Table A6.6.2/02-1 Types and Frequencies of Chromosome Aberrations

Concentration (mg/ml)	Chromatid Type Breaks	Chromatid Type Gaps	Chromosome Type Breaks	Chromosome Type Gaps	Rearrangements	Aberrant Cells	% Aberrant Cells
Within the Presence of S9							
0.03A	----------Not Evaluated----------						
0.03B	----------Not Evaluated----------						
1.0A	1	3	0	0	0	1	2.0
1.0B	2	0	1	0	0	3	6.0
3.0A	0	0	1	0	0	1	2.0
3.0B	1	1	0	0	0	1	2.0
10A	3	0	1	1	0	3	6.0
10B	1	3	0	1	0	1	2.0
30A	----------Not Evaluated----------						
30B	----------Not Evaluated----------						
Controls:							
Vehicle:							
Culture Medium A	1	0	0	0	0	1	2.0
Culture Medium B	1	1	0	0	0	1	2.0
Positive:							
CP [a]	12	3	5	2	1	17	38.6
Within the Absence of S9							
0.03A	----------Not Evaluated----------						
0.03B	----------Not Evaluated----------						
0.10A	0	0	0	0	0	0	0
0.10B	2	1	1	0	0	3	6.0
0.30A	0	0	0	0	0	0	0
0.30B	1	1	1	0	0	2	4.0
1.0A	0	4	0	1	0	0	0
1.0B	2	3	1	2	0	3	6.0
3.0A	----------Not Evaluated----------						
3.0B	----------Not Evaluated----------						
Controls:							
Vehicle:							
Culture Medium A	1	0	0	0	0	1	2.0
Culture Medium B	2	1	0	1	0	2	4.0
Positive:							
TEM [b]	16	4	6	2	3	18	36.0

a CP = cyclophosphamide, only 44 cells were suitable for evaluation.

b TEM = triethylenemelamine

Section A6.6.3(1) Annex Point IIA, VI.6.3 IUCLID 5.5/04	Genotoxicity *in vitro* Mutagenic potential in the CHO/HGPRT forward mutation assay	
	1 REFERENCE	Official use only
1.1 Reference	[redacted] (1994b) [redacted] (glutaraldehyde, 50% aqueous solution): Mutagenic potential in the CHO/HGPRT forward mutation assay, [redacted], Unpublished, 8 April 1994	
1.2 Data protection	Yes	
1.2.1 Data owner	The Dow Chemical Company	
1.2.2 Companies with letter of access	[redacted]	
1.2.3 Criteria for data protection	Data on an existing active substance for first entry to Annex I authorisation	
	2 GUIDELINES AND QUALITY ASSURANCE	
2.1 Guideline study	Yes US EPA OPP 84-2a	
2.2 GLP	Yes	
2.3 Deviations	Yes Analyses for stability and homogeneity of the test (and control) substances in the dosing solutions were not conducted. The vehicle and positive control substances were not analyzed for chemical purity, stability or uniformity.	
	3 MATERIALS AND METHODS	
3.1 Test material	[redacted] (Glutaraldehyde 50% aqueous solution)	
3.1.1 Lot/Batch number	[redacted]	
3.1.2 Specification	Not reported	
3.1.2.1 Description	Transparent, colorless, non-viscous liquid	X
3.1.2.2 Purity	[redacted]	
3.1.2.3 Stability	Assumed to be stable under normal storage conditions.	X
3.2 Study Type	Chinese Hamster Ovary Cell / Hypoxanthine-Guanine-Phosphoribosyl Transferase (CHO/HGPRT) Forward Mutation Assay	
3.2.1 Organism/cell type	Chinese Hamster Ovary Cell	
3.2.2 Deficiencies / Proficiencies	See Section 2.3	
3.2.3 Metabolic activation system	Metabolic activation was accomplished using rat liver S9 homogenate, prepared from Aroclor 1254-induced rats purchased from a commercial supplier.	
3.2.4 Positive controls	**In the absence of metabolic activation (S9)** Ethylmethanesulfonate (EMS), Lot-88F-0531 **In the presence of metabolic activation (S9)**	

Section A6.6.3(1) Annex Point IIA, VI.6.3 IUCLID 5.5/04	Genotoxicity *in vitro* **Mutagenic potential in the CHO/HGPRT forward mutation assay**	
	Dimethylnitrosamine (DMN), Lot- 38F0882	
3.3 Administration / Exposure; Application of test substance		
3.3.1 Concentrations	***Cytotoxicity study*** ***In the absence & presence of metabolic activation (S9)*** 0, 0.0003, 0.001, 0.003, 0.01, 0.03, 0.10, 0.30, 1.0, 3.0 and 10.0 mg/ml equivalent to 0.0002, 0.0005, 0.002, 0.005, 0.02, 0.05, 0.15, 0.51, 1.5 and 5.1 mg/ml glutaraldehyde, corrected for percent active ingredient. ***Main Assay*** ***In the absence of metabolic activation (S9)*** *Test 1* 0.0003, 0.001, 0.003, 0.006, 0.01 and 0.03 mg/ml equivalent to 0.0002, 0.0005, 0.002, 0.003, 0.005 and 0.02 mg/ml glutaraldehyde, corrected for percent active ingredient. *Test 2* 0.0001, 0.0003, 0.001, 0.003 and 0.006 mg/ml equivalent to 0.00005, 0.0002, 0.0005, 0.002 and 0.003 mg/ml glutaraldehyde, corrected for percent active ingredient. ***In the presence of metabolic activation (S9)*** *Test 1* 0.001, 0.003, 0.01, 0.03, 0.05 and 0.1 mg/ml equivalent to 0.0005, 0.002, 0.005, 0.02, 0.02 and 0.05 mg/ml glutaraldehyde, corrected for percent active ingredient. *Test 2* 0.003, 0.005, 0.01, 0.02 and 0.03 mg/ml equivalent to 0.002, 0.002, 0.005, 0.01 and 0.02 mg/ml glutaraldehyde, corrected for percent active ingredient. ***In the presence of metabolic activation (S9)*** *Test 3* 0.003, 0.005 and 0.01 mg/mL equivalent to 0.0015, 0.0025 and 0.005 mg/mL glutaraldehyde, corrected for percent active ingredient.	
3.3.2 Way of application	***Cytotoxicity study*** Approximately 24 hours prior to the mutation test, CHO cells were plated and incubated at 37°C. Immediately prior to treatment the medium in each flask was replaced with serum-free medium. S9 mixture was added to the appropriate cultures and the cell were then treated duplicate cultures were treated for approximately 4 hours in the presence or absence of S9, rinsed, and fresh medium was added. The cells were incubated for 18-24 hours. ***Main Assay***	

Section A6.6.3(1) **Annex Point IIA, VI.6.3** **IUCLID 5.5/04**		**Genotoxicity *in vitro*** **Mutagenic potential in the CHO/HGPRT forward mutation assay**	
		Approximately 20-24 hours prior to the mutation test, CHO cells were plated and incubated at 37 °C. On the day of testing, duplicate cultures were treated for approximately 4 hours in the presence or absence of S9, rinsed, and fresh medium was added. The cells were allowed to recover for 18-24 hours before chemically-induced toxicity was determined. At 2-3 day intervals following treatment, cells from each culture were subcultured in fresh dishes and incubated. Eight days later, cells were dissociated with trypsin, counted and plated. Colonies were allowed to grow an additional 6-8 days, then the new colonies were fixed, stained, and counted using manual methods or a colony counter.	
3.3.3	Pre-incubation time	24 hour incubation in medium before replacement with serum-free medium.	
3.3.4	Other modifications	None	
3.4	**Examinations**		
3.4.1	Number of cells evaluated	Not applicable. Mutant colonies were counted on agar plates.	
		RESULTS AND DISCUSSION	
3.5	**Genotoxicity**		
3.5.1	without metabolic activation	[redacted] is not considered mutagenic to cultured CHO cells in the absence of S9.	
3.5.2	with metabolic activation	[redacted] is not considered mutagenic to cultured CHO cells in the presence of S9.	
3.6	**Cytotoxicity**	No significant cytoxicity was observed at concentrations of 0.0003mg/ml or less in the absence of S9 or at concentrations of 0.003mg/ml or less in the presence of S9.	
		4 APPLICANT'S SUMMARY AND CONCLUSION	
4.1	**Materials and methods**	Forward mutation assay was performed using Chinese Hamster ovary cells measuring mutation at the hypoxanthine-guanine phosphoribosyl transferase loci (CHO/HGPRT). In the absence of metabolic activation (S9) Ethylmethanesulfonate (EMS) was used as the positive control and in the presence of metabolic activation (S9) Dimethylnitrosamine (DMN) was used as the positive control. Culture medium was the vehicle control. Rat liver S9 homogenate was prepared from Aroclor 1254-induced rats, and purchased from a commercial supplier. Two independent repetitions of the complete assay were performed. A partial repeat of cultures with S9 was also performed. CHO cells were obtained from Dr. Hsie at Oak Ridge National Laboratory, and cultured in the testing facility. A preliminary cytotoxicity study was conducted to set dose levels appropriate for the mutagenicity assay. For the mutagenicity evaluation, approximately 20-24 hours prior to the mutation test, CHO cells were plated and incubated at 37 °C. On the day of testing, duplicate cultures were treated for approximately 4 hours, rinsed, and fresh medium was added. The cells were allowed to recover for 18-24 hours before chemically-induced toxicity was determined. At 2-3 day intervals following treatment, cells	

Section A6.6.3(1) Annex Point IIA, VI.6.3 IUCLID 5.5/04	Genotoxicity *in vitro* **Mutagenic potential in the CHO/HGPRT forward mutation assay**	
	from each culture were subcultured in fresh dishes and incubated. Eight days later, cells were dissociated with trypsin, counted and plated. Colonies were allowed to grow an additional 6-8 days, when the new colonies were fixed, stained, and counted using manual methods or a colony counter. Mutation data were analyzed by the methods of Irr and Snee after transformation according to Box and Coxx. The criteria for interpretation of the test results depend upon the level of statistical significance relative to the control, and the evidence of a dose-response effect. Positive readings were required to meet both criteria, as well as be reproducible and an increase 2-fold above the vehicle control. All statistical analyses were performed using statistical software from Dixon (1990). For all statistical tests, the probability value of <0.05 (one-tailed) was used as the critical level of significance.	
4.2 Results and discussion	***Cytotoxicity study*** No significant cytoxicity was observed at concentrations of 0.0003mg/ml or less in the absence of S9 or at concentrations of 0.003mg/ml or less in the presence of S9. ***Main Assay*** ***Tables A6.6.3/01-1, 2, 3*** In the absence of S9 a statistically significant ($p<0.01$) increase in mutation frequency was observed with 0.0001mg/ml [redacted]. This increase was approximately 2.5 fold with respect to the mean vehicle control value and was consistent between replicates. However, since no statistically significant reproducible increases in mutation frequencies were observed at any of the other concentrations tested, [redacted] is not considered mutagenic to cultured CHO cells in the absence of S9. In the presence of S9 a statistically significant ($p<0.05$) increase in mutation frequency was observed with 0.003mg/ml [redacted]. However, since no statistically significant reproducible concentration related increases in mutation frequencies were observed during 3 independent repetitions of the test, Ucarcide® Antimicrobial 250 is not considered mutagenic to cultured CHO cells in the presence of S9. A full compendium of genotoxicity studies can be found in the following article, provided with the dossier: Zeiger, E., Gollapudi, B., and Spencer, P. (2005) Genetic toxicity and carcinogenicity studies of glutaraldehyde- a review, Mutation Research, 589, 136-151, Published.	
4.3 Conclusion	The test material is negative in the HGPRT assay with Chinese hamster ovary cells.	
4.3.1 Reliability	1	
4.3.2 Deficiencies	No	

Section A6.6.3(1) **Annex Point IIA, VI.6.3** **IUCLID 5.5/04**	**Genotoxicity *in vitro*** **Mutagenic potential in the CHO/HGPRT forward mutation assay**	
	Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE	
Date	October 26th, 2010	
Materials and Methods	3.1.2.1 It was mentioned in the study report that the test substance is a transparent, colourless liquid. 3.1.2.3 Stability of the test substance under storage conditions was not tested.	
Results and discussion	4.2. Results and discussion. There were increased mutation frequencies in several individual tests, as shown in Tables 6.6.3/01-1, 6.6.3/01-2 and 6.6.3/01-3. Only some of these are statistically significant, and many were seen in only one of the two duplicate cultures.	
Conclusion	The result of the study is equivocal, and would need further confirmation for a clear conclusion.	
Reliability	2	
Acceptability	Acceptable.	
Remarks		
	COMMENTS FROM DOW BENELUX B.V.	
Date	3 June 2009; 27 January 2011	
Materials and Methods		
Results and discussion		
Conclusion	* The lack of a dose-response and replicate response values suggest no treatment-related effect. Statistical and non-statistical increases in mutation frequency were not consistent with administered concentrations. * *See Zieger* et al *publication for other negative CHO/HGPRT assays with and without S9.*	
Reliability	*Discuss if deviating from view of rapporteur member state*	
Acceptability	*Discuss if deviating from view of rapporteur member state*	
Remarks		

Table 6.6.3/01-1 Results of the Gene Mutation Assay in CHO Cells Treated with the Test Material Test #1

	With S9		Without S9	
Test 1				
Concentration (mg/ml)	*Plating Efficiency (% of Control)*	*Mutation Frequency* [a]	*Plating Efficiency (% of Control)*	*Mutation Frequency* [a]
0.0003A	---	---	85	4.1
0.0003B	---	---	104	7.5
0.001A	90	5.9	91	6.7
0.001B	142	1.9	86	17.3
0.003A	77	4.6	78	0
0.003B	141	2.5	91	0
0.006A	---	---	T	T
0.006B	---	---	T	T
0.01A	56	4.8	T	T
0.01B	77	18.6	T	T
0.03A	T	T	T	T
0.03B	T	T	T	T
0.05A	T	T	---	---
0.05B	T	T	---	---
0.10A	T	T	---	---
0.10B	T	T	---	---
Controls:				
Vehicle:				
Culture Medium A	109	1.6	93	10.3
Culture Medium B	91	0	107	0
Positive:				
EMS (200 ug/mL)	---	---	109	139.5
DMN (200 ug/mL)	107	53.2	---	---

a Mutants/ 10^6 clonable cells; total number of mutant colonies divided by viable fraction.

--- Not Conducted

T Cytotoxic

Table 6.6.3/01-2 Results of the Gene Mutation Assay in CHO Cells Treated with the Test Material Test #2

	With S9		Without S9	
Test 2 *Concentration (mg/ml)*	*Plating Efficiency (% of Control)*	*Mutation Frequency* [a]	*Plating Efficiency (% of Control)*	*Mutation Frequency* [a]
0.0001A	---	---	65	13.7*
0.0001B	---	---	86	15.5
0.0003A	---	---	90	17.7
0.0003B	---	---	99	4.5
0.001A	---	---	84	9.5
0.001B	---	---	114	6.2
0.003A	84	17.3	75	10.7
0.003B	87	15.7	80	24.4
0.005A	87	35.8	---	---
0.005B	85	8.6	---	---
0.006A	---	---	T	T
0.006B	---	---	T	T
0.01A	45	71.3	---	---
0.01B	59	8.8	---	---
0.02A	T	T	---	---
0.02B	T	T	---	---
0.03A	T	T	---	---
0.03B	T	T	---	---
Controls:				
Vehicle:				
Culture Medium A	98	5.3	104	5.1
Culture Medium B	102	11.3	96	6.5
Positive:				
EMS (200 ug/mL)	---	---	99	206.3
DMN (200 ug/mL)	107	41.2	---	---

a Mutants/ 10^6 clonable cells; total number of mutant colonies divided by viable fraction.

