FINAL REGISTRATION REPORT

Part B

Section 10

Assessment of the relevance of metabolites in groundwater

Detailed summary of the risk assessment

Product code: CHR/H/FDF 574 SC

Product name(s): Cezaro 574 SC/ Huron 574 SC

Chemical active substance(s):

Florasulam, 12 g/L

Diflufenican, 250 g/L

Flufenacet, 312 g/L

Central Zone

Zonal Rapporteur Member State: Poland

CORE ASSESSMENT

(authorization)

Applicant: Innvigo Sp. z o.o.

Submission date: November 2021

MS Finalisation date: 21/11/2022

Version history

When	What
March 2022	Dossier sent for evaluation
June 2022	Updates based on feedback from zRMS Poland
September 2022	zRMS evaluation of dRR
November 2022	Final version prepared by zRMS after Commenting period

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zRMS comments:	The applicant´s text in this Section has not been amended by the zRMS. If necessary zRMS has crossed text and/or inserted new text written in gray.

10 Relevance of metabolites in groundwater

zRMS Comments:	The PECgw values for relevant metabolites of active substances were corrected, if relevant, in accordance with PECgw values presented in dRR Section 8. **Florasulam**. Based on PECgw assessment all relevant metabolites with concentration higher than trigger value of 0.1 µg/L were considered. **Flufenacet**. The only metabolite FOE-sulfonic acid has to be considered. The PECgw value for this metabolite was above the trigger value of 10 µg/L for winter cereals in Jokioinen scenario and was not taken into account as this scenario is not relevant for Central Zone. **Diflufenican**. The PECgw values for metabolites of active substance were below the trigger value of 0.1 µg/L. The relevance of metabolites will be discussed below. <u>ASTCA, TSA and 5-OH Florasulam</u> Genotoxicity studies were evaluated as part of the EU review process for florasulam. Results of these studies included in the EFSA Conclusions (EFSA Journal 2015;13(1): 3984). The metabolites did not demonstrate any genotoxic potential in an Ames test, a gen mutation assay *in vitro* with mammalian cells (CHO/HGPRT) and chromosomal aberration assay *in vitro*. Florasulam is not classified as acutely or chronically toxic or very toxic and is also not classified for reproductive toxicity and carcinogenicity. ASTCA, TSA and 5-OH Florasulam are not considered to be toxicologically relevant. <u>FOE-sulfonic acid</u> The metabolite did not demonstrate any genotoxic potential in an Ames test, a gene mutation assay *in vitro* with mammalian cells and *in vitro* micronucleus assay. The metabolite was exceed 0.75 µg/L in groundwater modelled scenario. Therefore, a refined exposure and risk assessment according to Step 5 was required. The estimated exposure for children and infants exceed the allocation factor of 20% based on the WHO Guidelines for drinking water. Therefore, exposure from other routes was assessed. Total exposure did not exceed the ADI.

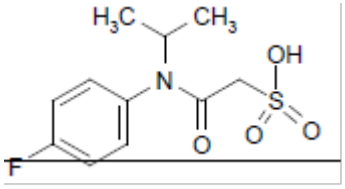
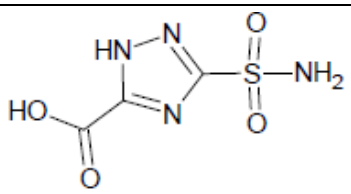
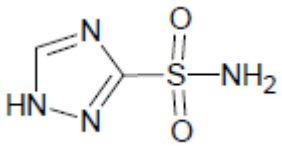
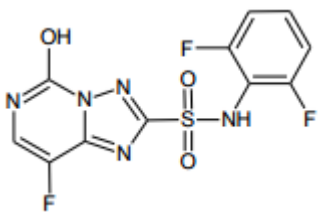
In the following document, data for active substances - diflufenican and flufenacet - was described during its inclusion on Annex 1 process in respectively 2009 and 2004 . Were reference to active substance data in the current risk assessment has been made, it was based on the data which protection for expired 10 years from date of inclusion of active substances on Annex I.
Data matching studies for florasulam have been evaluated by Poland. As a result of the assessment all reports were accepted and considered as equivalent to protected studies. Therefore, to support the authorization of CHR/H/FDF 574 SC INNVIGO is allowed to refer to EU approved reports

10.1 General information

The metabolites FOE sulfonic acid, 5-ASTCA, TSA and 5-OH Florasulam are predict predicted to occur in groundwater at concentrations above 0.1 µg/L (see PART B Section 8 of CHR/H/FDF 574 SC). Assessment of the relevance of these metabolites according to the stepwise procedure of the EC guidance document SANCO/221/2000 –rev.10 is therefore required.

General information on the metabolites provided in Table 10.1-1. The impact of the relevance assessment on whether a particular GAP use leads to acceptable risk or not is presented in the summary of the cGAP evaluation in chapter KCP 9.2.4 of the dRR Part B, Section 8 (Environmental fate and behaviour).

Table 10.1-1: General information on the metabolite(s)

Name of active substance	Metabolite name and code	Structural/molecular formula	Trigger for relevance assessmnent	
Flufenacet	FOE sulfonic acid		Max PEC_{gw} Based on:	8.679618 µg/L Focus PEARL 4.4.4 Jokoinen 14.045 µg/L 9.0484 µg/L Focus PEARL 4.4.4 ~~Jokoinen~~ Thiva Winter cereals
Florasulam	ASTCA		Max PECgw Based on:	~~0.4430~~ 0.433 µg/L Focus PEARL 4.4.4 ~~Thivia~~ Thiva Winter cereals
	TSA		Max PECgw Based on:	0.3364 µg/L Focus PEARL 4.4.4 ~~Thivia~~ Thiva Winter cereals
	5-OH Florasulam		Max PECgw Based on:	0.202 µg/L Focus PELMO 5.5.3 Porto Winter cereals

10.2 Relevance assessment of FOE sulfonic acid

Summary:

A summary of the relevance assessment is given in Table 10.2-1 and the corresponding studies are listed in the corresponding sections.

