX-W4189 technical, purity: 98.5%

Test organism: *Lemna minor*

Medium: algal nutrient medium pH = 5

GLP: Yes

Medium renewed on days 2, 5, 7, 9, 12

14-day E_rC_{50} = 0.11 µg/L

14-day NOE_rC = 0.04 µg/L

Regarding the Douglas *et al.* (1988) study, the endpoints are based on the growth rate. The NOE_rC for 14-day test duration was 0.04 µg/L and is the same as NOEC derived from recalculated data of the Boeri *et al.* (2002) study. In RAC's opinion, both NOEC results are derived from methods which are designed for compounds not stable in the test solution. On contrary, chlorsulfuron stability was proved during 21 day test period; the compound is not readily biodegradable and bioaccumulation is not expected because of the low log K_{ow} (at pH 7, log K_{ow} = -0.99). The very low concentration (below the LOQ) at the end of the 14 days study could be explained by uptake of the test compound to the test organism. Unfortunately this hypothesis cannot be verified since no information on the chlorsulfuron concentration in the test organisms is available.

6 OTHER INFORMATION

No other data available for consideration in determining the classification of chlorsulfuron.

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8 ANNEXES

IUCLID5 file