--- Not Conducted

* Significantly different from control group ($p < 0.01$)

T Cytotoxic

Table 6.6.3/01-3 Results of the Gene Mutation Assay in CHO Cells Treated with the Test Material Test #3

	With S9		Without S9	
Test 3				
Concentration (mg/ml)	*Plating Efficiency (% of Control)*	*Mutation Frequency* [a]	*Plating Efficiency (% of Control)*	*Mutation Frequency* [a]
0.003A	67	12.3*	---	---
0.003B	91	5.4	---	---
0.005A	65	2.5	---	---
0.005B	78	2.1	---	---
0.01A	75	0	---	---
0.01B	85	0	---	---
Controls:				
Vehicle:				
Culture Medium A	96	0.9	---	---
Culture Medium B	104	0	---	---
Positive:				
DMN (200 ug/mL)	79	61.1	---	---

a Mutants/ 10^6 clonable cells; total number of mutant colonies divided by viable fraction.

* Significantly different from control group ($p < 0.05$)

--- Not Conducted

[Cytotoxic

Section A6.6.4(1) **A6.6.4 Genotoxicity *in vivo***

Annex Point IIA, VI.6.4

***In Vivo* Mouse Micronucleus Assay**

IUCLID 5.6/01

			Official use only
		1 REFERENCE	
1.1	**Reference**	[redacted] (1993b), [redacted] (glutaraldehyde, 50% aqueous solution): *In-vivo* peripheral blood micronucleus test with [redacted] mice, [redacted]. Unpublished, 26 February 1993	
1.2	**Data protection**	Yes	
1.2.1	Data owner	The Dow Chemical Company	
1.2.2			
1.2.3	Criteria for data protection	Data submitted to the MS after 13 May 2000 on existing active substance for the purpose of its entry into Annex I	
		2 GUIDELINES AND QUALITY ASSURANCE	
2.1	**Guideline study**	Yes, OECD 474	
2.2	**GLP**	Yes	
2.3	**Deviations**	Yes The first sampling point was at 30h rather than the recommended 36h. This is not considered to have affected the results of the study. The site of blood sampling was not reported. The study director had no knowledge of the procedures used for the chemical analyses for interfering contaminants in the water conducted by the supplier, [redacted] or procedures used for diet analyses by [redacted] Analyses for stability, homogeneity, and concentration verification of the test (and control) substances in the dosing solutions were not conducted. The vehicle and positive control substances were not analyzed for chemical purity, stability, or uniformity. They exhibited the appropriate biological activities in this test system.	
		3 MATERIALS AND METHODS	
3.1	**Test material**	[redacted] (Glutaraldehyde 50% aqueous solution)	
3.1.1	Lot/Batch number	[redacted]	
3.1.2	Specification	As given in Section 2	
3.1.2.1	Description	Clear liquid	
3.1.2.2	Purity	[redacted]	
3.1.2.3	Stability	Reported to be stable under typical storage conditions	
3.1.2.4	Maximum tolerable dose	2000 mg/kg was reported to have exceeded the maximum tolerated dose based on a previously conducted acute peroral toxicity study in rats. The LD_{50} in rats was reported to be 246 in males, 154 in females and 317 mg/kg combined. Clinical signs of toxicity included sluggishness,	X

Section A6.6.4(1) **A6.6.4 Genotoxicity *in vivo***

Annex Point IIA, VI.6.4

***In Vivo* Mouse Micronucleus Assay**

IUCLID 5.6/01

		lacrimation, piloerection, diarrhoea, and a trace amount of blood in the urine of 2 animals. Red, perinasal soiling and perineal soiling was also noted. Deaths occurred at 1-2 days post-dosing	
3.2	**Test Animals**	*Non-entry field*	
3.2.1	Species	Mice	
3.2.2	Strain	[redacted]	
3.2.3	Source	[redacted]	
3.2.4	Sex	Male and Females	
3.2.5	Age/weight at study initiation	5 weeks old Male weight range 23.5 to 27.5 grams Female weight range 17.9 to 20.9 grams	
3.2.6	Number of animals per group	5 animals/sex/dose/sampling time	X
3.2.7	Control animals	Solvent/negative control (water) Positive control (triethylenemelamine) *via* intraperitoneal injection	
3.3	**Administration/ Exposure**	Peroral Intubation	
3.3.1	Number of applications	Single treatment	
3.3.2	Interval between applications	Not Applicable	
3.3.3	Postexposure period	Approximately 30, 48 and 72 hours after treatment.	
		Oral	
3.3.4	Type	Peroral Intubation	
3.3.5	Concentration	80, 160 and 250 mg [redacted]/kg equivalent to 40, 80, 125 mg a.i./kg	
3.3.6	Vehicle	Distilled Water	
3.3.7	Concentration in vehicle	Various, depending on the targeted dose level, to maintain a dose level of 10mL/kg body weight.	
3.3.8	Total volume applied	10mL/kg body weight	
3.3.9	Controls	Yes Solvent/negative control (water) Positive control (triethylenemelamine) *via* intraperitoneal injection	
3.3.10	Substance used as Positive Control	Triethylenemelamine	
3.3.11	Controls	Yes Solvent/negative control (water) Positive control (triethylenemelamine) via intraperitoneal injection	
3.4	**Examinations**	*Non-entry field*	
3.4.1	Clinical signs	Immediately post dose administration and twice daily thereafter.	
3.4.2	Tissue	Peripheral Blood Number of 5 per sex, per treatment	

		animals:
		Number of cells: 1000 polychromatic erythrocytes per animal
		Time points: Approximately 30, 48 and 72 hours after treatment.
		Type of cells: Erythrocytes in peripheral blood
		Parameters: Polychromatic/normochromatic erythrocytes ratio % micronucleated polychromatic erythrocytes
3.5	**Further remarks**	None

4 RESULTS AND DISCUSSION

4.1	**Clinical signs**	There were no clinical signs of toxicity in any female at any dose level. There were 4 deaths in males at the three dose levels; three were attributed to potential dosing errors and one possibly to treatment. Clinical signs in males were limited to unkempt appearance at 160 or 250 mg/kg.
4.2	**Haematology / Tissue examination**	**Table A6.6.4/01-1**
4.3	**Genotoxicity**	Negative
4.4	**Other**	None

5 APPLICANT'S SUMMARY AND CONCLUSION

5.1	**Materials and methods**	Thirty-five male and 35 female [redacted] mice were obtained from a commercial supplier, and were 4 weeks old at arrival to the testing facility. Animals were evaluated for health status, and uniquely identified by ear tags. Animals were group housed in shoebox caging in rooms designed to maintain adequate environmental conditions for the species. Tap water from a municipal source and commercial diet were provided ad libitum. Animals were acclimated to the laboratory for one week prior to testing, and weights were taken. Animals considered unfit for testing were excluded prior to study start. Animals were randomized to groups of 15 per sex per dose for the vehicle and positive controls, and the low and mid doses. Eight per sex were allocated to the high dose group. Distilled water was used as the vehicle control. The positive control was triethylenemelamine. Animals were dosed by peroral intubation in a single dose of 80, 160, or 250 mg/kg [redacted] (40, 80, 125 mg a.i./kg). The vehicle controls were given a single dose of water by the same route, and the positive control TEM was given as a single IP injection of 0.3 mg/kg. Peripheral blood was sampled at 30, 48, and 72 hours post-dosing for the [redacted] and negative controls, and at 30 hours for the positive control animals. One or two slides were prepared per animal at each sampling time. Micronuclei in peripheral blood polychromatophilic erythrocytes were stained. Slides were coded and read without knowledge of group assignments. The PCE:NCE ratio for 1000 cells for each animal was calculated to provide an estimate of

X

Section A6.6.4(1)	A6.6.4 Genotoxicity *in vivo*	
Annex Point IIA, VI.6.4	*In Vivo* Mouse Micronucleus Assay	
IUCLID 5.6/01		
	cytotoxicity. A minimum of 1000 PCE per animal was scored for the presence of micronuclei unless the toxicity of the test substance prevented such an evaluation. A maximum of 5 animals per sex per dose was evaluated. Data were analyzed using the Mann-Whitney U-test. All analyses were performed by computer. For all tests, the probability value of <0.05 (one-tailed) was used as the critical level of significance.	
5.2 Results and discussion	**Table A6.6.4/01-1** There were no clinical signs of toxicity in any female at any dose level. There were 4 deaths in males at the three dose levels; three were attributed to potential dosing errors and one possibly to treatment. Clinical signs in males were limited to unkempt appearance at 160 or 250 mg/kg. The pharmacokinetics of glutaraldehyde in the blood following oral gavage (IIA6.2) demonstrated glutaraldehyde was rapidly absorbed from the GI tract, reaching peak blood concentrations by 10-15 min. post-dosing. These data support the systemic bioavailability of glutaraldehyde to reach the target tissue, bone marrow in the current study.	X
5.3 Conclusion	[redacted] (Glutaraldehyde, 50% aqueous solution) did not produce statistically-significant changes in the PCE/NCE ratios in mice of either sex at any dose level or sampling time.	
5.3.1 Reliability	1	
5.3.2 Deficiencies	Only 1000 cells were evaluated, instead of 2000 per the current testing guidelines. Not all dose groups contained 5 animals as required by current testing guidelines.	

Evaluation by Competent Authorities

EVALUATION BY RAPPORTEUR MEMBER STATE

Date October 27th, 2010

Materials and Methods

2.3 One to a few drops of blood were collected by nicking the tail with a scalpel.

3.1.2.4 According to the study report, acute peroral LD_{50} in mice for the combined sexes was calculated to be 317 mg/kg [redacted]. The doses for the micronucleus assay were selected as 25, 50 and 80 % of this 3 day acute peroral LD_{50}, giving the concentrations of 80, 160 and 250 mg/kg. Maximum tolerable dose was reported according to the acute toxicity data on rat, which may not reflect the toxicity in mouse.

3.2.6. Eight mice/sex were treated at the high dose.

5.1 Animals were randomized to groups of 5/sex/dose for the vehicle and positive controls, and the low and mid doses.

Results and discussion

5.2 Results and discussion.

It is correct that glutaraldehyde absorbs rapidly after oral administration, but it mostly absorbs as molecules other than glutaraldehyde (or is rapidly converted

Section A6.6.4(1)	**A6.6.4 Genotoxicity *in vivo***
Annex Point IIA, VI.6.4	***In Vivo* Mouse Micronucleus Assay**
IUCLID 5.6/01	
	after absorption). At the time of C_{max} the rat blood contained approximately 0.16 % and 1.6 % of the total dose as glutaraldehyde following low and high dose, respectively (Dow A6.02/01, Dow A6.02/02). Three of the deaths occurred shortly after treatment and were most probably due to the dosing error. One male in the 80 mg/kg group was found dead on day 4. Necropsy examination of that animal revealed a tear in the esophagus. Other clinical signs of toxicity were limited to unkempt appearance in male mice of the mid and high dose groups. There were no significant changes in the PCE/NCE ratios in mice of either sex at any sampling time. In the light of very mild signs of toxicity and no changes in the PCE/NCE ratios, the dose levels chosen for the study might have been too low, or the test substance might not have reached the bone marrow.
Conclusion	Under the test conditions, GA was not genotoxic in the *in vivo* mouse micronucleus assay. Nevertheless, the validity of the result can be questioned because: • There were very mild signs of toxicity and therefore the doses selected based on the results from another species might have been too low. • It is quite possible that the test substance has not reached the bone marrow.
Reliability	2
Acceptability	The study is acceptable as such, but inconclusive.
Remarks	The test was performed according to the OECD guideline valid at the time. This guideline was replaced by a new one in 1997, requiring e.g. at least 2000 cells to be scored. Please note that the results in the Table below have not been checked in detail by the RMS.
	COMMENTS FROM DOW BENELUX B.V.
Date	3 June 2009; 27 January 2011
Materials and Methods	
Results and discussion	
Conclusion	Although this study deviates from the current guideline, it was conducted to the recommendations of the guideline at that time and the available toxicokinetic and ADME data support that Glutaraldehyde does reach the bone marrow. **RMS response**: It is correct that glutaraldehyde absorbs rapidly after oral administration, but it mostly absorbs as molecules other than glutaraldehyde, or is rapidly converted after absorption (see first comment to 5.2 Results and discussion).
Reliability	
Acceptability	* Significant toxicokinetic and ADME data on glutaraldehyde indicates oral bioavailability and that it can safely be assumed to have reached the bone marrow (please see dossier section A6.2.1-4). Dose levels tested (80% of LD50) would probably exceed a MTD for current testing parameters and it is likely that more specific clinical signs (vs. "unkempt appearance" used in the study) would

Section A6.6.4(1) **A6.6.4 Genotoxicity *in vivo***

Annex Point IIA, VI.6.4

***In Vivo* Mouse Micronucleus Assay**

IUCLID 5.6/01

indicate significant clinical toxicity. Alteration of PCE/NCE ratio does not necessarily reflect systemic toxicity, depending on the mode of action for the chemical.

RMS response: Glutaraldehyde genotoxicity testing would require glutaraldehyde to reach the target organ. The RMS does not believe that sufficient glutaraldehyde exposure at the target organ has occurred - please see the previous RMS comment above. The RMS does nevertheless not request further testing as it appears that such testing would not be relevant taking into account the possible exposure routes.

*Although not all dose groups had 5 animals, the number of time points that were analyzed (3) exceeds current requirements and the additional analysis points supplement these data. Furthermore, 5 animals were used in the highest dose tested (250 mg/kg/day) and no trend or dose-related increase in micronuclei were observed.

* Previous guidelines [OECD 474, Adopted 26 May, 1983] allowed for justification of fewer number of animals/group, *e.g.*, at the lower dose levels and also stated that 1000 PCE should be counted versus current requirement of 2000.

Remarks

Table A6.6.4/01-1 Results for Micronucleus Test *In Vivo*

Dose Group	Sex	N[a]	30 hours			48 hours			72 hours		
			Mean MN-PCE/1000 PCE		% PCE[b]	Mean MN-PCE/1000 PCE		% PCE[b]	Mean MN-PCE/1000 PCE		% PCE[b]
			Mean	S.D.	Mean	Mean	S.D.	Mean	Mean	S.D.	Mean
0[c] mg/kg	Male	5	2.2	1.30	---	2.6	2.07	---	1.8	0.84	---
	Female	5	1.6	0.89	---	3.6	2.61	---	2.8	1.48	---
80 mg/kg	Male	4	1.5	1.91	100.9	1.3	0.50	96.8	2.3	2.63	91.6
	Female	5	1.6	1.14	101.8	3.2	2.17	98.7	2.6	1.34	99.4
160 mg/kg	Male	4	3.3	0.50	103.1	2.8	2.75	130.6	2.5	1.29	146.0
	Female	5	1.8	0.84	122.7	3.0	1.00	108.6	1.6	1.34	100.6
250 mg/kg	Male	5	1.8	1.64	93.0	1.0	1.22	93.2	1.8	1.64	85.5
	Female	5	1.6	1,14	81.6	4.4	0.55	95.4	2.8	2.17	87.9
0.3 mg/kg TEM[d]	Male	5	17.2*	4.38	41.2	**	**	**	**	**	**
	Female	5	8.8*	3.19	42.3	**	**	**	**	**	**

a N is the number of animals per dose group at the time of scheduled sacrifice. 1000 PCE were examined/animal for MN incidence, and expressed as MN per 1000 PCE (% MNPCE)

b % PCE = % PCE of the control values calculated and reported separately.

c Mice were dosed with the vehicle (distilled water)

d TEM = triethylenemelamine

* Indicates statistically significant results, $p < 0.05$.