Table 10.2-1: Summary of the relevance assessment for FOE sulfonic acid

	Assessment step	Result of assessment

	STEP 1		Metabolite of no concern?	~~Yes~~ No
for groundwater	STEP 2		Max PEC_{gw}	~~14.045~~ 9.0484 µg/L
			Based on	FOCUS PEARL, ~~Jokioinen~~ Thiva
Hazard assessment	STEP 3	Stage 1	Biological activity comparable to the parent?	No
		Stage 2	Genotoxic properties of metabolite	Non-genotoxic
		Stage 3	Toxic properties of metabolite;	Not toxic or very toxic (T or T+)
			Classification of parent	not currently classified as toxic or very toxic
			Classification of metabolite	not currently classified as toxic or very toxic
Consumer health risk assessment	STEP 4		Estimated consumer exposure via drinking water and other sources; threshold of concern approach	>0.75 µg/L
	STEP 5		Refined risk assessment	Required
			Predicted exposure (% of ADI)	~~57.2%~~ 35.8 % for infant ~~38.2%~~ 23.88 % for toddler ~~12.8%~~ 7.96 % for adult
			ADI based on	Active substance Flufenacet

* N/A: not applicable

10.2.1 STEP 1: Exclusion of degradation products of no concern

The major metabolites, FOE sulfonic acid contain a phenyl ring and therefore are not aliphatic compounds. Hence, these metabolites are not automatically metabolite of no concern.
Therefore an assessment has been conducted as given in the EU Guidance Document on the assessment of the relevance of metabolites in groundwater (Anonymous, 2003; Sanco/221/2000 - rev. 10, 25th February 2003).

10.2.2 STEP 2: Quantification of potential groundwater contamination

The quantity of metabolites potentially present in the soil water at 1m depth with the potential to move to groundwater depends upon the active ingredient, the application rate, the timing, the crop intercept, the plant uptake factor, the soil degradation rate and the soil adsorption constant and a range of other factors.
Hence the values in groundwater for all metabolites prone to leaching are quite variable depending upon their degradation properties and of cause the use pattern and application rate.
Therefore the risk assessments will be performed for a range of possible concentrations. Therefore the quantities of the metabolites reaching soil water at 1 m depth are grouped into classes of concentrations ranging from < 0.1 µg/L, >0.1 µg/L and ≤0.75 mg/L, >0.75 µg/L and ≤ 10 µg/L, >10 µg/L and ≤ 20 µg/L. The risk assessments will consider the upper limit of each class of possible concentrations per metabolite. Metabolites with groundwater concentration < 0.1 µg/L are not required to be further assessed according the SANCO/221/2000 assessment scheme and thus will not be subject to further assessments.

PEC_{gw} for flufenacet and metabolite(s) on winter cereals (with FOCUS PEARL 4.4.4)

Crop	**Scenario**	**80th Percentile PEC_{gw} at 1 m Soil Depth (µg/L)**		
		Flufenacet	**FOE Sulfonic acid**	**FOE oxalate**
Winter	Châteaudun	<0.0001	8.6327	0.0004

cereals	Hamburg	<0.0001	8.5117	0.0111
	Jokioinen	<0.0001	14.045	0.007117
	Kremsmünster	<0.0001	5.2870	0.0019
	Okehampton	<0.0001	4.856	0.01817
	Piacenza	<0.0001	5.1420	0.0017
	Porto	<0.0001	4.2630	0.0135
	Sevilla	<0.0001	4.034	<0.0001
	Thiva	<0.0001	9.0484	~~14.045~~ <0.0001

PEC_{gw} for ~~penoxsulam~~ flufenacet and metabolite(s) on winter cereals (with FOCUS PELMO 5.5.3)

Crop	Scenario	80th Percentile PEC_{gw} at 1 m Soil Depth (µg/L)		
		Flufenacet	FOE Sulfonic acid	FOE oxalate
Winter cereals	Châteaudun	<0.001	5.973	<0.001
	Hamburg	<0.001	6.354	0.001
	Jokioinen	<0.001	7.677	<0.001
	Kremsmünster	<0.001	4.837	<0.001
	Okehampton	<0.001	3.731	0.001
	Piacenza	<0.001	5.466	<0.001
	Porto	<0.001	3.258	0.001
	Sevilla	<0.001	2.637	<0.001
	Thiva	<0.001	4.713	<0.001

Lysimeter studies:

- **corn/wheat rotation (480 + 180 g a.s./ha)**

Total mean 2.5 µg/l, max. 5 µg/l (year 1)
mean 0.24 µg/l (year 2)
a.s. not identified
FOE oxalate and thioglycolate < 0.1 µg/l
FOE sulfonic acid :
mean 1.49 µg/l, **max. 3.7 µg/l (year 1)**
mean 0.015 µg/l (year 2)

Conclusions: The maximum concentration of FOE 5043 in single leachates were measured to be lower than 0.01 µg/l, however, a defined peak of the parent compound could not be observed. Therefore, FOE 5043 was not positively identified in the leachate. The same is true for FOE oxalate (M1, ≤0.04 µg/l). FOE alcohol (M3) was only positively identified in lysimeter #18, especially in the early leachates in maximum concentrations of 0.16 µg/l. Whereas FOE thioglycolate sulfoxide was only detected in amounts of 0.028 µg/l, the maximum residues of FOE sulfonic acid (M2) in the leachates of February 1994 were 3.4 µg/l (lys #17) and 3.7 µg/l (lys #18). Maximum concentrations of radioactivity remaining at the start and being spread over the whole TLC plate were 0.31 and 0.37 µg/l, respectively, and unknown single metabolites were below 0.08 µg/l.

The results of this study proved that even under worst case conditions a contamination of soil layers below 1.2 m depth by parent compound can be precluded with high probability. Most of ist relevant degradation products in soil showed a similar behaviour. Furthermore, the data confirmed that FOE 5043 is well degradable in soil.

10.2.3 STEP 3: Hazard assessment – identification of relevant metabolites

FOE sulfonic acid is not relevant according to the hazard screening outlined in Step 3. See assessment below (point 10.2.3.1 – 10.2.3.3)

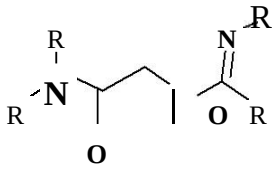
10.2.3.1 STEP 3, Stage 1: screening for biological activity

No screening information was submitted but an assessment based on metabolism and structure-activity relationship based on the two following publications :

Förster, H., Schmidt, R.R., Santel, H.J., Andree, R., 1997, FOE 5043 - a new selective herbicide from the oxyacetamide group; Pflanzenschutz-Nachrichten Bayer 50/1997, 2, pp 105-116

Bieseler, B., Fedtke, C., Neuefeind, T., Etzel, W., Prade, L., Reinemer, P., 1997, Maize selectivity of FOE 5043: Degradation of active ingredient by glutathion-S-transferases, Pflanzenschutz-Nachrichten Bayer 50/1997, 2, pp 117-140.