*** Not evaluated

Section A6.6.4(2) **Genotoxicity *in vivo***

Annex Point IIA6.6.4 Bone Marrow Chromosomal Aberration Assay in Rats

IUCLID 5.6/04

Official use only

1 REFERENCE

1.1	**Reference**	[redacted] (1993c), [redacted] (Glutaraldehyde, 50% Aqueous solution): Bone Marrow Chromosomal Aberration Assay in Rats; [redacted], 27 May 1993.
1.2	**Data protection**	Yes
1.2.1	Data owner	The Dow Chemical Company
1.2.2		
1.2.3	Criteria for data protection	Data submitted to the MS after 13 May 2000 on existing active substance for the purpose of its entry into Annex I

2 GUIDELINES AND QUALITY ASSURANCE

2.1	**Guideline study**	Yes, Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) Section 10 (d) (1) (A), (B) or (C), comparable to OECD Guideline 475.
2.2	**GLP**	Yes
2.3	**Deviations**	Yes • The mitotic index was not measured in at least 1000 cells per animal. • At least 100 cells were not analysed for each animal. These deviations were not considered to have impacted on the results of the study.

3 MATERIALS AND METHODS

3.1	**Test material**	[redacted] (Glutaraldehyde, 50% aqueous solution)
3.1.1	Lot/Batch number	[redacted]
3.1.2	Specification	As given in Section 2
3.1.2.1	Description	Transparent colourless non-viscous liquid
3.1.2.2	Purity	[redacted]
3.1.2.3	Stability	Stability not confirmed
3.1.2.4	Maximum tolerable dose	Doses for this study based on a peroral LD50 study [redacted] giving LD50 for males of 246 mg/kg bw and for females 154mg/kgbw.
3.2	**Test Animals**	*Non-entry field*
3.2.1	Species	Rat
3.2.2	Strain	[redacted]
3.2.3	Source	[redacted]
3.2.4	Sex	Male and female
3.2.5	Age/weight at study initiation	5 weeks; 176-210g (males) and 126-154g (females)
3.2.6	Number of animals	15 male and 15 female per dose, 5 male and 5 female in positive control,

Section A6.6.4(2) — **Genotoxicity *in vivo***
Annex Point IIA6.6.4 — Bone Marrow Chromosomal Aberration Assay in Rats
IUCLID 5.6/04

	per group	5 male and 5 female at each sampling time (12, 24, 48hours post dosing)
3.2.7	Control animals	Yes, negative and positive controls.
3.3	**Administration/ Exposure**	Oral
3.3.1	Number of applications	1
3.3.2	Interval between applications	Not applicable
3.3.3	Postexposure period	12, 24, 48 h after treatment
		Oral
3.3.4	Type	Gavage
3.3.5	Concentration	Males 25, 60 and 120 mg/kg bw Females 15, 40 and 80mg/kg bw
3.3.6	Vehicle	Water
3.3.7	Concentration in vehicle	0.25, 0.60 and 1.2%w/v [redacted] for males and 0.15, 0.40 and 0.80%w/v [redacted] in females equivalent to 2.5, 6.0 and 12.0mg/mL in males and 1.5, 4.0 and 8.0mg/mL in females. This is equivalent to 1.25, 3.0 and 6.0 mg a.i./mL for males and 0.75, 2.0 and 4.0mg a.i./mL.
3.3.8	Total volume applied	10mL/kg bw based on body weights on the day prior to treatment.
3.3.9	Controls	Vehicle for negative controls and cyclophosphamide monohydrate for positive controls.
3.4	**Examinations**	*Non-entry field*
3.4.1	Clinical signs	Yes
3.4.2	Tissue	Bone marrow
		Number of animals: — All animals
		Number of cells: — 500/animal where possible for assessment of mitotic index
		Time points: — 12, 24, 48 h after treatment
		Type of cells — erythrocytes in bone marrow
		Parameters: — numbers and types of structural aberrations
3.5	**Further remarks**	None
		4 RESULTS AND DISCUSSION
4.1	**Clinical signs**	No effects
4.2	**Haematology / Tissue examination**	Not determined
4.3	**Genotoxicity**	No

Section A6.6.4(2) **Genotoxicity *in vivo***

Annex Point IIA6.6.4 Bone Marrow Chromosomal Aberration Assay in Rats

IUCLID 5.6/04

4.4	**Other**	None	
		5 APPLICANT'S SUMMARY AND CONCLUSION	
5.1	**Materials and methods**	Fifteen male and 15 female rats were dosed orally at 25, 60 or 120 mg/kg [redacted] (males) or 15, 40 or 80 mg/kg [redacted] (females). A control group of 15 males and 15 females were treated with the vehicle (water). Five males and 5 females were treated intraperitoneally with the positive control substance, 30 mg/kg cyclophosphamide monohydrate. Bone marrow was collected 12, 24 and 48h after treatment and at 24h only for the positive control group. Bone marrow was collected and fixed prior to being placed on slides and stained. The slides were then evaluated for incidence and type of chromosomal damage.	
5.2	**Results and discussion**	[redacted] (Glutaraldehyde, 50% aqueous solution) did not produce significant or dose related increases in the incidence of chromosomal aberrations among male or female Sprague Dawley rats assessed at 12, 24, and 48 hours after treatment with [redacted] as a single oral dose. The pharmacokinetics of glutaraldehyde in the blood following oral gavage (IIA6.2) demonstrated glutaraldehyde was rapidly absorbed from the GI tract, reaching peak blood concentrations by 10-15 min. post-dosing. These data support the systemic bioavailability of glutaraldehyde to reach the target tissue, bone marrow in the current study.	X
5.3	**Conclusion**	[redacted] (Glutaraldehyde 50% aqueous solution) is not considered clastogenic in [redacted] rats under the conditions of this *in vivo* study.	
5.3.1	Reliability	1	
5.3.2	Deficiencies	No	

Evaluation by Competent Authorities
Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
EVALUATION BY RAPPORTEUR MEMBER STATE
Date October 27th, 2010

Section A6.6.4(2) Genotoxicity *in vivo*

Annex Point IIA6.6.4

Bone Marrow Chromosomal Aberration Assay in Rats

IUCLID 5.6/04

Materials and Methods	Doses selected for the chromosome aberration test in males were 25, 60 and 120 mg/kg bw. Dose selection for this study was based on an acute peroral toxicity study where 1/5 males died at 200 mg/kg bw. No gross lesions were detected in other animals at this dose level. There were no deaths or gross lesions in males at 100 mg/kg bw. Doses selected for the chromosome aberration test in females were 15, 40 and 80 mg/kg bw. 2/5 female rats died at 141 mg/kg bw in an acute peroral study. No gross lesions were detected in other animals at this dose level. There were no deaths or gross lesions in females at 100 mg/kg bw. The sampling intervals are according to the OECD guideline valid at the time of the study. However, it is now known that the times are not appropriate: for rodents, the first sampling interval is 1.5 normal cell cycle length (the latter being normally 12-18 h) following treatment. The next sampling time is recommended at 24 h after the first (acceptable) sampling time. According to the current OECD Guideline 475, mitotic index should be determined as a measure of cytotoxicity in at least 1000 cells per animal (500 were scored). At least 100 cells for each animal should be analysed for chromosomal aberrations (50 were evaluated).
Results and discussion	5.2 Results and discussion. The toxicokinetics studies indicate that glutaraldehyde is bioavailable but is either absorbed as molecules other than glutaraldehyde, or rapidly converted to other molecules. Therefore glutaraldehyde concentration in the bone marrow was low. One male treated with 120 mg/kg bw was found dead on the morning of Day 2. The cause of the death was not investigated. There were no other mortalities or noteworthy clinical signs observed during the study. There were no clear or dose-related signs of cytotoxicity (decrease in a mitotic index) in males or females. In the absence of clinical signs and bone marrow cytotoxicity, it is unclear whether the test substance has reached the bone marrow.
Conclusion	Under the test conditions, GA did not cause significant or dose related increases in the incidence of chromosomal aberrations. In the absence of clinical signs and bone marrow cytotoxicity, it is unclear whether the test substance has reached the bone marrow.
Reliability	2
Acceptability	The study is acceptable as such, but inconclusive.
Remarks	

Section A6.6.4(2) — **Genotoxicity *in vivo***

Annex Point IIA6.6.4 — Bone Marrow Chromosomal Aberration Assay in Rats

IUCLID 5.6/04

	COMMENTS FROM DOW BENELUX B.V.
Date	3 June 2009; 27 January 2011
Materials and Methods	OECD 475, (Adopted 4 April 1984) states under Performance of the test, "*Animals are treated with the test substance once at the selected dose(s). Samples are taken at three times after treatment. For rodents the central sampling interval is 24 hours. Since cell cycle kinetics can in influenced by the test substance, one earlier and one later sampling interval adequately spaced within the range of 6 to 48 hours are applied.*" In this assay, the sampling times and chromosomal aberrations were evaluated at 12, 24, and 48 hours. **RMS response**: Agreed. This is not a deviation from the guidelines, but does affect the scientific reliability. When this test was conducted, the OECD guideline at that time [OECD 475, Adopted 4 April 1984] stated, "The number of cells to be analyzed per animal should be based upon the number of animals used, the negative control frequency, the predetermined sensitivity and the power chosen for the test." The test protocol stated that when possible, 500 cells/animal will be scored to determine a mitotic index and when possible, 50 metaphase cells/animals will be evaluated for incidence and type of chromosome damage. Although these values are less than recommended in the current guideline, they were fully acceptable when evaluated and performed by the guideline at that time. **RMS response**: Agreed. This is not a deviation from the guidelines, but does affect the scientific reliability.
Results and discussion	
Conclusion	The study in question fully met the guidelines that were in effect at the time the study was conducted and should be fully accepted.
Reliability	
Acceptability	As previously mentioned for the *in vivo* micronucleus assay above, there is sufficient TK and ADME data for glutaraldehyde which indicates oral bioavailability and uptake. From these data, we conclude that the test material does reach the target tissue. **RMS response**: The studies indicate that glutaraldehyde is bioavailable but is either absorbed as molecules other than glutaraldehyde, or rapidly converted to other molecules. Therefore glutaraldehyde concentration in the bone marrow was low, and the test concerns metabolites of glutaraldehyde.
Remarks	

Section A6.6.4(2) **Genotoxicity *in vivo***

Annex Point IIA6.6.4 Bone Marrow Chromosomal Aberration Assay in Rats

IUCLID 5.6/04

Table A6_6_4-1. **Table for Bone Marrow Test *In Vivo* (modify if necessary)**

State mean ± standard deviation state individual numbers for critical findings		control group			low dose			mid dose			high dose		
Number of cells evaluated		500[a]			500[a]			500[a]			500[a]		
Sampling time (h)		12	24	48	12	24	48	12	24	48	12	24	48
Chromatid aberrations	**Gaps**	0	1	0	0	2	2	0	3	1	0	0	3
	Breaks	5	9	1	4	8	7	1	15	5	5	9	8
	Interchanges	0	0	0	0	0	0	0	0	0	0	0	0
Mitotic index (%)	**Males**	5.1	4.9	7.6	6.9	4.5	5.1	5.6	4.1	5.4	6.2	6.0	3.5
	Females	2.2	3.8	2.4	2.6	1.3	3.9	3.6	1.2	2.3	2.8	1.4	4.0

a = 250 cells from male rats and 250 cells from female rats i.e. 50 cells per animal (except 24h high dose males 200 cells examined; 24h low dose females 201 cells examined and 48h high dose females 200 cells examined)

The positive control group showed 130 breaks at 24h

Section A6.8.1(1) Annex Point IIA, VI.6.8.1 IUCLID 5.8.2/01	A6.8.1 Teratogenicity Study (Rabbit) **Study of the prenatal toxicity of glutaraldehyde in rabbits after oral administration (gavage)**	
	1 REFERENCE (A6.8.1/01)	**Official use only**
1.1 Reference	[redacted] (1991a) Study of the prenatal toxicity of glutaraldehyde in rabbits after oral administration (gavage), [redacted], Unpublished, 31 January 1991	
1.2 Data protection	Yes	
1.2.1 Data owner	[redacted] The Dow Chemical Company (Dow)	
1.2.2 Companies with letter of access	Dow [redacted]	
1.2.3 Criteria for data protection	Data submitted to the MS after 13 May 2000 on an existing a.s. for the purpose of its entry to Annex I	
	2 GUIDELINES AND QUALITY ASSURANCE	
2.1 Guideline study	Yes OECD 414	
2.2 GLP	Yes	
2.3 Deviations	No	
	3 MATERIALS AND METHODS	
3.1 Test material	Glutaraldehyde, 50%	
3.1.1 Lot/Batch number	[redacted]	
3.1.2 Specification	Not reported	
3.1.2.1 Description	Watery solution	
3.1.2.2 Purity	[redacted]	
3.1.2.3 Stability	Assumed to be stable under normal storage conditions	
3.2 Test Animals		
3.2.1 Species	Rabbit	
3.2.2 Strain	[redacted]	
3.2.3 Source	[redacted]	
3.2.4 Sex	Female	
3.2.5 Age/weight at study initiation	26-27 weeks of age Approximately 2596 grams	
3.2.6 Number of animals per group	15	

Section A6.8.1(1) Annex Point IIA, VI.6.8.1 IUCLID 5.8.2/01		A6.8.1 Teratogenicity Study (Rabbit) **Study of the prenatal toxicity of glutaraldehyde in rabbits after oral administration (gavage)**			
3.2.7	Control animals	Yes, concurrent vehicle			
3.2.8	Mating period	Artificial insemination following an injection of 0.2 mL of a synthetic hormone to release LH and FSH from the anterior pituitary lobe. Rabbits were inseminated with pooled ejaculate samples from male rabbits of the same breed, kept under comparable living conditions as the females.			
3.3	**Administration/ Exposure**				
3.3.1	Duration of exposure	Dosed on gestation days 7-19, sacrificed on gestation day 29			
		rabbit	day 7-19	Post- insemination	
3.3.2	Postexposure period	10 days			
		Oral			
3.3.3	Type	Gavage			
3.3.4	Concentration	100%			
3.3.5	Vehicle	Water			
3.3.6	Concentration in vehicle	0, 99, 298, or 895 mg/100mL			
3.3.7	Dose Confirmation	Analytical verification of the homogeneity and stability were conducted by the testing facility prior to study initiation (by colorimetry and titrimetry). Reanalysis of the test substance for verification of stability was determined following the termination of the other toxicological studies with the same test substance. Analytical verification of the test concentrations were conducted prior to start, twice during the study period. Additional analyses were conducted following the study termination on frozen samples.			
3.3.8	Total volume applied	10 mL/kg			
3.3.9	Controls	Yes, concurrent vehicle			
3.4	**Examinations**				
3.4.1	Body weight	Days 0, 2, 4, 7, 9, 11, 14, 16, 19, 21, 23, 25, 29 post-insemination			
3.4.2	Food consumption	Recorded daily			
3.4.3	Clinical signs	Animals were examined for clinical symptoms at least once daily, or more often when signs of toxicity were noted. A mortality check was performed twice daily on work days, and once daily on weekends.			
3.4.4	Examination of uterine content	The uterus and ovaries were removed and the following data recorded: weight of uterus before opening, number of corpora lutea, live fetuses, dead implantations, early resorptions, late resorptions, and dead fetuses.			
3.4.5	Examination of foetuses	Fetuses were dissected from the uterus and further investigated with different methods. They were weighed, sexed, and examined macroscopically for external abnormalities. Half of the fetuses per dam were fixed in ethyl alcohol, and the other in Bouin's solution for further			X