As outlined by Förster *et al.* (1997) the herbicidal activity of Flufenacet is closely connected to the following active central structure:



Any change in this central structure leads to a significant decrease or even loss of the herbicidal activity.

The tolerance of crops towards Flufenacet depends mainly on a glutathion-conjugation of Flufenacet within the metabolic degradation in the plant. In this conjugation step a cystein conjugate is formed and the central structure, where the herbicidal activity is located, is irreversibly destroyed. Further metabolic steps are of no further importance for selectivity (Bieseler et al.,1997). Studies on the metabolism of Flufenacet in animals, soil and water showed that all tested organisms, i.e. mammals, soil and water microorganisms, used the same conjugation with glutathion as the initial step of the metabolism and detoxification of Flufenacet.

It could be demonstrated that the metabolite M2 Flufenacet-sulfonic acid are formed later in the metabolic pathway (see Annex II, point 5, 6, 7). If the formula for parent and metabolites are compared, it is evident that the biological activity of the metabolite M2 is expected to be at least lower than for the active ingredient, if not completely lost.

Therefore herbicidal activity of M2 Flufenacet-sulfonic acid is very unlikely. Furthermore it could be demonstrated that M2 (Flufenacet-sulfonic acid) is clearly less toxic to non-target organisms (fish, Daphnia, algae, Lemna, earthworm, mammals) than the active ingredient.

Based on these results it is concluded that FOE sulfonic acid has no herbicidal activity.

10.2.3.2 STEP 3, Stage 2: screening for genotoxicity

zRMS	The metabolites FOE sulfonic acid was screened for genotoxicity activity by the following data package of *in vitro* genotoxicity studies: Ames test, gene mutation test

comments:

with mammalian cells and *in vitro* mammalian cell micronucleus test. The results are negative. It can be concluded that FOE sulfonic acid is considered to be non-genotoxic. Summary of genotoxicity data are presented in table below.

Study	Test object/concentration	Results
Bacterial reverse mutation assay [Herbold B., 2000; M-019064-01-1]	*S. typhimurium*, TA1535, TA1537, TA98, TA100, TA102/ 16 - 5000 µg/plate (+/- S9 mix)	Negative
In vitro micronucleus assay (MNA) [Antonik, J, 2016, Study Number: K81/JA/01]	CHO-K1 cells 250-2000 µg/ml (+/- S9 mix)	Negative
In vitro mammalian cell gene mutation (MLA) [Antonik J.; Study number: K82/JA/01]	L5178Y TK+/− / 250 – 2000 µg/mL (+/- S9 mix)	Negative

The genotoxic studies:
- in vitro micronucleus assay (MNA); Antonik, J, 2016, Study Number: K81/JA/01
- in vitro mammalian cell gene mutation (MLA); Antonik J.; Study number: K82/JA/01

are evaluated in Part B, Section 6, Point A 2.11.2.

According to DAR Flufenacet and Section B6, Point 6.4.4.in core dossier:
FOE 5043 sulfonic acid was initially investigated using the salmonella/microsome plate incorporation test (*Salmonella/microsome test with FOE 5043 sulfonic acid. Report Bayer PH 29473. GLP. Unpublished. B. Herbold, 2000.*) for point mutagenic effects in doses of up to and including 5000 µg per plate on five Salmonella typhimurium LT2 mutants. These comprised the histidine-auxotrophic strains TA 1535, TA 100, TA 1537, TA 98 and TA 102. The independent repeat was performed as preincubation for 20 minutes at 37°C. Other co ditions remained unchanged.
Doses up to and including 5000µg per plate did not cause any bacteriotoxic effects : total bacteria counts remained unchanged and no inhibition of growth was observed. Evidence of mutagenic activity of FOE 5043 sulfonic acid was not seen. No biologically relevant increase in the mutant count, in comparison with the negative controls was observed.
The positive controls sodium azide, nitrofurantoin, 4-nitro-1,2-phenylene diamine, cumene hydroperoxyde and 2-aminoanthracene had a marked mutagenic effect, as was seen by a biologically relevant increase in mutant colonies compared to the corresponding negative controls.

Additional two studies were provided ~~by PUH Chemiro~~ Section B6, Point 6.4.4 1 in core dossier~~:~~:
- *In vitro evaluation of Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid, Trifluoroacetic acid and Flufenacet methylsulfone genotoxicity using the micronucleus assay (MNA).* J.Antonik, 2016.; SELVITA. Study Number: K81/JA/01.Method: OECD 487
- *In vitro Mammalian Cell Gene Mutation test (OECD 490) - genotoxicity determination of Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid and Trifluoroacetic acid by Mouse Lymphoma Assay.* J.Antonik; 2016; J.Antonik; Study number: K82/JA/01; SELVITA; OECD 490

The formation of MN is a consequence of chromosomal breakage and/or spindle-fiber dysfunction induced by clastogens and/or aneuploidogens. The present study was performed in accordance with the OECD 487 and under GLP requirements. In order to assess genotoxic potential CHO-K1 cells were exposed to test items (Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid, Trifluoroacetic acid and Flufenacet methylsulfone) and appropriate reference items in system with (+S9) and without (-S9 short and extended treatment) an exogenous metabolic activation. Statistical analysis of the MN frequency and binucleate cells with MN was performer using the Chi-square test with Yates'

correction. To examine the dose-response relationship in frequencies of the micronuclei Chi-square test for trend was performed.
None of tested concentration of Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid, Trifluoroacetic acid and Flufenacet methylsulfone exhibit a statistically significant increase in MN frequency compared with the concurrent negativecontrol (P>0.05, T bles IX-XIII, Figure I-VII). Chi-square test for trend revealed no dose-related increase in MN frequency (P>0.05).
Results for positive reference items (mitomycin C and cyclophosphamide) demonstrated reproducibility and sensitivity of system.

In summary, the present research has demonstrated that items Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid, Trifluoroacetic acid and Flufenacet methylsulfone did not produce dose-dependent genetic toxicity in the CHO-K1 cells.