Section A6.8.1(1) Annex Point IIA, VI.6.8.1 IUCLID 5.8.2/01	A6.8.1 Teratogenicity Study (Rabbit) **Study of the prenatal toxicity of glutaraldehyde in rabbits after oral administration (gavage)**	
	evaluation. Evaluation criteria **Table A6.8.1/01-1**.	
3.4.5.1 General	None	
3.4.5.2 Skeletal	A full skeletal examination was performed after the soft tissue examination on all fetuses. The stained skeletons were placed on an illuminated plate and examined, evaluated, and assessed. Evaluation criteria **Table A6.8.1/01-1**.	
3.4.5.3 Soft tissue	The viability of the fetuses and the condition of the placentae, umbilical cords, fetal membranes, and fluids were examined. Individual placental weights were measured. The abdomen and thorax were opened for an in situ examination. The heart and kidneys were removed and sectioned to assess the internal structure. Fetuses were sexed by an internal examination. Heads with abnormalities were detached, sectioned, and examined. Evaluation criteria **Table A6.8.1/01-1**.	
3.5 Statistics	Dunnett's test was used for evaluation of food consumption, water consumption, body weight, weight change, corrected weight gain, weight of the uterus before opening, weight of fetuses, weight of placentae, corpora lutea, implantations, pre-and post-implantation losses, resorptions, and live fetuses. Fisher's exact test was used for statistical evaluation of conception rate, mortality of dams, and fetal findings. Significance was set at $p<0.05$ or $p<0.01$ and was specified with the raw data and in summary tables.	
	4 RESULTS AND DISCUSSION	
4.1 Maternal toxic Effects	There were spontaneous deaths of 5 does in the high dose group (post-insemination days9-11), as well as soft or absent feces and blood in the bedding, but no mortality or unusual behavior noted at the other dose levels. A statistically-significant reduction in food consumption was noted at 45 mg/kg, as well as a related decrease (22%) in body weight. Food consumption and body weights of the other treated groups were practically similar to control animals. Corrected weight gains were not affected. **Table A6.8.1/01-2, 3, 4.** Victims showed GI tract irritation upon necropsy, thinkened pylorus, ulceration, and distended cecum or colon. No such findings were seen in survivors, even at the same dose level.	
4.2 Teratogenic / embryotoxic effects	There were no substantial changes in uterine weight. Conception rates, number of corpora lutea, implantation sites, were unaffected by treatment. In the 45 mg/kg dose group, there was a drastic increase in resorption rates and post-implantation loss (9/15 dams had no viable fetuses), and was assessed as a clear substance-induced finding. **Table A6.8.1/01-6**. Conception rates were 93-100% for all treatment groups. The sex distribution was not markedly affected by treatment, but all fetal indices were difficult to assess at the high dose due to limited number of live fetuses. The weights of placentae were largely unaffected, but were	X

Section A6.8.1(1) Annex Point IIA, VI.6.8.1 IUCLID 5.8.2/01	A6.8.1 Teratogenicity Study (Rabbit) **Study of the prenatal toxicity of glutaraldehyde in rabbits after oral administration (gavage)**	
	lowest at the high dose. There were no differences in fetal weights in the low and mid dose groups, but weights were lower, and outside the historical control range for high dose fetuses. **Table A6.8.1/01-6, 7.** There was one malformation (cheiloschisis) and one variation (pseudoankylois) noted in the control. Sporadic soft tissue observations were made, but incidences were not dose-dependent and were within the historical control range. **Table A6.8.1/01-8, 9, 10.** Skeletal examinations were noted across dose groups, and the only statistically-significant findings were lower incidences of skeletal malformations. Variations and retardations likewise occurred across dose groups, and were within historical control variation. **Table A6.8.1/01-8, 11, 12.**	
4.3 Other effects	None noted	
	5 APPLICANT'S SUMMARY AND CONCLUSION	
5.1 Materials and methods	Animals were dosed via oral gavage from gestation day (GD) 7 to day 19 at dose levels of 0, 5, 15, 45 mg/kg/day. Analytical verification of the test solutions was performed pre-study and twice during the in-life phase. The homogeneity and stability of the solutions was determined pre-study. Food was analyzed by the supplier for contaminants, and the drinking water was analyzed for contaminants by the municipal water authority. The test material was administered to the animals orally by gavage from day 7 post-coitum to day 19 post-coitum, to include the period of major organogenesis. Dose volume was maintained at 10 mL/kg, and gavage doses were given each morning based on the initial, pre-study body weights. Food consumption was recorded daily. Body weights were recorded on days 0, 2, 4, 7, 9, 11, 14, 16, 19, 21, 23, 25, 29. Animals were examined for clinical symptoms at least once daily, or more often when signs of toxicity were noted. A mortality check was performed twice daily on work days, and once daily on weekends. Animals were sacrificed on post-coitum day 29. Dams were assessed by gross pathology. The uterus and oaries were removed and the following data recorded: weight of uterus before opening, number of corpora lutea, live fetuses, dead implantations, early resorptions, late resorptions, and dead fetuses. Conception rate and pre- and post- implantation loss % were calculated. Dams showing signs of abortion were sacrificed immediately and uterine contents were evaluated if possible. Fetuses were dissected from the uterus and further investigated with different methods. They were weighed, sexed, and examined macroscopically for external abnormalities. The viability of the fetuses and the condition of the placentae, umbilical cords, fetal membranes, and fluids were examined. Individual placental weights were measured. Half of the fetuses per dam were fixed in ethyl alcohol, and the other in	

Section A6.8.1(1) **Annex Point IIA, VI.6.8.1** **IUCLID 5.8.2/01**	**A6.8.1 Teratogenicity Study (Rabbit)** **Study of the prenatal toxicity of glutaraldehyde in rabbits after oral administration (gavage)**	
	Bouin's solution for further evaluation. Fetuses were examined for any organ or soft-tissue abnormalities (heart and kidney were sectioned and evaluated), and a skeletal examination was done. Criteria for evaluation of malformations, variations, retardations, and unclassified observations were pre-stated.	
5.2 Results and discussion	Administration of glutaraldehyde produced marked signs of maternal toxicity (decreased food consumption, body weight decrease, spontaneous death, adverse clinical symptoms, irritations of the GI tract, and reduced uterus weights at 45 mg/kg. At this dose, the test material also produced high embryolethality, increased resorption rate, an increase in post-implantation loss, and reduction in mean placental and fetal weights in the few live fetuses that were developed. There were no signs of maternal, embryo, or fetal toxicity at 5 or 15 mg/kg, and no indications of any teratogenicity were present up to and including the highest dose (however few fetuses could be assessed).	
5.3 Conclusion	Glutaraldehyde is not teratogenic to rabbits following oral gavage dosing on gestation days 7-19.	
5.3.1 LO(A)EL maternal toxic effects	45 mg/kg/day based on decreased food consumption and body weight, and the deaths of 5 dams.	
5.3.2 NOEL maternal toxic effects	15 mg/kg/day	
5.3.3 LO(A)EL embryotoxic / teratogenic effects	45 mg/kg/day based on increased resorption rate and increased post-implantation loss	
5.3.4 NOEL embryotoxic / teratogenic effects	15 mg/kg/day	
5.3.5 Reliability	1	
5.3.6 Deficiencies	No	
	Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE	
Date	November 1st, 2010	
Materials and Methods	3.4.5 Examination of foetuses. All foetuses were placed in ethyl alcohol after the soft tissue examination. 5.1 Materials and methods. Same comment as for 3.4.5 above.	
Results and discussion	4.2 Teratogenic/embryotoxic effects: • The uterine weight was drastically reduced in the high dose group (to about 7 % of the control value), while no such effect was seen for the other dose groups.	

Section A6.8.1(1) Annex Point IIA, VI.6.8.1 IUCLID 5.8.2/01	A6.8.1 Teratogenicity Study (Rabbit) **Study of the prenatal toxicity of glutaraldehyde in rabbits after oral administration (gavage)**
	• There was a slightly increased incidence of total foetal skeletal variations (12, 14, 18, 25 %). The 25 % indicates one affected foetus out of the 4 foetuses that survived in the high dose group, and can be disregarded in any analyses. The 18 % at the mid-dose group is mainly due to the findings of sternebrae of irregular shape (6 %, n=5, historical control range 0-8.6 %) and rudimentary cervical ribs (6 %, n=5, historical control range 0-4.5 %). This finding is concluded incidental.
Conclusion	Glutaraldehyde was not teratogenic to Himalayan rabbits. LOAEL (maternal): 45 mg/kg bw/day NOAEL (maternal): 15 mg/kg bw/day LOAEL (teratogenicity): 45 mg/kg bw/day NOAEL (teratogenicity): 15 mg/kg bw/day
Reliability	2
Acceptability	Acceptable
Remarks	The study would not satisfy the requirements of the current OECD 414 guideline with regard to the animals used, and because of the high toxicity at the highest dose level and the lack of toxic effects at the mid-dose level. The requirements are however satisfied of the guideline that was valid at the time of the test (OECD 414 guideline, 1981). The selection of the dose levels (5, 15 and 45 mg/kg bw/day) is poor because of the high toxicity at the high dose level (maternal mortality 5/15, only 4 viable foetuses) and no signs of toxicity at the mid-dose level. The rationale for the dose spacing was based on the two dose levels in the range finding study, where the lower dose level caused no toxicity and was used as the low dose in the main study (5 mg/kg bw/day), and the higher dose was 25 mg/kg bw/day and caused maternal toxicity (gastritis, reduced food consumption). The RMS considers this problem to be due to the very steep dose-mortality curve, and considers that the high dose and mid-dose are sufficiently close to each other for the acceptability of the study. The deficiencies do not compromise the essential results of the study. Please note that the tabulated results in the Tables have not been checked in detail by the RMS.
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*

Section A6.8.1(1) **Annex Point IIA, VI.6.8.1** **IUCLID 5.8.2/01**	**A6.8.1 Teratogenicity Study (Rabbit)** **Study of the prenatal toxicity of glutaraldehyde in rabbits after oral administration (gavage)**	
Reliability	*Discuss if deviating from view of rapporteur member state*	
Acceptability	*Discuss if deviating from view of rapporteur member state*	
Remarks		

Table A6.8.1/01-1 Evaluation Criteria for Assessing Skeletons and Organs of the Fetuses

Classification	Criteria
Malformation *(Concerns external, soft tissue, and skeletal observations)*	Rare and/or probably lethal changes (examples, exencephaly, atresia ani, hernia umbilicalis)
Variations *(Concerns external, soft tissue, and skeletal observations)*	Changes which occur regularly in control animals, and generally have no adverse effect on survival
Retardations *(Concerns skeletal observations only)*	Delays in skeletal development compared with the controls at the time of examination
Unclassified Observations *(concerning external and soft tissue observations only)*	Changes which are not classified as any of the above (examples, focal liver necrosis in fetuses)

Table A6.8.1/01-2 **Mean Maternal Food Consumption & Body Weights (Gestation) – Grams**

		CONTROL		5 mg/kg/day		15 mg/kg/day		45 mg/kg/day	
		FC	BW	FC	BW	FC	BW	FC	BW
DAYS 0 TO 1	MEAN	122.9	2598	110.5	2607	121.0	2616	118.4	2594
	S.D.	16.55	139.8	15.85	112.7	20.76	111.8	17.45	124.5
	N	15	15	14	14	14	14	15	15
DAYS 1 TO 2	MEAN	128.9		118.6		123.9		124.2	
	S.D.	14.63		12.51		18.07		17.52	
	N	15		14		14		15	
DAYS 2 TO 3	MEAN	131.4	2672	120.4	2672	126.0	2671	123.6	2659
	S.D.	15.27	131.6	11.70	129.2	16.01	120.5	14.95	144.5
	N	15	15	14	14	14	14	15	15
DAYS 3 TO 4	MEAN	127.7		115.3		124.5		115.6	
	S.D.	14.84		12.59		15.89		33.46	
	N	15		14		14		15	
DAYS 4 TO 5	MEAN	121.2	2682	112.0	2666	117.6	2677	113.7	2653
	S.D.	16.38	129.0	12.63	129.3	14.07	119.0	25.22	156.4
	N	15	15	14	14	14	14	15	15
DAYS 5 TO 6	MEAN	119.6		107.1		116.5		118.7	
	S.D.	15.43		18.63		16.06		12.27	
	N	15		14		14		15	
DAYS 6 TO 7	MEAN	123.3		118.2		122.6		121.7	
	S.D.	13.50		12.87		14.89		13.82	
	N	15		14		14		15	
DAYS 7 TO 8	MEAN	116.9	2665	108.8	2655	109.2	2671	32.5[b]	2653
	S.D.	12.77	127.9	11.68	118.6	16.28	114.4	24.02	146.0
	N	15	15	14	14	14	14	15	15
DAYS 8 TO 9	MEAN	113.5		108.4		104.8		1.8[b]	
	S.D.	10.51		12.43		19.20		3.60	
	N	15		14		14		13	
DAYS 9 TO 10	MEAN	114.8	2669	109.3	2659	112.1	2674	2.2[b]	2631
	S.D.	9.70	127.1	12.42	126.1	16.04	116.0	2.42	171.0
	N	15	15	14	14	14	14	12	13
DAYS 10 TO 11	MEAN	113.1		106.2		114.5		1.6b	
	S.D.	10.93		12.65		13.36		2.26	
	N	15		14		14		12	
DAYS 11 TO 12	MEAN	106.2	2678	99.1	2663	105.1	2686	1.8[b]	2414[b]
	S.D.	9.20	120.1	8.74	113.9	15.09	114.1	1.96	155.8
	N	15	15	14	14	14	14	10	12
DAYS 12 TO 13	MEAN	101.7		93.3		101.7		4.1b	
	S.D.	14.39		10.44		15.38		3.03	
	N	15		14		13		10	
DAYS 13 TO 14	MEAN	101.9		92.6		101.8		9.0b	
	S.D.	22.35		20.88		21.91		6.66	
	N	15		14		14		10	
DAYS 14 TO 15	MEAN	95.6	2694	84.8	2682	94.4	2698	15.0[b]	2241[b]
	S.D.	15.70	116.2	12.40	123.7	24.95	1123	10.22	125.0
	N	15	15	14	14	14	14	10	10
DAYS 15 TO 16	MEAN	96.5		84.9		90.4		15.7[b]	
	S.D.	11.66		15.23		30.51		10.30	

	N	15		14		14		10	
		CONTROL		**5 mg/kg/day**		**15 mg/kg/day**		**45 mg/kg/day**	
		FC	**BW**	**FC**	**BW**	**FC**	**BW**	**FC**	**BW**
DAYS 16 TO 17	MEAN	99.9	2737	85.7	2717	95.8	2737	17.5[b]	2208[b]
	S.D.	9.89	117.7	20.54	127.4	28.55	107.0	17.60	137.4
	N	15	15	14	14	14	14	10	10
DAYS 17 TO 18	MEAN	105.3		88.6a		95.8		24.7[b]	
	S.D.	7.69		14.89		19.41		24.51	
	N	15		14		14		10	
DAYS 18 TO 19	MEAN	106.0		88.7a		97.5		32.6[b]	
	S.D.	11.68		15.35		16.60		29.24	
	N	15		14		14		10	
DAYS 19 TO 20	MEAN	99.5	2728	88.5	2706	100.4	2730	37.0[b]	2153[b]
	S.D.	13.76	113.5	25.18	119.3	17.96	113.5	31.98	165.8
	N	14	15	14	14	13	14	10	10
DAYS 20 TO 21	MEAN	109.2		97.6		106.3		70.3[b]	
	S.D.	13.27		18.91		22.90		45.09	
	N	15		14		14		10	
DAYS 21 TO 22	MEAN	104.0	2733	98.4	2708	105.5	2736	86.0	2123[b]
	S.D.	15.08	117.4	17.97	121.7	19.82	121.5	48.30	203.9
	N	15	15	14	14	14	14	10	10
DAYS 22 TO 23	MEAN	100.0		99.6		108.7		95.1	
	S.D.	11.14		11.56		12.99		47.92	
	N	15		14		13		10	
DAYS 23 TO 24	MEAN	108.4	2758	110.1	2743	114.4	2763	110.7	2151[b]
	S.D.	13.07	123.0	16.14	128.0	8.69	115.5	55.17	216.2
	N	15	15	14	14	14	14	10	10
DAYS 24 TO 25	MEAN	112.6		112.8		122.4		110.7	
	S.D.	17.92		15.03		11.69		50.22	
	N	15		14		14		10	
DAYS 25 TO 26	MEAN	116.8	2810	110.7	2808	121.6	2817	110.4	2198[b]
	S.D.	21.09	122.9	28.14	136.1	23.54	116.1	40.39	228.5
	N	15	15	14	14	14	14	10	10
DAYS 26 TO 27	MEAN	120.9		116.6		133.6		116.3	
	S.D.	16.68		29.22		14.14		41.58	
	N	15		14		13		10	
DAYS 27 TO 28	MEAN	119.7		119.5		126.0		116.5	
	S.D.	24.13		15.72		16.36		39.67	
	N	15		14		14		10	
DAYS 28 TO 29	MEAN	127.2		123.7		129.2		131.7	
	S.D.	18.61		21.70		17.68		29.87	
	N	15		14		14		10	
DAY 29	MEAN		2901		2911		2908		2299[b]
	S.D.		138.5		148.5		119.9		221.8
	N		15		14		14		10