Mutagenic potential of Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid and Trifluoroacetic acid was evaluated trough Mouse Lymphoma Assay (MLA) in L5178Y cells. Tested items were analyzed in MLA, in the presence and absence of exogenous metabolic activation.Obtained results have shown that tested item did not exceed MF above a value termed as
Global Evaluation Factor 126 ×10−6 in any of the tested doses both in the presence and absence of S9 exogenous activation system.
Obtained results indicate that tested items (Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid and Trifluoroacetic acid) or their metabolic derivatives were not positive in Mouse Lymphoma Assay under the protocol described and according to the acceptability criteria defined in OECD guideline 490 and SPB-19.

Conclusion :
FOE 5043 sulfonic acid is considered to be non-mutagenic and have no genotoxicity potential in all of these assays.

10.2.3.3 STEP 3, Stage 3: screening for toxicity

zRMS comments:	The parent compound flufenacet is not classified as acutely or chronically toxic or very toxic and is also not classified for reproductive toxicity and carcinogenicity. According to the CLP regulation flufenacet is classified as follows: - Acute Tox 4; H302 - Skin Sens. 1, H317 - STOT RE 2, H373 Metabolite FOE sulfonic acid is considered not relevant and is further evaluated in Step 4.

FOE 5043 sulfonic acid plasmakinetics and excretion in urine in a rat study with single oral vs intravenous administration. Report PH 30052. GLP. Unpublished. F. Krötinger, U Schmidt, 2000.
A study for the comparison of the plasmakinetics and excretion in urine after the single oral versus intravenous administration of FOE 5043 sulfonic acid, a metabolite of FOE 5043, was conducted in male Wistar rats.
The treatment and observation time of the study has been designed to follow the principles of the following guidelines : OECD-Guideline for Testing of Chemicals No 423.

The oral AUC was despite the 10-fold higher dose slightly lower than the intravenous AUC, which also argued in favour of low oral absorption.
The t1/2 after iv administration was short (about 30mn) which suggests the major role of the renal clearance.
FOE 5043 Sulfonsäure : study for acute oral toxicity in rats. Report 27137. GLP. Unpublished. F. Krötinger, 1998.

A study for acute oral toxicity in male and female Wistar rats was conducted with the test substance FOE 5043 Sulfonsäure (soil metabolite of FOE 5043).
The method used complied with the OCDE-guideline for Testing of Chemicals; section4: Health effects, No. 401 –"Acute oral Toxicity".
Clinical findings :
Doses of 500 and 2000 mg/kg body weight were tolerated by male and female rats without mortalities and 500mg/kg b.w. also without clinical signs. At 2000 mg/kg b.w. in both sexes diarrhea occurred and anuses were moistened. The signs observed started 4 hours and lasted up to 5 hours after administration.
Body weight and body weight gain were not affected by treatment.
The acute oral LD50 of FOE 5043 sulfonic acid is > 2000 mg /kg b.w.

Conclusion on toxicological significance of the metabolite FOE sulfonic acid:

The goal of the additional physico-chemical and biological experiments which were requested from the applicant was to demonstrate that the metabolite M2 (FOE sulfonic acid) was poorly absorbed orally, and had a low potential toxicity.

Based firstly on the physical properties that show a high hydrosolubility which suggests a low biological absorption,
- The solubility in water is 55g/l at 20°C at pH 4 to 9 ; under the same conditions, its Kow is 0.0019, leading to a log Kow = -2.72 ; the pKa of sulfonic acid is <1

and secondly on the biological investigations that show poor biological disposition and low toxicity, the metabolite FOE 5043 sulfonic acid is considered of no toxicological relevance.

10.2.4 STEP 4: Exposure assessment – threshold of concern approach

Metabolites which have not been identified as being relevant according to the hazard screening outlined in Step 3, should be further tested in an exposure assessment to make sure that any contamination of groundwater will not lead to unacceptable exposure of consumers via their drinking water.

The potential exposure to metabolite FOE sulfonic acid is > 0.75 µg/L but <10 µg/L. A further assessment in Step 5 is required.

Concentrations in groundwater for the metabolites prone to leaching are quite variable depending upon their degradation properties, the use pattern and application rates. Therefore the risk assessments are be performed for a range of possible concentrations. The quantities of the metabolites reaching soil water at 1 m depth are grouped into classes of concentrations ranging from >0.1 µg/L to ≤0.75 mg/L, >0.75 µg/L to ≤ 10 µg/L, and >10 µg/L to ≤ 20 µg/L. The risk assessments will consider the upper limit of each class of possible concentrations per metabolite.

Based on the concentration predicted by groundwater modeling the intake of FOE Sulfonic acid by means of drinking water could potentially exceed the threshold of 0.75 µg/L. Therefore the "threshold of concern" is applicable for these metabolites and FOE sulfonic acid is subject to refined risk assessments (Step 5).

10.2.5 STEP 5: Refined risk assessment

zRMS comments:

Calculations based on max PECgw = 9.0482 µg/L (FOCUS PEARL, Thiva)

Conversion of gw concentration expressed as metabolite to parent equivalents

Metabolite	Estimated upper limit gw concentration expressed as metabolite [µg/L]	Estimated upper limit gw concentration expressed as parent equivalent [µg/L]
FOE sulfonic acid (M02)	9.0484	11.94

Molar mass flufenacet 363.3 g/mol
Molar mass FOE 5043 sulfonic acid 275.3 g/mol

Calculation of risk for 5-kg bottle-fed infant (consuming 0.75 l/day)

Infant	TMDI (mg/kg bw/day)	% of ADI
	0.00179	35.8

Calculation of risk (% ADI) for 10-kg child (consuming 1.0 l/day):

Child	TMDI (mg/kg bw/day)	% of ADI
	0.00119	23.88

Calculation of risk (% ADI) for 60-kg adult (consuming 2.0 l/day):

Adult	TMDI (mg/kg bw/day)	% of ADI
	0.000398	7.96

The maximum potential exposure of this metabolite *via* ground water would be above 20% (infant, child) of the ADI. Considering other routes of uptake for flufenacet sulfonic acid:

- worst case value for consumer exposure (parent value) - 15% of ADI (Section B7)

Groundwater metabolite concentration and resulting % ADI including residues from consumer risk assessment

Scenario	TMDI (mg/kg bw/day)	% of ADI	Consumer risk assessment	Total (% ADI)
Infant	0.00179	35.8	15%	50.8
Child	0.00119	23.88	15%	38.8
Adult	0.000398	7.96	15%	23

Metabolites which have passed steps 1 to 3 and for which levels of estimated concentrations of metabolites in groundwater (as defined in Step 2) exceed 0.75 µg/L will require a refined assessment of their potential toxicological significance for consumers.
Estimation of an acceptable daily intake (ADI) FOE sulfonic acid (M02)

On the basis of the findings from toxicological and animal metabolism studies it is concluded that the bioavailability for FOE sulfonic acid (M02) is low compared to the parent substance.