FC food consumption
BW body weight
SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/01-3 Mean Maternal Food Consumption During Gestation – grams/animal/day

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
DAYS 0 TO 7	MEAN OF MEANS	125.0	114.6b	121.7	119.4
	S.D.	4.34	4.86	3.55	3.98
	N	7	7	7	7
DAYS 7 TO 19	MEAN OF MEANS	105.9	95.9a	101.9	13.2[b]
	S.D.	7.22	9.97	7.48	11.79
	N	12	12	12	12
DAYS 19 TO 29	MEAN OF MEANS	111.8	107.8	116.8	98.5
	S.D.	9.30	11.25	11.31	27.77
	N	10	10	10	10
DAYS 0 TO 29	MEAN OF MEANS	112.6	104.5	111.8	68.2[b]
	S.D.	10.49	12.08	11.88	50.84
	N	29	29	29	29

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/01-4 Mean Maternal Body Weight Change During Gestation – grams

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
DAYS 0 TO 7	MEAN	67.0	47.2	55.6	58.7
	S.D.	41.63	37.82	24.33	43.81
	N	15	14	14	15
DAYS 7 TO 19	MEAN	62.6	51.2	59.3	-446.0[b]
	S.D.	30.83	38.89	49.46	125.28
	N	15	14	14	10
DAYS 19 TO 29	MEAN	173.7	205.2	177.4	146.0
	S.D.	68.18	68.99	79.97	96.97
	N	15	14	14	10
DAYS 0 TO 29	MEAN	303.3	303.5	292.3	-257.8[b]
	S.D.	107.20	101.46	92.75	177.18
	N	15	14	14	10

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/01-5 Mean Gravid Uterine Weights and Net Maternal Body Weight Change – grams

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
GRAVID UTERUS	MEAN	337.9	383.4	325.3	24.4[b]
	S.D.	81.49	78.91	108.80	51.69
	N	15	14	14	10
CARCASS	MEAN	2563.4	2527.6	2582.5	2274.9[b]
	S.D.	100.23	111.06	143.90	198.17
	N	15	14	14	10
NET WEIGHT CHANGE FROM DAY 7	MEAN	-101.6	-127.0	-88.6	-324.4[b]
	S.D.	61.60	42.17	82.79	163.71
	N	15	14	14	10

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.
CARCASS WEIGHT = TERMINAL BODY WEIGHT MINUS UTERINE WEIGHT
NET WEIGHT CHANGE FROM DAY 7 = CARCASS WEIGHT MINUS DAY 7 BODY WEIGHT

Table A6.8.1/01-6 Summary of Reproduction Data

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Females Mated	N	15	15	15	15
Pregnant	N	15	14	14	15
	%	100	93	93	100
Aborted	N	0	0	0	0
Premature Births	N	0	0	0	0
Dams with Viable Fetuses	N	15	14	14	1
Dams with all Resorptions	N	0	0	0	9
Female Mortality	N	0	0	0	5[a]
	%	0.0	0.0	0.0	33
Pregnant at C-section	N	15	14	14	10[a]
	%	100	93	93	67
Corpora Lutea	MEAN	8.5	8.7	8.1	7.6
	S.D.	1.19	1.54	1.07	1.71
	TOTAL	128	122	113	76
Implantation Sites	MEAN	6.9	7.6	6.6	7.2
	S.D.	1.64	1.78	2.10	1.23
	TOTAL	103	107	92	72
Preimplantation Loss	MEAN %	19.2	12.1	19.0	4.0
	S.D.	17.71	13.66	21.85	9.08
Postimplantation Loss	MEAN %	13.6	7.4	12.0	94.3[b]
	S.D.	14.60	13.86	19.12	18.07
Pregnant at C-section	N	15	14	14	10
Resorptions: Total	MEAN	0.9	0.4	0.6	6.8[b]
	S.D.	1.03	0.65	0.84	1.81
	TOTAL	14	6	9	68
	MEAN %	13.6	6.2	12.0	94.3[b]
	S.D.	14.60	10.13	19.12	18.07
Early	MEAN	0.5	0.3	0.2	6.6[b]
	S.D.	0.64	0.47	0.43	2.32
	TOTAL	7	4	3	66
	MEAN %	7.6	4.2	4.3	91.4[b]
	S.D.	10.89	7.16	9.66	27.11
Late	MEAN	0.5	0.1	0.4	0.2
	S.D.	0.83	0.36	0.65	0.63
	TOTAL	7	2	6	2
	MEAN %	6.0	2.0	7.7	2.9
	S.D.	9.54	5.16	11.87	9.04
Dead Fetuses	N	0	1	0	0
Dams with Viable Fetuses	N	15	14	14	1
Live Fetuses	MEAN	5.9	7.1	5.9	4.0
	S.D.	1.71	2.11	2.37	0.00
	TOTAL	89	100	83	4
	MEAN %	86.4	92.6	88.0	57.1
	S.D.	14.60	13.86	19.12	0.00

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Females Mated	N	15	15	15	15
Females	MEAN	3.6	3.4	3.4	4.0
	S.D.	1.24	1.95	1.86	0.00
	TOTAL	54	48	47	4
	MEAN %	52.8	43.9	51.0	57.1
	S.D.	14.38	22.21	27.13	0.00
Males	MEAN	2.3	3.7	2.6	0.0
	S.D.	1.23	1.64	1.83	0.00
	TOTAL	35	52	36	0
	MEAN %	33.6	48.7	37.0	0.0
	S.D.	15.49	18.54	20.37	0.00
PERCENT LIVE FEMALES		60.7	48.0	56.6	100.0
PERCENT LIVE MALES		39.3	52.0	43.4	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/01-7 Mean Placental and Fetal Body Weights

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
PLACENTAL WEIGHTS (grams)					
of all Viable Fetuses	MEAN	4.9	4.6	4.6	4.1
	S.D.	0.67	0.74	0.70	0.00
	N	15	14	14	1
of Male Fetuses	MEAN	5.1	4.6	4.9	
	S.D.	0.89	0.83	0.71	
	N	15	14	13	
of Female Fetuses	MEAN	4.8	4.4	4.3	4.1
	S.D.	0.65	0.81	0.54	0.00
	N	15	14	13	1
FETAL WEIGHTS (grams)					
of all Viable Fetuses	MEAN	41.7	40.2	41.3	26.9[b]
	S.D.	2.68	4.87	3.92	0.00
	N	15	14	14	1
of Male Fetuses	MEAN	42.1	40.3	42.3	
	S.D.	4.05	5.79	4.10	
	N	15	14	13	
of Female Fetuses	MEAN	41.5	39.2	40.2	26.9[b]
	S.D.	3.03	4.66	3.66	0.00
	N	15	14	13	1

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/01-8 **Summary of All Classified Fetal Observations (External, Soft Tissue, and Skeletal)**

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Litters Evaluated	N	15	14	14	1
Fetuses Evaluated	N	89	101	83	4
Live	N	89	100	83	4
Dead	N	0	1	0	0
TOTAL MALFORMATIONS					
Fetal Incidence	N	4	2	2	0
	%	4.5	2.0	2.4	0.0
Litter Incidence	N	2	2	2	0
	%	13	14	14	0.0
Affected Fetuses/ Litter	MEAN %	3.7	2.1	1.7	0.0
	S.D.	11.30	5.79	4.30	0.00
TOTAL VARIATIONS					
Fetal Incidence	N	53	62	51	3
	%	60	61	61	75
Litter Incidence	N	15	14	14	1
	%	100	100	100	100
Affected Fetuses/ Litter	MEAN %	61.1	64.3	62.1	75.0
	S.D.	21.10	26.51	20.34	0.00
TOTAL RETARDATIONS					
Fetal Incidence	N	68	77	64	4
	%	76	76	77	100
Litter Incidence	N	15	14	14	1
	%	100	100	100	100
Affected Fetuses/ Litter	MEAN %	74.6	75.7	76.6	100.0
	S.D.	25.80	15.36	19.57	0.00

Table A6.8.1/01-9 Summary of All Fetal External Observations

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Litters Evaluated	N	15	14	14	1
Fetuses Evaluated	N	89	101	83	4
Live	N	89	100	83	4
Dead	N	0	1	0	0
SUMMARY- ALL CLASSIFIED OBSERVATIONS					
TOTAL MALFORMATIONS					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
Affected Fetuses/Litter	MEAN %	1.0	0.0	0.0	0.0
	S.D.	3.69	0.00	0.00	0.00
TOTAL VARIATIONS					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
Affected Fetuses/Litter	MEAN %	0.8	0.0	0.0	0.0
	S.D.	3.23	0.00	0.00	0.00
EXTERNAL MALFORMATIONS					
CHEILOSCHISIS					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
TOTAL FETAL EXTERNAL MALFORMATIONS					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
TOTAL FETAL EXTERNAL UNCLASSIFIED OBSERVATIONS					
Fetal Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0

Table A6.8.1/01-10 Summary of All Fetal Soft Tissue Observations

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Litters Evaluated	N	15	14	14	1
Fetuses Evaluated	N	89	101	83	4
Live	N	89	100	83	4
Dead	N	0	1	0	0
FETAL SOFT TISSUE MALFORMATIONS					
HYDROCEPHALY					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
TRUNCUS ARTERIOSUS COMMUNIS					
Fetal Incidence	N	0	0	1	0
	%	0.0	0.0	1.2	0.0
Litter Incidence	N	0	0	1	0
	%	0.0	0.0	7.1	0.0
AGENESIA OF GALLBLADDER					
Fetal Incidence	N	0	2	1	0
	%	0.0	2.0	1.2	0.0
Litter Incidence	N	0	2	1	0
	%	0.0	14	7.1	0.0
TOTAL FETAL SOFT TISSUE MALFORMATIONS					
Fetal Incidence	N	1	2	1	0
	%	1.1	2.0	1.2	0.0
Litter Incidence	N	1	2	1	0
	%	6.7	14	7.1	0.0
FETAL SOFT TISSUE VARIATIONS					
SEPARATED ORIGIN OF CAROTIDS					
Fetal Incidence	N	33	32	32	2
	%	37	32	39	50
Litter Incidence	N	14	14	12	1
	%	93	100	86	100
HEART: TRACES OF INTERVENTRIC. FORAMEN/SEPTUM MEMBRANACEUM					
Fetal Incidence	N	19	24	21	2
	%	21	24	25	50
Litter Incidence	N	12	10	10	1
	%	80	71	71	100
TOTAL FETAL SOFT TISSUE VARIATIONS					
Fetal Incidence	N	45	52	41	3
	%	51	51	49	75
Litter Incidence	N	15	14	12	1
	%	100	100	86	100

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Litters Evaluated	N	15	14	14	1
Fetuses Evaluated	N	89	101	83	4
Live	N	89	100	83	4
Dead	N	0	1	0	0
FETAL SOFT TISSUE UNCLASSIFIED OBSERVATIONS					
POST MORTEM AUTOLYSIS					
Fetal Incidence	N	0	1	0	0
	%	0.0	1.0	0.0	0.0
Litter Incidence	N	0	1	0	0
	%	0.0	7.1	0.0	0.0
LIVER: FOCAL NECROSIS					
Fetal Incidence	N	0	3	0	0
	%	0.0	3.0	0.0	0.0
Litter Incidence	N	0	2	0	0
	%	0.0	14	0.0	0.0
TOTAL FETAL SOFT TISSUE UNCLASSIFIED OBSERVATIONS					
Fetal Incidence	N	0	4	0	0
	%	0.0	4.0	0.0	0.0
Litter Incidence	N	0	3	0	0
	%	0.0	21	0.0	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/01-11 Summary of All Classified Fetal Skeletal Observations

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Litters Evaluated	N	15	14	14	1
Fetuses Evaluated	N	89	101	83	4
Live	N	89	100	83	4
Dead	N	0	1	0	0
TOTAL MALFORMATIONS					
Fetal Incidence	N	4	0a	1	0
	%	4.5	0.0	1.2	0.0
Litter Incidence	N	2	0	1	0
	%	13	0.0	7.1	0.0
Affected Fetuses/ Litter	MEAN %	3.7	0.0	0.8	0.0
	S.D.	11.30	0.00	2.97	0.00
TOTAL VARIATIONS					
Fetal Incidence	N	11	14	15	1
	%	12	14	18	25
Litter Incidence	N	8	9	10	1
	%	53	64	71	100
Affected Fetuses/ Litter	MEAN %	12.1	14.9	22.2	25.0
	S.D.	13.06	15.10	26.48	0.00
TOTAL RETARDATIONS					
Fetal Incidence	N	68	77	64	4
	%	76	76	77	100
Litter Incidence	N	15	14	14	1
	%	100	100	100	100
Affected Fetuses/ Litter	MEAN %	74.6	75.7	76.6	100.0
	S.D.	25.80	15.36	19.57	0.00

Table A6.8.1/01-12 Summary of All Fetal Skeletal Observations

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Litters Evaluated	N	15	14	14	1
Fetuses Evaluated	N	89	101	83	4
Live	N	89	100	83	4
Dead	N	0	1	0	0
MALFORMATIONS					
SKULL BONES FUSED					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
LUMBAR VERTEBRA ABSENT					
Fetal Incidence	N	2	0	1	0
	%	2.2	0.0	1.2	0.0
Litter Incidence	N	1	0	1	0
	%	6.7	0.0	7.1	0.0
STERNEBRAE WITH VARIOUS MALFORMATIONS					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
STERNEBRAE SEVERELY FUSED (BONY PLATE)					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
TOTAL FETAL SKELETAL MALFORMATIONS					
Fetal Incidence	N	4	0a	1	0
	%	4.5	0.0	1.2	0.0
Litter Incidence	N	2	0	1	0
	%	13	0.0	7.1	0.0
EPACTAL BONE BETWEEN NASAL AND FRONTAL BONES					
Fetal Incidence	N	1	1	2	0
	%	1.1	1.0	2.4	0.0
Litter Incidence	N	1	1	2	0
	%	6.7	7.1	14	0.0
SPLITTING OF SKULL BONE(S)					
Fetal Incidence	N	0	3	1	0
	%	0.0	3.0	1.2	0.0
Litter Incidence	N	0	3	1	0
	%	0.0	21	7.1	0.0
ACCESSORY THORACIC VERTEBRA					
Fetal Incidence	N	0	0	1	0
	%	0.0	0.0	1.2	0.0
Litter Incidence	N	0	0	1	0
	%	0.0	0.0	7.1	0.0