FOE sulfonic acid has low oral bioavailability with absorption of about 10% in the rat. Together with a fast renal excretion this resulted in very low systemic exposure (Kroetlinger and Schmidt (2000), M-042251-01-1, see Monograph and baseline dossier KCA 5.8.1).

Comparing absorption of FOE sulfonic acid to the parent compound for which an absorption rate of 75-89% (low dose 1 mg/kg; m/f) of the applied rate was determined in the rat ADME study, the bioavailability of both metabolite is considerably less.
Therefore, the ADI of the parent compound is considered adequate for the assessment of consumer exposure to ~~FOE oxalate and~~ FOE sulfonic acid via drinking water and other sources.

Table 10.2.5-1 Toxicological endpoints for flufenacet

Active	**Endpoint**	**Value (mg/kg bw/day)**	**Study**	**Safety factor**	**Reference**

substance					
Flufenact	Acceptable Daily Intake (ADI)	0.005	2 year rat study (LOEL)	250	**Review Report (7469/VI/98-Final – 3rd July 2003)**
	Acute Reference Dose (ARfD)	0.017	90 day, 1 year dog study	100	

Risk assessment: Consumer exposure via ground-or drinking water:
The exposure of consumers to FOE sulfonic acid (M02) via drinking water and the resulting usage of the toxicological reference values (ADI, ARfD) is assessed based on the following worst case assumptions which lead to a very conservative approach:
- The ADI of 0.005 mg/kg bw/day and ARfD of 0.017 mg/kg bw/d for flufenacet are used as reference values.
Because the groundwater concentrations are compared to toxicological reference values expressed as flufenacet, also the concentration of the metabolites contained in ground-or drinking water must be expressed as parent equivalents.

Table 10.2.5-2 Conversion of gw concentration expressed as metabolite to parent equivalents

Metabolite	Estimated upper limit gw concentration expressed as metabolite [µg/L]	Estimated upper limit gw concentration expressed as parent equivalent [µg/L]
FOE sulfonic acid (M02)	14.045	19.07

Molar mass flufenacet 363.3 g/mol
Molar mass FOE 5043 sulfonic acid 275.3 g/mol

a) Calculation risk for infant:

Based on these considerations an intake of 0.75 liters of water/day the theoretical ingestion of flufenacet would be to 14.3 µg/L of flufenacet, coreresponding to 2.86 µg//kg bw/d taking into account a weight of 5 kg/person.

The following amounts for flufenacet metabolites by means of intake from drinking water and the corresponding ADI / ARfD usages are calculated:

Table 10.2.5-3 Upper limit intake of FOE sulfonic acid through drinking water

Metabolite	Intake [µg/kg bw/d] expressed as parent equivalent	**Usage of ADI [%]**	**Usage of ARfD [%]**
FOE sulfonic acid (M02)	2.86	57.2	16.8

a) Calculation risk for toodler:

Based on these considerations an intake of 1 liters of water/day the theoretical ingestion of flufenacet would be to 19.7 µg/L of flufenacet, coreresponding to 1.91 µg//kg bw/d taking into account a weight of 10 kg/person.

The following amounts for flufenacet metabolites by means of intake from drinking water and the corresponding ADI / ARfD usages are calculated:

Table 10.2.5-3 Upper limit intake of FOE sulfonic acid through drinking water

Metabolite	Intake [µg/kg bw/d] expressed as parent equivalent	**Usage of ADI [%]**	**Usage of ARfD [%]**
FOE sulfonic acid	1.91	38.2	11.2

(M02)			

a) Calculation risk for adult:

Based on these considerations an intake of 2 liters of water/day the theoretical ingestion of flufenacet would be to 38.14 µg/L of flufenacet, coreresponding to 0.64 µg//kg bw/d taking into account a weight of 60 kg/person.

The following amounts for flufenacet metabolites by means of intake from drinking water and the corresponding ADI / ARfD usages are calculated:

Table 10.2.5-3 Upper limit intake of FOE sulfonic acid through drinking water

Metabolite	Intake [µg/kg bw/d] expressed as parent equivalent	**Usage of ADI [%]**	**Usage of ARfD [%]**
FOE sulfonic acid (M02)	0.64	12.8	3.8

From the long-term and short-term exposure calculations above it can be concluded that possible intakes of FOE sulfonic acid (M02) by means of drinking water do not present a consumer health concern. The calculations are based on several worst case assumptions.

According to Guideline for Drinking-water Quality, WHO 2017, 20% of ADI value should be used only when the consumer exposure is unknown. In case of CHR/H/PENDIF 599.5 SC and metabolite M02 all of these data (ways of consumer exposure) were provided in dRR Part B7 – residue section - and this document. Therefore, the limitation of 20% of ADI for drinking water shouldn't be taken into account. The metabolite M02 is not included in plant residue definition for risk assessment and should not have influence on risk assessment for consumer. Although the worst case value for consumer exposure – parent value: 15% of ADI – can be used in assessment of consumer exposure for Metabolite M02. If the consumer exposure value from food (section B7 - 15% of ADI) and consumer exposure value from water calculated above are taken into account, total exposure for adult, toddler and infant is below 100% of ADI. According to information above, the metabolite M02 can no pose unacceptable risk to consumer.

Conclusion:
In summary the metabolite FOE sulfonic acid M02 is considered to be biologically, toxicologically and ecotoxicologically non relevant.

10.3 Relevance assessment of ASTCA

Summary:

The relevance of the groundwater metabolite ASTCA has already been assessed and the assessment agreed at EU level (RAR Florasulam-2013, Vol3 – B6) , and the relevance assessment is applicable as well for the GAP and groundwater scenarios considered in this dRR (i.e., the conclusions reached at Step 4 and 5 of the relevance assessment made at the EU-level are valid also with regard to the PEC_{gw} calculated for the GAP and groundwater scenarios considered in this dRR). ASTCA is not considered relevant according to the criteria laid down in the EC guidance document SANCO/221/2000 –rev.10.
A summary of the relevance assessment is given in Table 10.2-1 and the corresponding studies are listed in the corresponding sections.