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Litters Evaluated	N	15	14	14	1
Fetuses Evaluated	N	89	101	83	4
Live	N	89	100	83	4
Dead	N	0	1	0	0
STERNEBRAE FUSED					
Fetal Incidence	N	4	0a	2	0
	%	4.5	0.0	2.4	0.0
Litter Incidence	N	2	0	2	0
	%	13	0.0	14	0.0
ACCESORY STERNEBRA					
Fetal Incidence	N	0	3	1	0
	%	0.0	3.0	1.2	0.0
Litter Incidence	N	0	2	1	0
	%	0.0	14	7.1	0.0
STERNEBRA(E) OF IRREGULAR SHAPE					
Fetal Incidence	N	1	4	5	1
	%	1.1	4.0	6.0	25
Litter Incidence	N	1	4	4	1
	%	6.7	29	29	100
ACCESSORY 13TH RIB(S)					
Fetal Incidence	N	1	3	1	0
	%	1.1	3.0	1.2	0.0
Litter Incidence	N	1	2	1	0
	%	6.7	14	7.1	0.0
VARIATIONS					
12TH RIB(S) SHORTENED					
Fetal Incidence	N	1	0	1	0
	%	1.1	0.0	1.2	0.0
Litter Incidence	N	1	0	1	0
	%	6.7	0.0	7.1	0.0
WAVY RIB(S)					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
RUDIMENTARY CERVICAL RIB(S)					
Fetal Incidence	N	2	1	5	0
	%	2.2	1.0	6.0	0.0
Litter Incidence	N	2	1	3	0
	%	13	7.1	21	0.0
TOTAL FETAL SKELETAL VARIATIONS					
Fetal Incidence	N	11	14	15	1
	%	12	14	18	25
Litter Incidence	N	8	9	10	1
	%	53	64	71	100

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Litters Evaluated	N	15	14	14	1
Fetuses Evaluated	N	89	101	83	4
Live	N	89	100	83	4
Dead	N	0	1	0	0
SKULL INCOMPLETELY OSSIFIED					
Fetal Incidence	N	2	1	1	0
	%	2.2	1.0	1.2	0.0
Litter Incidence	N	2	1	1	0
	%	13	7.1	7.1	0.0
ENLARGED FRONTAL FONTANELLE					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
THORACIC VERTEBRAL BODY/BODIES DUMBBELL-SHAPED (SYMMETR.)					
Fetal Incidence	N	2	0	1	0
	%	2.2	0.0	1.2	0.0
Litter Incidence	N	2	0	1	0
	%	13	0.0	7.1	0.0
LUMBAR VERTEBRAL ARCH(ES) INCOMPLETELY OSSIFIED					
Fetal Incidence	N	2	3	2	0
	%	2.2	3.0	2.4	0.0
Litter Incidence	N	2	3	1	0
	%	13	21	7.1	0.0
SACRAL VERTEBRAL ARCH(ES) INCOMPLETELY OSSIFIED					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
STERNEBRA(E) NOT OSSIFIED					
Fetal Incidence	N	31	40	25	3
	%	35	40	30	75
Litter Incidence	N	11	13	11	1
	%	73	93	79	100
STERNEBRA(E) INCOMPLETELY OSSIFIED OR REDUCED IN SIZE					
Fetal Incidence	N	34	37	38	0
	%	38	37	46	0.0
Litter Incidence	N	13	14	12	0
	%	87	100	86	0.0
PHALANGES NOT OSSIFIED (FORELIMB)					
Fetal Incidence	N	1	0	0	1
	%	1.1	0.0	0.0	25
Litter Incidence	N	1	0	0	1
	%	6.7	0.0	0.0	100

		Control	5 mg/kg/day	15 mg/kg/day	45 mg/kg/day
Litters Evaluated	N	15	14	14	1
Fetuses Evaluated	N	89	101	83	4
Live	N	89	100	83	4
Dead	N	0	1	0	0
PHALANGES INCOMPLETELY OSSIFIED (FORELIMB)					
Fetal Incidence	N	1	3	1	0
	%	1.1	3.0	1.2	0.0
Litter Incidence	N	1	2	1	0
	%	6.7	14	7.1	0.0
CALCANEUS INCOMPLETELY OSSIFIED					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
TALUS NOT OSSIFIED					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
TALUS INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	2	0	0
	%	0.0	2.0	0.0	0.0
Litter Incidence	N	0	1	0	0
	%	0.0	7.1	0.0	0.0
PHALANGES NOT OSSIFIED (HINDLIMB)					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
OS PUBIS INCOMPLETELY OSSIFIED					
Fetal Incidence	N	1	0	0	0
	%	1.1	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	6.7	0.0	0.0	0.0
TOTAL FETAL SKELETAL RETARDATIONS					
Fetal Incidence	N	68	77	64	4
	%	76	76	77	100
Litter Incidence	N	15	14	14	1
	%	100	100	100	100

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Section A6.8.1(2) Annex Point IIA, VI.6.8.1 IUCLID 5.8.2/02	A6.8.1 Teratogenicity Study	
	1 REFERENCE (A6.8.1/02)	**Official use only**
1.1 Reference	[redacted] (1991b) Study of the prenatal toxicity of glutaraldehyde in rats after oral administration (drinking water), [redacted], Unpublished, 8 February 1991	
1.2 Data protection	Yes	
1.2.1 Data owner	[redacted] The Dow Chemical Company (Dow)	
1.2.2 Companies with letter of access	Dow [redacted]	
1.2.3 Criteria for data protection	Data on an existing active substance for first entry to Annex I authorisation	
	2 GUIDELINES AND QUALITY ASSURANCE	
2.1 Guideline study	Yes OECD 414	
2.2 GLP	Yes	
2.3 Deviations	Not applicable	
	3 MATERIALS AND METHODS	
3.1 Test material	Glutaraldehyde, 50%	
3.1.1 Lot/Batch number	[redacted]	
3.1.2 Specification	Not reported	
3.1.2.1 Description	watery solution	
3.1.2.2 Purity	[redacted]	
3.1.2.3 Stability	Assumed to be stable under typical storage conditions	X
3.2 Test Animals		
3.2.1 Species	Rat	
3.2.2 Strain	[redacted]	
3.2.3 Source	[redacted]	
3.2.4 Sex	Female	
3.2.5 Age/weight at study initiation	68-77 days of age 230 grams (mean)	
3.2.6 Number of animals per group	25	
3.2.7 Control animals	Yes	

Section A6.8.1(2) Annex Point IIA, VI,6.8.1 IUCLID 5.8.2/02		A6.8.1 Teratogenicity Study			
3.2.8	Mating period	One male was allowed to mate with up to 5 females. The day a sperm-positive vaginal smear was taken was considered GD0.			X
3.3	**Administration/ Exposure**				
3.3.1	Duration of exposure	11 days, with necropsies on GD 20.			
		rat	Day 6-16	Post-mating	
3.3.2	Postexposure period	Animals were sacrificed and necropsied on Day 20 of gestation.			
		Oral			
3.3.3	Type	Drinking water			
3.3.4	Concentration	Pure water			
3.3.5	Vehicle	Drinking water			
3.3.6	Concentration in vehicle	50, 250 or 750 ppm (0, 5, 26, 68 mg/kg/day)			
3.3.7	Dose Confirmation	Analytical verification of the test solutions was performed pre-study and twice during the in-life phase. The homogeneity and stability of the solutions was determined pre-study.			
3.3.8	Total volume applied	Not applicable. Drinking water containing the test material was offered *ad libitum*.			
3.3.9	Controls	Yes, concurrent vehicle			
3.4	**Examinations**				
3.4.1	Body weight	Gestation Days 0, 1, 3, 6, 8, 10, 13, 15, 17, 20			
3.4.2	Food consumption	Gestation Days 0, 1, 3, 6, 8, 10, 13, 15, 17, 20			
3.4.3	Water consumption	Gestation Days 1, 3, 6, 8, 10, 13, 15, 16, 17, 20			
3.4.4	Clinical signs	Animals were examined for clinical symptoms at least once daily, or more often when signs of toxicity were noted. A mortality check was performed twice daily on work days, and once daily on weekends.			
3.4.5	Examination of uterine content	Dams were assessed by gross pathology. The uterus and ovaries were removed and the following data recorded: weight of uterus before opening, number of corpora lutea, live fetuses, dead implantations, early resorptions, late resorptions, and dead fetuses. Conception rate and pre- and post- implantation loss % were calculated.			
3.4.6	Examination of foetuses	Fetuses were dissected from the uterus and further investigated with different methods. They were weighed, sexed, and examined macroscopically for external abnormalities. The viability of the fetuses and the condition of the placentae, umbilical cords, fetal membranes, and fluids were examined. Individual placental weights were measured. Half of the fetuses per dam were fixed in ethyl alcohol, and the other in BOUIN's solution for further evaluation			
3.4.6.1	General	None			
3.4.6.2	Skeletal	A skeletal examination was done under a stereomicroscope. ***Table A6.8.1/02-1.***			

Section A6.8.1(2) Annex Point IIA, VI,6.8.1 IUCLID 5.8.2/02	A6.8.1 Teratogenicity Study	
3.4.6.3 Soft tissue	Fetuses were examined for any organ or soft-tissue abnormalities. *Table A6.8.1/02-1.*	
3.5 Statistics	Dunnett's test was used for evaluation of food consumption, water consumption, body weight, weight change, corrected weight gain, weight of the uterus before opening, weight of fetuses, weight of placentae, corpora lutea, implantations, pre-and post-implantation losses, resorptions, and live fetuses. Fisher's exact test was used for statistical evaluation of conception rate, mortality of dams, and fetal findings. Significance was set at $p<0.05$ or $p<0.01$ and was specified with the raw data.	
	4 RESULTS AND DISCUSSION	
4.1 Maternal toxic Effects	There was no observable difference between treated and control dams for food consumption. Water consumption was less for high and mid dose dams, and was attributed to an aversion to the taste or smell of the dose solutions, **Table A6.8.1/02-2**. All body weights were statistically similar to the controls, and are fully in the range of normal variation. Body weight gains were likewise similar to the control animals. None of the dams showed any remarkable clinical observations during the study period. All observations noted at gross pathology were spontaneous in origin, and not attributed to treatment. Uterine weight was not affected by treatment, **Table A6.8.1/02-3.** There were no adverse, substance-related, or statistically significant findings in reproductive endpoints for dams, **Table A6.8.1/02-4**.	
4.2 Teratogenic / embryotoxic effects	The sex distribution of the fetuses was comparable to the control litters, weights of placentae and fetuses were not affected by treatment, **Table A6.8.1/02-5**. External examinations yielded no findings, **Table A6.8.1/02-6, 7**. There was one aglossotomia and one fused placentae noted, but historical control supports a low frequency of such observations, and they were not considered attributable to treatment. Soft tissue exams revealed one situs inversus, and dilated renal pelvis across all dose groups without any clear dose-response relationship. **Table A6.8.1/02-6, 8.** Skeletal examinations revealed a number of malformations of the skull, but the only statistically significant finding was a lower number of 250ppm fetuses with malformations. Variations that were noted were related to the ribs, sternum, vertebral column, and were found in all dose groups. The only statistically significant notation was a decreased occurrence of variations in the low dose group, but is considered without any biological relevance. Retardations of vertebral bodies, sternebrae, and/or hyoid bone were noted, but incidences were within the historical control range. **Table A6.8.1/02-6, 9, 10**.	X
4.3 Other effects	None	
	5 APPLICANT'S SUMMARY AND CONCLUSION	
5.1 Materials and	Animals were dosed via the drinking water from gestation day (GD) 6	

Section A6.8.1(2) Annex Point IIA, VI.6.8.1 IUCLID 5.8.2/02		A6.8.1 Teratogenicity Study	
	methods	to day 16 at dose levels of 0, 50, 250, or 750 ppm. Analytical verification of the test solutions was performed pre-study and twice during the in-life phase. The homogeneity and stability of the solutions was determined pre-study. Food was analyzed by the supplier for contaminants, and the drinking water was analyzed for contaminants by the municipal water authority. The test material was administered to the animals orally in the drinking water from day 6 post-coitum to day 16 post-coitum, to include the period of major organogenesis. Body weights, food consumption, and water consumption was recorded on days 0, 1, 3, 6, 8, 10, 13, 15, 16 (water only) 17, and 20. Animals were examined for clinical symptoms at least once daily, or more often when signs of toxicity were noted. A mortality check was performed twice daily on work days, and once daily on weekends. Animals were sacrificed on post-coitum day 20. Dams were assessed by gross pathology. The uterus and ovaries were removed and the following data recorded: weight of uterus before opening, number of corpora lutea, live fetuses, dead implantations, early resorptions, late resorptions, and dead fetuses. Conception rate and pre- and post-implantation loss % were calculated. Fetuses were dissected from the uterus and further investigated with different methods. They were weighed, sexed, and examined macroscopically for external abnormalities, soft tissue and skeletal changes. The viability of the fetuses and the condition and weight of the placentae, umbilical cords, fetal membranes, and fluids were examined. Criteria for evaluation of malformations, variations, retardations, and unclassified observations were pre-stated.	
5.2	**Results and discussion**	Clear signs of maternal toxicity could not be induced in the study due to water aversion at high doses. There were no signs of embryo-/fetotoxicity, and especially no indications of any teratogenic effect in any of the dose groups.	
5.3	**Conclusion**	Glutaraldehyde was negative for teratogenicity in rats when dosed via the drinking water up to 750 ppm.	
5.3.1	LO(A)EL maternal toxic effects	26 mg/kg based on decreased body weights	
5.3.2	NOEL maternal toxic effects	5 mg/kg based on water consumption	
5.3.3	LO(A)EL embryotoxic / teratogenic effects	>68 mg/kg (>750 ppm)	
5.3.4	NOEL embryotoxic / teratogenic effects	68 mg/kg (750 ppm)	
5.3.5	Reliability	1	

Section A6.8.1(2) **Annex Point IIA, VI,6.8.1** **IUCLID 5.8.2/02**	**A6.8.1 Teratogenicity Study**	
5.3.6 Deficiencies	The high dose level was limited by palatability of the drinking water solution. Clear signs of maternal toxicity could not be induced in the study due to water aversion at high doses.	
	Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE	
Date	November 2nd, 2010	
Materials and Methods	3.1.2.3 Stability. The stability was verified analytically. 3.2.8 Mating period. One male was mated with 3-4 females (not 5).	
Results and discussion	4.2 Teratogenic/embryotoxic effects. The following details should be added/corrected: • Soft tissue findings. The foetus with aglossotomia was in the high dose group and the fused placenta in the low dose group. These findings are considered incidental. *Situs inversus* was in the control group. • Total skeletal retardations. The foetal incidence was slightly increased in all dose groups, but without statistical significance or dose relation (37, 52, 45, 46 %). The historical mean value is 41 %. • Sternebrae not ossified. There was an increase in foetal incidence (7, 12, 12, 10 %) and litter incidence (32, 55, 48, 52 %). There was no statistical significance or dose relation. The foetal incidences are within historical control range (4.1-14.7 %), and the litter incidence slightly above the control range (28-50 %). • Sternebrae incompletely ossified or reduced in size. There was an increase in foetal incidence (11, 25, 19, 20 %) and litter incidence (42, 73, 48, 83 %). Statistical significance was reached for foetal incidence in the low dose group, and for litter incidence in the high dose group, but the values are within the historical control range (8.6-32 % and 40-90 %). • Note that the findings above are selected as possible indications of teratogenic effects. For several other effects the dose relation is reversed, i.e. the highest incidences are in the control group. All findings listed in these bullet points are considered incidental.	
Conclusion	Glutaraldehyde was not teratogenic or embryotoxic when tested at concentrations up to 35 mg GA/kg bw/day, equivalent to 68 mg test substance/kg bw/day (see Remarks below). NOAEL for maternal toxicity, embryotoxicity and teratogenicity is the highest dose used, 35 mg GA/kg bw/day. LOAEL was not established for maternal or foetal effects.	
Reliability	2 The value of the study would be higher if maternal toxicity were reached. This could have been achieved by using other dosing methods, as it is known that GA causes aversion when administered in the drinking water.	
Acceptability	Acceptable	

Section A6.8.1(2) Annex Point IIA, VI,6.8.1 IUCLID 5.8.2/02	A6.8.1 Teratogenicity Study	
Remarks	RMS concludes that the study report indicates the final results for the test substance, not GA, regardless of indicating the GA concentrations in drinking water as well. The corrected concentrations are given only in the RMS Conclusion above. An explanation follows since this might otherwise be controversial: 1. High dose group received 750 ppm GA in water, containing 149.1 mg/100 ml. 2. The high dose group consumed approximately 25 g water per day (equal to 25 ml). 3. An estimation of the high dose group GA intake per day can be calculated as follows: 25 ml × 149.1 /100 mg/ml = 37 mg. 4. The calculation above is approximated, and the GA intake brought forward is calculated using 51 % of the reported 68 mg/kg bw/day = 35 mg/kg bw/day. Please note that the tabulated results in the Tables have not been checked in detail by the RMS.	
	COMMENTS FROM ...	
Date	*Give date of comments submitted*	
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*	
Results and discussion	*Discuss if deviating from view of rapporteur member state*	
Conclusion	*Discuss if deviating from view of rapporteur member state*	
Reliability	*Discuss if deviating from view of rapporteur member state*	
Acceptability	*Discuss if deviating from view of rapporteur member state*	
Remarks		

Table A6.8.1/02-1 Evaluation Criteria for Assessing Skeletons and Organs of the Fetuses

Classification	Criteria
Malformation *(Concerns external, soft tissue, and skeletal observations)*	**Rare and/or probably lethal changes (examples, exencephaly, atresia ani, hernia umbilicalis)**
Variations *(Concerns external, soft tissue, and skeletal observations)*	**Changes which occur regularly in control animals, and generally have no adverse effect on survival (example, dilated renal pelvis)**
Retardations *(Concerns skeletal observations only)*	**Delays in skeletal development compared with the controls at the time of examination (example, sternebrae not ossified)**
Unclassified Observations *(concerning external and soft tissue observations only)*	**Changes which are not classified as any of the above (examples, blood coagulum around placentae)**

Table A6.8.1/02-2 Mean Maternal Water Consumption During Gestation – grams/animal/day

		Control	50 ppm	250 ppm	750 ppm
DAYS 0 TO 1	MEAN	22.4	21.4	21.1	21.1
	S.D.	4.63	4.47	4.06	4.85
	N	20	22	23	23
DAYS 1 TO 3	MEAN	26.4	26.0	25.9	27.1
	S.D.	3.05	3.53	3.12	4.30
	N	20	22	23	23
DAYS 3 TO 6	MEAN	27.1	27.3	26.7	27.8
	S.D.	3.71	3.70	3.79	4.03
	N	20	22	23	23
DAYS 6 TO 8	MEAN	26.4	26.6	26.8	22.1[a]
	S.D.	4.76	3.36	6.66	3.04
	N	19	21	22	23
DAYS 8 TO 10	MEAN	28.2	26.2	26.2	23.1[b]
	S.D.	5.53	3.95	4.88	2.52
	N	20	22	23	23
DAYS 10 TO 13	MEAN	32.5	30.4	28.9a	26.3[b]
	S.D.	4.15	4.63	4.70	2.80
	N	20	22	23	22
DAYS 13 TO 15	MEAN	33.5	31.4	29.5a	27.8[b]
	S.D.	4.39	6.65	4.90	3.06
	N	19	22	23	23
DAYS 15 TO 16	MEAN	36.5	32.5	32.7	30.8[b]
	S.D.	6.28	6.28	5.11	6.57
	N	20	22	23	23
DAYS 16 TO 17	MEAN	41.7	38.5	40.2	40.7
	S.D.	5.21	6.69	7.21	4.86
	N	20	22	23	23
DAYS 17 TO 20	MEAN	39.4	37.4	39.6	40.8
	S.D.	4.75	5.99	5.73	4.54
	N	20	22	23	23

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/02-3 Mean Gravid Uterine Weights and Net Maternal Body Weight Change

– grams

		Control	50 ppm	250 ppm	750 ppm
GRAVID UTERUS	MEAN	74.9	77.8	82.2	80.4
	S.D.	25.13	16.32	16.14	14.73
	N	20	22	23	23
CARCASS	MEAN	309.7	304.1	302.5	310.8
	S.D.	22.57	17.67	17.01	17.13
	N	20	22	23	23
NET WEIGHT CHANGE FROM DAY 6	MEAN	44.9	43.9	44.8	48.5
	S.D.	8.53	8.45	7.23	9.32
	N	20	22	23	23

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.
CARCASS WEIGHT = TERMINAL BODY WEIGHT MINUS UTERINE WEIGHT
NET WEIGHT CHANGE FROM DAY 6 = CARCASS WEIGHT MINUS DAY 6 BODY WEIGHT

Table A6.8.1/02-4 Summary of Reproduction Data

		Control	50 ppm	250 ppm	750 ppm
Females Mated	N	25	25	25	25
Pregnant	N	20	22	23	23
	%	80	88	92	92
Aborted	N	0	0	0	0
Premature Births	N	0	0	0	0
Dams with Viable Fetuses	N	19	22	23	23
Dams with all Resorptions	N	1	0	0	0
Female Mortality	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Pregnant at C-section	N	20	22	23	23
	%	80	88	92	92
Corpora Lutea	MEAN	17.3	16.1	16.2	16.1
	S.D.	1.75	2.20	1.64	1.35
	TOTAL	346	354	372	370
Implantation Sites	MEAN	15.4	14.6	15.0	14.7
	S.D.	3.90	3.03	2.85	2.65
	TOTAL	307	321	344	338

Preimplantation Loss	MEAN %	11.6	9.4	8.2	8.9
	S.D.	19.52	13.95	12.08	13.54
Postimplantation Loss	MEAN %	14.3	5.9	4.0[a]	5.1a
	S.D.	22.04	5.72	5.82	7.33
Pregnant at C-section	N	20	22	23	23
Resorptions: Total	MEAN	2.4	0.9[a]	0.6[a]	0.7[a]
	S.D.	3.78	0.89	0.84	1.05
	TOTAL	48	19	13	17
	MEAN %	14.3	5.9	4.0[a]	5.1[a]
	S.D.	22.04	5.72	5.82	7.33
Early	MEAN	2.2	0.7a	0.6[a]	0.7[a]
	S.D.	3.76	0.77	0.84	1.06
	TOTAL	44	16	13	16
	MEAN %	13.1	5.1	4.0a	4.8
	S.D.	22.00	5.17	5.82	7.39
Late	MEAN	0.2	0.1	0.0	0.0
	S.D.	0.41	0.35	0.00	0.21
	TOTAL	4	3	0	1
	MEAN %	1.2	0.9	0.0	0.3
	S.D.	2.53	2.22	0.00	1.39
Dead Fetuses	N	0	0	0	0
Dams with Viable Fetuses	N	19	22	23	23
Live Fetuses	MEAN	13.6	13.7	14.4	14.0
	S.D.	3.53	2.98	2.90	2.87
	TOTAL	259	302	331	321
	MEAN %	90.2	94.1	96.0[a]	94.9
	S.D.	9.10	5.72	5.82	7.33

		Control	50 ppm	250 ppm	750 ppm
Females Mated	N	25	25	25	25
Females	MEAN	6.2	7.1	6.5	7.5
	S.D.	2.85	1.96	2.31	2.56
	TOTAL	117	157	149	172
	MEAN %	40.4	49.9	44.6	50.7
	S.D.	13.24	13.16	16.49	13.86
Males	MEAN	7.5	6.6	7.9	6.5
	S.D.	2.22	2.95	3.12	2.29
	TOTAL	142	145	182	149
	MEAN %	49.9	44.2	51.4	44.2
	S.D.	9.98	15.10	16.49	13.55
Percent Live Females		45.2	52.0	45.0	53.6
Percent Live Males		54.8	48.0	55.0	46.4

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/02-5 Mean Placental and Fetal Body Weights

		Control	50 ppm	250 ppm	750 ppm
		PLACENTAL WEIGHTS UNITS: GRAMS			
of all viable fetuses	MEAN	0.47	0.47	0.46	0.46
	S.D.	0.036	0.049	0.044	0.047
	N	19	22	23	23
of Male Fetuses	MEAN	0.48	0.47	0.47	0.47
	S.D.	0.038	0.055	0.046	0.051
	N	19	22	23	23
of Female Fetuses	MEAN	0.47	0.46	0.46	0.46
	S.D.	0.039	0.052	0.059	0.048
	N	19	22	23	23
		FETAL WEIGHTS UNITS: GRAMS			
of all viable fetuses	MEAN	3.9	3.8	3.9	3.9
	S.D.	0.27	0.18	0.20	0.22

	N	19	22	23	23
of Male Fetuses	MEAN	3.9	3.9	4.0	3.9
	S.D.	0.25	0.23	0.21	0.24
	N	19	22	23	23
of Female Fetuses	MEAN	3.8	3.7	3.8	3.8
	S.D.	0.29	0.18	0.22	0.20
	N	19	22	23	23

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/02-6 Summary of All Classified Fetal Observations (External, Soft Tissue, and Skeletal)

		Control	50 ppm	250 ppm	750 ppm
Litters Evaluated	N	19	22	23	23
Fetuses Evaluated	N	259	302	331	321
Live	N	259	302	331	321
Dead	N	0	0	0	0
		TOTAL MALFORMATIONS			
Fetal Incidence	N	10	8	2[b]	9
	%	3.9	2.6	0.6	2.8
Litter Incidence	N	8	8	2[a]	7
	%	42	36	8.7	30
Affected Fetuses/ Litter	MEAN %	4.1	2.7	0.6a	2.6
	S.D.	5.68	3.87	2.07	4.29
		TOTAL VARIATIONS			
Fetal Incidence	N	76	60[a]	84	75
	%	29	20	25	23
Litter Incidence	N	19	18	23	22
	%	100	82	100	96
Affected Fetuses/ Litter	MEAN %	30.1	19.5	26.1	22.7
	S.D.	13.83	13.80	16.81	14.44
		TOTAL RETARDATIONS			
Fetal Incidence	N	49	80[a]	77	75
	%	19	26	23	23
Litter Incidence	N	18	19	20	22
	%	95	86	87	96
Affected Fetuses/ Litter	MEAN %	19.7	25.5	23.6	22.9
	S.D.	10.45	16.57	14.95	12.33

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/02-7 Summary of All Fetal External Observations

		Control	50 ppm	250 ppm	750 ppm
Litters Evaluated	N	19	22	23	23
Fetuses Evaluated	N	259	302	331	321
Live	N	259	302	331	321
Dead	N	0	0	0	0
TOTAL MALFORMATIONS					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.3
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	4.3
Affected Fetuses/Litter	MEAN %	0.0	0.0	0.0	0.3
	S.D.	0.00	0.00	0.00	1.39
TOTAL VARIATIONS					
Fetal Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Affected Fetuses/Litter	MEAN %	0.0	0.0	0.0	0.0
	S.D.	0.00	0.00	0.00	0.00
UNCLASSIFIED OBSERVATIONS					
PLACENTAE FUSED					
Fetal Incidence	N	0	1	0	0
	%	0.0	0.3	0.0	0.0
Litter Incidence	N	0	1	0	0
	%	0.0	4.5	0.0	0.0
TOTAL FETAL EXTERNAL UNCLASSIFIED OBSERVATIONS					
Fetal Incidence	N	0	1	0	0
	%	0.0	0.3	0.0	0.0
Litter Incidence	N	0	1	0	0
	%	0.0	4.5	0.0	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/02-8 Summary of All Fetal Soft Tissue Observations

		Control	50 ppm	250 ppm	750 ppm
Litters Evaluated	N	19	22	23	23
Fetuses Evaluated	N	127	147	158	157
Live	N	127	147	158	157
Dead	N	0	0	0	0
MALFORMATIONS					
SITUS INVERSUS					
Fetal Incidence	N	1	0	0	0
	%	0.8	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	5.3	0.0	0.0	0.0
TOTAL FETAL SOFT TISSUE MALFORMATIONS					
Fetal Incidence	N	1	0	0	0
	%	0.8	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	5.3	0.0	0.0	0.0
VARIATIONS					
DILATED RENAL PELVIS					
Fetal Incidence	N	20	18	26	23
	%	16	12	16	15
Litter Incidence	N	10	10	14	14
	%	53	45	61	61
HYDROURETER					
Fetal Incidence	N	6	5	4	6
	%	4.7	3.4	2.5	3.8
Litter Incidence	N	3	5	4	5
	%	16	23	17	22
TOTAL FETAL SOFT TISSUE VARIATIONS					
Fetal Incidence	N	20	18	26	23
	%	16	12	16	15
Litter Incidence	N	10	10	14	14
	%	53	45	61	61

UNCLASSIFIED OBSERVATIONS

Fetal Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0
Litter Incidence	N	0	0	0	0
	%	0.0	0.0	0.0	0.0

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/02-9 Summary of All Classified Fetal Skeletal Observations

		Control	50 ppm	250 ppm	750 ppm
Litters Evaluated	N	19	22	23	23
Fetuses Evaluated	N	132	155	173	164
Live	N	132	155	173	164
Dead	N	0	0	0	0
		TOTAL MALFORMATIONS			
Fetal Incidence	N	9	8	2[a]	9
	%	6.8	5.2	1.2	5.5
Litter Incidence	N	7	8	2	7
	%	37	36	8.7	30
Affected Fetuses/ Litter	MEAN %	7.2	5.3	1.3	5.0
	S.D.	11.02	7.48	4.25	8.37
		TOTAL VARIATIONS			
Fetal Incidence	N	56	42[b]	58	52
	%	42	27	34	32
Litter Incidence	N	18	16	21	20
	%	95	73	91	87
Affected Fetuses/ Litter	MEAN %	44.6	26.0a	33.6	29.6
	S.D.	24.80	21.92	23.12	19.68
		TOTAL RETARDATIONS			
Fetal Incidence	N	49	80[a]	77	75
	%	37	52	45	46
Litter Incidence	N	18	19	20	22
	%	95	86	87	96
Affected Fetuses/ Litter	MEAN %	38.0	49.6	44.9	44.5
	S.D.	19.65	31.98	28.09	23.19

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Table A6.8.1/02-10 Summary of All Fetal Skeletal Observations

		Control	50 ppm	250 ppm	750 ppm
Litters Evaluated	N	19	22	23	23
Fetuses Evaluated	N	132	155	173	164
Live	N	132	155	173	164
Dead	N	0	0	0	0
MALFORMATIONS					
FETUS WITH MULTIPLE MALFORMATIONS (SKULL)					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.6
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	4.3
THORACIC VERTEBRAL BODY/BODIES BIPARTITE (ASYMMETRICAL)					
Fetal Incidence	N	1	1	1	1
	%	0.8	0.6	0.6	0.6
Litter Incidence	N	1	1	1	1
	%	5.3	4.5	4.3	4.3
THORACIC VERTEBRAL BODY/BODIES DUMBBELL-SHAPED (ASYMMETR.)					
Fetal Incidence	N	6	7	1[a]	7
	%	4.5	4.5	0.6	4.3
Litter Incidence	N	6	7	1[a]	6
	%	32	32	4.3	26
THORACIC VERTEBRAL BODY/BODIES DISLOCATED					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.6
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	4.3
DIFF. THORAC. VERT. BOD. AND/OR ARCHES SEVERELY MALFORMED					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.6
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	4.3
LUMBAR VERTEBRAE FUSED AND/OR OF IRREGULAR SHAPE					
Fetal Incidence	N	1	0	0	0

	%	0.8	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	5.3	0.0	0.0	0.0
STERNEBRA(E) BIPARTITE, OSSIFICATION CENTERS DISLOCATED					
Fetal Incidence	N	2	0	0	1
	%	1.5	0.0	0.0	0.6
Litter Incidence	N	2	0	0	1
	%	11	0.0	0.0	4.3
FUSED RIBS					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.6
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	4.3