Table 10.2-2: Summary of the relevance assessment for ASTCA according to the RAR Florasulam-2013, Vol3 – B6

	Assessment step		Result of assessment	
	STEP 1		Metabolite of no concern?	Yes
Quantification of groundwater contamination	STEP 2		Max PEC_{gw}	0.4330 µg/L
			Based on	FOCUS PEARL, Thivia,
Hazard assessment	STEP 3	Stage 1	Biological activity comparable to the parent?	no
		Stage 2	Genotoxic properties of metabolite	Non-genotoxic
		Stage 3	Toxic properties of metabolite;	Not toxic or very toxic (T or T+)
			Classification of parent	not currently classified as toxic or very toxic
			Classification of metabolite	not currently classified as toxic or very toxic
Consumer health risk assessment	STEP 4		Estimated consumer exposure via drinking water and other sources; threshold of concern approach	Acceptable <0.75 µg/L
	STEP 5		Refined risk assessment	Not required
			Predicted exposure (% of ADI)	Not required
			ADI based on	Not required

* N/A: not applicable

10.4 Relevance assessment of TSA

Summary:

The relevance of the groundwater metabolite TSA has already been assessed and the assessment agreed at EU level (RAR Florasulam-2013, Vol3 – B6) , and the relevance assessment is applicable as well for the GAP and groundwater scenarios considered in this dRR (i.e., the conclusions reached at Step 4 and 5 of the relevance assessment made at the EU-level are valid also with regard to the PEC_{gw} calculated for the GAP and groundwater scenarios considered in this dRR). TSA is not considered relevant according to the criteria laid down in the EC guidance document SANCO/221/2000 –rev.10.
A summary of the relevance assessment is given in Table 10.2-1 and the corresponding studies are listed in the corresponding sections.

Table 10.2-3: Summary of the relevance assessment for TSA according to the RAR Florasulam-2013, Vol3 – B6

	Assessment step		Result of assessment	
	STEP 1		Metabolite of no concern?	Yes
Quantification of groundwater contamination	STEP 2		Max PEC_{gw}	0.3364 µg/L
			Based on	FOCUS PEARL, Thivia,
Hazard assessment	STEP 3	Stage 1	Biological activity comparable to the parent?	no

		Stage 2	Genotoxic properties of metabolite	Non-genotoxic
		Stage 3	Toxic properties of metabolite;	Not toxic or very toxic (T or T+)
			Classification of parent	not currently classified as toxic or very toxic
			Classification of metabolite	not currently classified as toxic or very toxic
Consumer health risk assessment	STEP 4		Estimated consumer exposure via drinking water and other sources; threshold of concern approach	Acceptable <0.75 µg/L
	STEP 5		Refined risk assessment	Not required
			Predicted exposure (% of ADI)	Not required
			ADI based on	Not required

* N/A: not applicable

10.5 Relevance assessment of 5-OH-Florasulam

Summary:

The relevance of the groundwater metabolite 5-OH Florasulam has already been assessed and the assessment agreed at EU level (RAR Florasulam-2013, Vol3 – B6) , and the relevance assessment is applicable as well for the GAP and groundwater scenarios considered in this dRR (i.e., the conclusions reached at Step 4 and 5 of the relevance assessment made at the EU-level are valid also with regard to the PEC_{gw} calculated for the GAP and groundwater scenarios considered in this dRR). 5-OH Florasulam is not considered relevant according to the criteria laid down in the EC guidance document SANCO/221/2000 –rev.10.
A summary of the relevance assessment is given in Table 10.2-1 and the corresponding studies are listed in the corresponding sections.

Table 10.2-4: Summary of the relevance assessment for 5-OH Florasulam according to the RAR Florasulam-2013, Vol3 – B6

	Assessment step		**Result of assessment**	
	STEP 1		Metabolite of no concern?	Yes
Quantification of groundwater contamination	STEP 2		Max PEC_{gw}	0.202 µg/L
			Based on	FOCUS PELMO, Porto,
Hazard assessment	STEP 3	Stage 1	Biological activity comparable to the parent?	no

		Stage 2	Genotoxic properties of metabolite	Non-genotoxic
		Stage 3	Toxic properties of metabolite;	Not toxic or very toxic (T or T+)
			Classification of parent	not currently classified as toxic or very toxic
			Classification of metabolite	not currently classified as toxic or very toxic
Consumer health risk assessment	STEP 4		Estimated consumer exposure via drinking water and other sources; threshold of concern approach	Acceptable <0.75 µg/L
	STEP 5		Refined risk assessment	Not required
			Predicted exposure (% of ADI)	Not required
			ADI based on	Not required

Appendix 1 Lists of data considered in support of the evaluation

List of data submitted by the applicant and relied on

Data point	Author(s)	Year	**Title** **Company Report No.** **Source (where different from company)** **GLP or GEP status** **Published or not**	**Vertebrate study Y/N**	Owner
KCP 10	J. Antonik	2016	In vitro evaluation of Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid, Trifluoroacetic acid and Flufenacet methylsulfone genotoxicity using the micronucleus assay (MNA). Selvita S.A. Park Life Science, Poland Study code: K81/JA/01 GLP- yes Unpublished	N	Chemirol
KCP 10	J.Antonik	2016	In vitro Mammalian Cell Gene Mutation test (OECD 490) - genotoxicity determination of Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid and Trifluoroacetic acid by Mouse Lymphoma Assay Selvita S.A. Park Life Science, Poland Study code: K82/JA/01 GLP- yes Unpublished	N	Chemirol

List of data submitted or referred to by the applicant and relied on, but already evaluated at EU peer review

Cross reference to the section B6 of the dRR

Appendix 2 Additional information

Reference:	KCP 10.1/01
Report	In vitro evaluation of Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid, Trifluoroacetic acid and Flufenacet methylsulfone genotoxicity using the micronucleus assay (MNA) Justyna Antonik, 2016 Selvita S.A. Park Life Science, Poland Study code: K81/JA/01 GLP Unpublished
Guideline(s):	Organization for Economic Cooperation and Development (OECD) 487 and under GLP requirements
Deviations:	No
GLP:	Yes
Acceptability:	Yes
Duplication (if vertebrate study)	-

Materials and Methods:

Test System:

CHO-K1 cell line was cultivated according to the previously established SOP-01 in 25 cm2 or 75 cm2 tissue culture flasks at 37ºC in a humidified atmosphere containing 5% CO2 using Ham's F12 medium supplemented with 10% v/v h.i. FBS and antibiotics (Penicillin and Streptomycin). The doubling time of CHO-K1 determined at Selvita is approximately 18h. The cultures were tested regularly for the absence of mycoplasma infections.

Test Item:

Item	Provider	CAS	Batch no	MW [g/mol]	Mfg. date	Exp. date
Flufenacet oxalate	Key Organics	201668-31-7	EXP-15-EE1619	225.2	01.12.2015	12.2017
Flufenacet sulfonic acid	Key Organics	n/a	EXP-15-DFO394	275.3	01.12.2015	12.2017
Trifluoroethanesulfonic acid	Apollo Scientific	1827-97-0	A5457345	164.1	10.2015	10.2017

Method:

The *in vitro* Micronucleus Assay (MNA) is a mutagenic test system for the detection of chemicals that induce the formation of small membrane-bound DNA fragments (micronuclei - MN) in the cytoplasm of interphase cells. The MNA, used for regulatory purposes measures formation of chromosomal changes following DNA damage induced by the items under test, and is used to predict the genotoxic potential of pharmaceuticals, industrial chemicals, food additives and cosmetic ingredients. MN originate from chromosome fragments or whole chromosomes that are not included in the main daughter nuclei during nuclear division. They reflect chromosome damage and may thus provide a marker of genotoxicity and even early-stage carcinogenesis. The most commonly used method in mammalian cells is the cytokinesis-block micronucleus (CBMN) assay. In the CBMN assay, MN are scored after a single cell division using binucleated cultured cells (accumulated using cytochalasin B) to eliminate the confounding effect of altered cell division kinetics on the MN index.

Test procedure:

1. CHO-K1 cells were maintained in Ham's F12 medium supplemented with 10% v/v h.i. FBS , 1000 U/mL penicillin and 1000 U/mL streptomycin.

2. For experiments cells were plated at 50 000 cells/well into the wells of a 24-well plate, in a volume of 500 µL per well (-S9 incubation), 100 000 cells/well into the wells of a 12-well plate, in a volume of 1000 µL per well (+S9 incubation) and cultured overnight (18–22h) prior to the start of the assay.

3. The following day, the medium was removed and replaced with 500 µL (-S9 incubation) or 600 µL (+S9 incubation) per well of medium with test items and appropriate positive and negative controls. For details see Table 1.
4. For the short treatment (+/- S9), cells were treated with items for 3h, after which the medium was removed, the cells were washed once with warm medium, and fresh medium containing cytochalasin B (3 µg/mL) was added for 27h.
5. At the end of the incubation period, the medium was removed, the cells were washed once with warm PBS, than were detached by trypsinization, collected to 15- ml falcons in 3 mL medium and centrifuged for 8 minutes in 160 x g.
6. Medium was discarded and cells were washed with 2 mL of PBS. Cultures were centrifuged for 8 minutes in 160 x g.
7. Cells were treated with 1 mL of warm 75 mM KCl hypotonic solution for 20-30 seconds and then they were fixed by adding 2 mL of cold fixative (acetic acid:methanol in proportions 1:3 v/v).
8. Cultures were centrifuged for 8 minutes in 160 x g, then treated with 3 mL of fixative and centrifuged again.
9. The cells were incubated in fresh fixative for 30 minutes at room temperature, after which they were centrifuged for 8 minutes in 160 x g.
10. After last centrifugation, the supernatant was gently discarded, whilst cel suspension (approx. 150 µL fixative) was gently resuspended and a few drops of suspension was placed on a cold clean glass slide in humid chamber (45°C in water bath) and air dried.
11. Next day, the slides were stained by with 15% Giemsa stain for approx. 5 minutes, then washed twice in distilled water and air dried.

Results:
The formation of MN is a consequence of chromosomal breakage and/or spindle-fiber dysfunction induced by clastogens and/or aneuploidogens. The present study was performed in accordance with the OECD 487 and under GLP requirements.
In order to assess genotoxic potential CHO-K1 cells were exposed to test items (Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid, Trifluoroacetic acid and Flufenacet methylsulfone) and appropriate reference items in system with (+S9) and without (-S9 short and extended treatment) an exogenous metabolic activation.
Statistical analysis of the MN frequency and binucleate cells with MN was performer using the Chi-square test with Yates' correction. To examine the dose-response relationship in frequencies of the micronuclei Chi-square test for trend was performed.
None of tested concentration of Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid, Trifluoroacetic acid and Flufenacet methylsulfone exhibit a statistically significant increase in MN frequency compared with the concurrent negative
control ($P>0.05$, Tables IX-XIII, Figure I-VII). Chi-square test for trend revealed no dose-related increase in MN frequency ($P>0.05$). Results for positive reference items (mitomycin C and cyclophosphamide) demonstrated reproducibility and sensitivity of system.
In summary, the present research has demonstrated that items Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid, Trifluoroacetic acid and Flufenacet methylsulfone did not produce dose-dependent genetic toxicity in the CHO-K1 cells.

Reference:	KCP 10.1/02
Report	In vitro Mammalian Cell Gene Mutation test (OECD 490) - genotoxicity determination of Flufenacet oxalate, Flufenacet sulfonic acid, Trifluor ethanesulfonic acid and Trifluoroacetic acid by Mouse Lymphoma Assay Justyna Antonik, 2016 Selvita S.A. Park Life Science, Poland Study code: K82/JA/01 GLP Unpublished
Guideline(s):	Organization for Economic Cooperation and Development (OECD) 490 and under GLP requirements

Deviations: No

GLP: Yes

Acceptability: Yes

Duplication (if vertebrate study) -

Materials and Methods:

Test System:

The L5178Y TK+/− (clone 3.7.2C) cell line was purchased from American Type Culture Collection (ATCC) and maintained in log phase growth by serial subculturing. The cells were routinely cultured in RPMI 1640 supplemented with 10% (v/v) heat inactivated horse serum hereafter referred to as the medium growth (Medium10). To reduce the frequency of spontaneous TK−/− mutants, cell cultures were cleansed of the pre-existing TK−/− mutants by exposing them to the thymidine, hypoxanthine, methotrexate and glutamine (THMG) for approximately 24 hours to select against the TK−/− phenotype. The concentration of heat inactivated horse serum was reduced to 5% (v/v) prior to treatment with tested item.

The cloning medium (Medium 20) consisted of RPMI 1640 supplemented with 20% (v/v) heat inactivated horse serum. For selection, the cloning media were supplemented with 3 µg/mL 3-trifluorothymidine (TFT).

Test Item:

Samples of the test items were provided by the Study Sponsor (Table 1). Dimethyl sulfoxide (DMSO) was selected as a solvent for compounds: Flufenacet oxalate, Flufenacet sulfonic acid, and Trifluoroethanesulfonic acid, while water (H2O) was selected as a solvent for Trifluoroacetic acid.

Flufenacet oxalate and Flufenacet sulfonic acid were soluble at 2000 µg/mL concentration – the highest concentration recommended by OECD 490 for compounds with molecular mass above 200 g/M. However, it was noted that Flufenacet sulfonic acid at this concentration quickly polymerizes in DMSO, while after addition to culture medium it rapidly dissolutes. Trifluoroethanesulfonic acid was soluble at 10 mM concentration – the highest concentration recommended by OECD 490 for compounds with molecular mass below 200 g/M. Compound was soluble at all concentrations used. Trifluoroacetic acid was tested at concentrations up to 2 µL/mL – the highest concentration recommended by OECD 490 for liquid compounds. It was noted that Trifluoroacetic acid at 2 µL/mL concentration rapidly changes pH of the culture medium.

Name	Provider	CAS	Batch no	MW [g/mol]	Mfg. date	Exp. date
Flufenacet oxalate	Key Organics	201668-31-7	EXP-15-EE1619	225.2	01.12.2015	12.2017
Flufenacet sulfonic acid	Key Organics	n/a	EXP-15-DFO394	275.3	01.12.2015	12.2017
Trifluoroethanesulfonic acid	Apollo Scientific	1827-97-0	A5457345	164.1	10.2015	10.2017
Trifluoroacetic acid	FluoroChem	76-05-1	20120730	114.0	n/a	n/a

Control item

Methyl methanosulfonate (MMS) and Cyclophosphamide (Cp) were selected to be used in the assay as positive controls.

MMS was used in the absence of metabolic activation (−S9) and Cp in the presence of metabolic activation (+S9). MMS is a direct acting mutagen, while Cp is promutagen that requires biotransformation with the liver enzymes to elicit a mutagenic response. PBS without or with S9 treated cultures were used as vehicle (negative) controls for tested item.

Positive controls demonstrated effectiveness of the assay.

Method:

The Mouse Lymphoma Assay (MLA) is a short-term assay designed to detect forward gene mutations induced by mutagens at the heterozygous thymidine kinase (TK) locus. It is capable of quantifying genetic alterations. The system recommended by OECD 490 employed L5178Y TK+/− cells and the TK (thymidine kinase) locus. 5-Trifluorothymidine (TFT) is a toxic pyrimidine analogue that interferes with DNA metabolism causing cell death. However, if the functional copy of the TK gene is lost (TK−/−) through mutation, the TFT is not metabolized and is no longer toxic. The L5178Y TK+/− cells are

sensitive to the cytotoxic effects of the TFT. When L5178Y TK+/− cells are exposed on mutagenic and/or carcinogenic agents, TK+/− is mutated to the TK−/− genotype which is causing TFT resistance. The mutant cells when cloned in medium containing the selective agent TFT, proliferate and form colonies. The mouse lymphoma TK assay uses the thymidine kinase (TK) gene (reporter of mutation) d tects a broad spectrum of genetic damage, including point mutations, large scale chromosomal changes and recombination. That is why it is often recommended and widely used to determine the genotoxic potential of various chemicals. This is also the Gene Mutation Assay of choice at Selvita laboratory as a suitable short-term mutagenicity screening assay to predict chemical carcinogenicity. The studies were performed according to Standard Research Procedure SPB-19.

Exposure:

On day 1, L5178Y TK−/−-clean cells growing in logarithmic phase were treated in individual 50mL falcon tubes for 4 hours exposition and in T75 cm2 culture flasks for extended exposition. Each tube contained 8.5 mL of cell suspension (6×106 cells in total) in Medium 5. In the next step, 0.5 mL of S9 mixture or medium 5 was added. Then 100 µL solution of test items (20 µL for Trifluoroacetic acid), 100 µL of positive control or vehicle was added. Each tube was fulfilled to the 10 mL volume (short incubation). Each culture flask contained 19.8 mL of cell suspension (4×106 cells in total) in Medium 5. Then 200 µL solution of vehicle and test items (40 µL for Trifluoroacetic acid), 200 µL of positive control or vehicle was added. Each culture flask was fulfilled to the 20 mL volume. Following addition of the test item, the cell suspensions were gently mixed and placed in a CO2 incubator at 37°C for the exposure period. At the end of the exposure time, the cells were pelleted, washed with Medium A and collected by centrifugation, and then resuspended in 20 mL of Medium 10. Cultures were transferred to flasks for growth through the expression period and placed in the CO2 incubator (5% CO2, 37°C).

Cell suspension from each culture was used for counting (post-treatment) and for plating immediately after treatment to obtain Relative Viability (RV) and Relative Total Growth (RTG) values. Portion from the cell suspension was used to prepare 3-step dilution with non-selective (without TFT) Medium 20 to obtain concentration of 8 cells/mL. Using a multichannel pipette, 200 µL of cell suspension was dispensed to each well of two 96-well sterile flat-bottom plates for each tested dose and controls.

Results:

Mutagenic potential of Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid and Trifluoroacetic acid was evaluated trough Mouse Lymphoma Assay (MLA) in L5178Y cells. Tested items were analyzed in MLA, in the presence and absence of exogenous metabolic activation. Obtained results have shown that tested item did not exceed MF above a value termed as Global Evaluation Factor 126 ×10−6 in any of the tested doses both in the presence and absence of S9 exogenous activation system (Table 9-11).

Obtained results indicate that tested items (Flufenacet oxalate, Flufenacet sulfonic acid, Trifluoroethanesulfonic acid and Trifluoroacetic acid) or their metabolic derivatives were not positive in Mouse Lymphoma Assay under the protocol described and according to the acceptability criteria defined in OECD guideline 490 and SPB-19.

zRMS comments	zRMS has evaluated these studies in Section 6.