		Control	50 ppm	250 ppm	750 ppm
Litters Evaluated	N	19	22	23	23
Fetuses Evaluated	N	132	155	173	164
Live	N	132	155	173	164
Dead	N	0	0	0	0
TOTAL FETAL SKELETAL MALFORMATIONS					
Fetal Incidence	N	9	8	2a	9
	%	6.8	5.2	1.2	5.5
Litter Incidence	N	7	8	2	7
	%	37	36	8.7	30
VARIATIONS					
ACCESSORY THORACIC VERTEBRA					
Fetal Incidence	N	1	0	0	0
	%	0.8	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	5.3	0.0	0.0	0.0
ACCESSORY LUMBAR VERTEBRA					
Fetal Incidence	N	3	0	0	0
	%	2.3	0.0	0.0	0.0
Litter Incidence	N	1	0	0	0
	%	5.3	0.0	0.0	0.0
STERNEBRA(E) OF IRREGULAR SHAPE					
Fetal Incidence	N	36	24[a]	39	32
	%	27	15	23	20
Litter Incidence	N	17	12[a]	19	17
	%	89	55	83	74
STERNEBRA(E) BIPARTITE					
Fetal Incidence	N	9	5	6	4
	%	6.8	3.2	3.5	2.4
Litter Incidence	N	6	4	5	3
	%	32	18	22	13
ACCESSORY STERNEBRA					
Fetal Incidence	N	0	0	0	1

	%	0.0	0.0	0.0	0.6
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	4.3
		13^{TH} RIB(S) SHORTENED			
Fetal Incidence	N	10	11	13	18
	%	7.6	7.1	7.5	11
Litter Incidence	N	5	6	9	8
	%	26	27	39	35
		RUDIMENTARY CERVICAL RIB(S)			
Fetal Incidence	N	0	4	4	3
	%	0.0	2.6	2.3	1.8
Litter Incidence	N	0	3	3	2
	%	0.0	14	13	8.7

		Control	50 ppm	250 ppm	750 ppm
Litters Evaluated	N	19	22	23	23
Fetuses Evaluated	N	132	155	173	164
Live	N	132	155	173	164
Dead	N	0	0	0	0
ACCESSORY 14TH RIB(S)					
Fetal Incidence	N	3	0	0	0
	%	2.3	0.0	0.0	0.0
Litter Incidence	N	2	0	0	0
	%	11	0.0	0.0	0.0
TOTAL FETAL SKELETAL VARIATIONS					
Fetal Incidence	N	56	42[b]	58	52
	%	42	27	34	32
Litter Incidence	N	18	16	21	20
	%	95	73	91	87
RETARDATIONS					
HYOID BONE INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	1	2	0
	%	0.0	0.6	1.2	0.0
Litter Incidence	N	0	1	2	0
	%	0.0	4.5	8.7	0.0
HYOID BONE NOT OSSIFIED					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.6
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	4.3
THORACIC VERTEBRAL BODY/BODIES DUMBBELL-SHAPED (SYMMETR.)					
Fetal Incidence	N	12	13	12	16
	%	9.1	8.4	6.9	9.8
Litter Incidence	N	9	9	9	9
	%	47	41	39	39
THORACIC VERTEBRAL BODY/BODIES INCOMPLETELY OSSIFIED					
Fetal Incidence	N	0	0	0	2

	%	0.0	0.0	0.0	1.2
Litter Incidence	N	0	0	0	2
	%	0.0	0.0	0.0	8.7
THORACIC VERTEBRAL BODY/BODIES-ONLY ONE OSSIFICATION CENTER					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.6
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	4.3
THORACIC VERTEBRAL BODY/BODIES BIPARTITE (SYMMETRICAL)					
Fetal Incidence	N	1	2	0	0
	%	0.8	1.3	0.0	0.0
Litter Incidence	N	1	2	0	0
	%	5.3	9.1	0.0	0.0

		Control	50 ppm	250 ppm	750 ppm
Litters Evaluated	N	19	22	23	23
Fetuses Evaluated	N	132	155	173	164
Live	N	132	155	173	164
Dead	N	0	0	0	0
THORACIC VERTEBRAL BODY/BODIES NOT OSSIFIED					
Fetal Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	0.6
Litter Incidence	N	0	0	0	1
	%	0.0	0.0	0.0	4.3
STERNEBRA(E) NOT OSSIFIED					
Fetal Incidence	N	9	19	21	16
	%	6.8	12	12	9.8
Litter Incidence	N	6	12	11	12
	%	32	55	48	52
STERNEBRA(E) INCOMPLETELY OSSIFIED OR REDUCED IN SIZE					
Fetal Incidence	N	15	39b	33	33
	%	11	25	19	20
Litter Incidence	N	8	16	11	19b
	%	42	73	48	83
STERNEBRA(E) – ONLY ONE OSSIFICATION CENTER					
Fetal Incidence	N	21	23	24	26
	%	16	15	14	16
Litter Incidence	N	12	14	15	14
	%	63	64	65	61
TOTAL FETAL SKELETAL RETARDATIONS					
Fetal Incidence	N	49	80a	77	75
	%	37	52	45	46
Litter Incidence	N	18	19	20	22
	%	95	86	87	96

SIGNIFICANTLY DIFFERENT FROM CONTROL: a = P<0.05; b = P<0.01.

Section A6.8.2 Annex Point IIA, VI.6.8.2 IUCLID 5.8.1/01	A6.8.2 Multigeneration Reproduction Toxicity Study Two –Generation Drinking Water Reproduction Study in Rats	
	1 REFERENCE (A6.8.2/01)	Official use only
1.1 Reference	[redacted] (1994) Glutaraldehyde: Two-generation reproduction study in the drinking water of [redacted] rats, [redacted], Unpublished, 24 March 1994	
1.2 Data protection	Yes	
1.2.1 Data owner	The Dow Chemical Company	
1.2.2 Companies with letter of access	[redacted]	
1.2.3 Criteria for data protection	Data on an existing active substance for first entry to Annex I authorisation	
	2 GUIDELINES AND QUALITY ASSURANCE	
2.1 Guideline study	Yes US EPA OPP 83-4	
GLP	Yes	
Deviations	Yes Verified data collected on heat sensitive paper and calculator tapes were kept, original data was discarded.	
	3 MATERIALS AND METHODS	
3.1 Test material	Glutaraldehyde [redacted]	
3.1.1 Lot/Batch number	[redacted]	
3.1.2 Specification	Not reported	
3.1.2.1 Description	Clear, colourless, non-viscous liquid	
3.1.2.2 Purity	[redacted]	
3.1.2.3 Stability	Assumed to be stable under normal storage conditions.	X
3.2 Test Animals		
3.2.1 Species	Rat	
3.2.2 Strain	[redacted]	
3.2.3 Source	[redacted]	
3.2.4 Sex	Male and female	
3.2.5 Age/weight at study initiation	7 weeks of age Weight range (males) 236.5-237 grams Weight range (females) 165-166.7 grams	
3.2.6 Number of animals per group	28/sex/dose	
3.2.7 Husbandry	**Table A6.8.2/01-1**	
3.2.8 Mating	After the 10-week prebreeding exposure period, animals were randomly mated (established via computer randomization protocol) 1:1 within their dose group. They were paired for 7 days. After the 7 days, unmated females and unmated males were repaired for an additional 7 days (and repeated a third time if still unsuccessful for a maximum mating period of 21 days). The observation of a dropped vaginal copulation plug was used as evidence of successful mating (checked	

Section A6.8.2 Annex Point IIA, VI.6.8.2 IUCLID 5.8.1/01	A6.8.2 Multigeneration Reproduction Toxicity Study Two –Generation Drinking Water Reproduction Study in Rats	
	twice daily). Vaginal smears were made once daily. The day on which a copulation plug or sperm-positive smear was found was considered Gestation Day (GD) 0. Once positive signs of mating were seen, the pair was singly housed.	
3.2.9 Duration of mating	Up to 21 days depending on pregnancy status	
3.2.10 Deviations from standard protocol	The water content in three test samples was inadvertently not analyzed.	
3.2.11 Control animals	Yes, concurrent vehicle	
3.3 Administration/ Exposure	Oral/drinking water	
3.3.1 Animal assignment to dosage groups	Animals were weighed on the first day of treatment, and assigned randomly to groups based on body weights, 28 per sex per dose.	
3.3.2 Duration of exposure before mating	10 weeks	
3.3.3 Duration of exposure in general F_0, F_1, F_2 males, females	F_0 Parental breeding period of 10 weeks F_0 Mating period of 21 days F_0-F_1 Gestation and lactation period through post-partum day 21 F_1 Parental breeding period of 10 weeks F_1-F_2 Gestation and lactation period through post-partum day 21	
3.3.4 Type	Oral, drinking water	
3.3.5 Concentration	Mixed as received (51% glutaraldehyde in water) into the drinking water.	
3.3.6 Vehicle	Deionized Water	
3.3.7 Concentration in vehicle	50, 250 and 1000 ppm	
3.3.8 Total volume applied	50 ppm group (4.3-6.7 mg/kg/day) 250 ppm group (19.5-28.3 mg/kg/day) 1000 ppm group (69.1-98.4 mg/kg/day)	X
3.3.9 Dose Confirmation	***Homogeneity*** was established at 50 ppm and 1000 ppm. ***Stability*** was established prior to study start at 50, 1000 ppm ***Concentrations*** were verified by gas chromatography for all dose levels for the first 4 weeks of the study, and every 4 weeks thereafter.	
3.3.10 Controls	Yes, concurrent vehicle (tap or deionized water)	
3.4 Examinations		
3.4.1 Clinical signs	Animals were observed twice daily for mortality and a detailed clinical observation was recorded weekly during the pre-breeding period. During mating, gestation, and lactation, animals were checked twice daily for mortality. Detailed observations were recorded weekly.	
3.4.2 Body weight	Mated females were weighed on GD 0, 6, 15, and 20. Dams with litters were weighed on Lactation Day (LD) 0, 7, 14, 21.	
3.4.3 Food/water consumption	Food consumption and water consumption were recorded weekly during the pre-breeding period. Water and food consumption was measured for females at 3-4 day intervals throughout gestation. Water consumption was measured on LD 0-14, and food consumption	

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		was measured LD 0-21.	
3.4.4	Oestrus cycle	Not evaluated	
3.4.5	Sperm parameters	Not evaluated	
3.4.6	Offspring	All pups in both generations were examined as soon as possible on the day of birth to determine the numbers of viable and stillborn males and females. Litters were evaluated twice daily for mortality. On day 4, the litter sizes were adjusted (by random elimination) to yield 4 males and 4 females. Culled pups were examined externally only. Pup survival indices were calculated at LD 0, 4, 7, 14, and 21, and one week post-weaning on day 28. Pups were examined for physical abnormalities at birth and throughout lactation. All spontaneous deaths were necropsied if possible to determine the cause of death. One week after weaning, parents of the F_2 generation were chosen. Ten pups per sex per dose were examined during necropsy for gross external abnormalities. The vagina, uterus, ovaries, testes with epididymis, seminal vesicles, prostate, and other tissues with gross lesions were evaluated. All pups dying spontaneously were examined, and a complete internal examination was conducted on all pups with external abnormalities.	
3.4.7	Organ weights P1, P2 and F1and F2	Not evaluated	
3.4.8	Histopathology P1 and P2	Adult F_0 males and females, and F_1 females were sacrificed after weaning their litters. F_1 males were sacrificed after delivery of their litters. Necropsies of all animals included evaluations of the brain, spinal cord, thoracic, abdominal, and pelvic cavities, viscera including reproductive organs, and cervical tissue and organs. Histopathology on the same organs plus vagina, uterus, ovaries, testes, epididymis, seminal vesicles, prostate, and any tissues with gross lesions was evaluated for control and high dose animals only. Testes and epididymis were evaluated in the low and mid dose groups for males which did not sire. A complete necropsy with histopathology was conducted on any parental animal sacrificed in a moribund condition or dying prior to scheduled sacrifice. Fixed uteri from parental females failing to produce a litter were stained for confirmation of pregnancy status.	X
3.4.9	Histopathology F1 not selected for mating, F2	Same as 3.4.8	X
3.5	Statistics	The unit of comparison was the male, pregnant dam, or litter. The data for quantitative continuous variables were inter-compared for the 3 dose groups and the control group by use of Levene's test for equality of variances, ANOVA, and t-tests. The t-tests were used when the F value from the ANOVA was significant. When Levene's test indicated similar variances, and the ANOVA was significant, a pooled t-test was used for pair-wise comparison. When Levene's test indicated heterogeneous variances, all groups were compared by an ANOVA for unequal variances followed, when appropriate, by a separate variance t-test for pair-wise comparisons. Non-parametric data were statistically evaluated using the Kruskal-Wallis test followed by the Mann-Whitney U test. Incidence data were compared using a Fischer's Exact Test. The critical level of	

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	significance for all tests was $p<0.05$ (two-tailed).	
	4 RESULTS AND DISCUSSION	
4.1 Effects		
4.1.1 Mortality	There was no treatment-related mortality or clinical signs of toxicity observed during any phase of the study.	
4.1.2 Clinical Observations	With the exception of decreased food and water consumption, no in-life observations were noted in any dose level, in either the parents or offspring. There were no overt signs of systemic toxicity.	
4.1.3 Body Weights (Adults)	**F_0 Adults** Mean body weights in males were unaffected by treatment. Body weight gains in males were depressed in the 250 ppm and 1000 ppm dose groups for the first week of study, and sporadic decreases were noted in later weeks, but not in a dose-related manner. There was a significant, treatment-related decrease in body weights of high-dose (1000 ppm) females; sporadic changes in the low and mid dose groups were not attributed to treatment and did not occur in a dose-related manner. There was no effect on gestation body weights at any dose. A decrease in lactation body weights was noted in high dose animals. **F_1 Adults** Body weight means, and occasionally weight gains, were decreased slightly in the 1000 ppm males; overall, cumulative weight gains were comparable to control animals over the 10-week pre-breeding period. Sporadic decreases in females were noted, but did not occur in a treatment-related pattern and were therefore not considered related to treatment. Gestation and lactation body weights were unaffected by treatment.	
4.1.4 Food & Water Consumption	**F_0 Pre-Breeding** Water consumption was decreased in the 250 and 1000 ppm groups. There was additionally a slight decrease in water consumption for females from the 50ppm group weeks 2-6. Food consumption was reduced for males and females at 1000ppm, though the increase was not consistent through the pre-breeding phase. **F_0 Gestation & Lactation** Decreases in water consumption were noted in the 250 and 1000ppm groups (20-35%), and transiently in the 50ppm group during the first week of gestation. A 5% decrease in food consumption in females from the high dose was noted in late gestation. There were no effects on food consumption during lactation. **F_1 Pre-Breeding** Similar observations to the F_0 pre-breeding were made for food and water consumption as well as body weights. **F_1 Gestation & Lactation** Water consumption continued to be decreased in the mid and high dose groups for gestation and lactation. A slight decrease in water consumption was also noted in the low dose during gestation. A slight decrease in food consumption was noted for the high dose group late in gestation. No effects on lactational food consumption were noted in any group.	

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4.1.5 Test Material Intake	50 ppm group (4.3-6.7 mg/kg/day) 250 ppm group (19.5-28.3 mg/kg/day) 1000 ppm group (69.1-98.4 mg/kg/day)	X
4.1.6 Organ Weights	Not applicable	
4.1.7 Gross Pathology	There were no treatment-related findings from necropsies of F_1 males and females and F_2 weanlings.	X
4.1.8 Histopathology	There were no treatment-related findings from histopathologic examinations of F_1 males and females and F_2 weanlings.	X
4.1.9 Sperm Parameters	Not applicable	
4.1.10 Estrous Cyclicity	Not applicable	
4.1.11 Reproductive Indices, Pup Survival and Sex Ratio	**F_0 Mating and Reproductive Effects at F_1 Breed (Table A6.8.2/01-2)** Mating, fertility, and gestational indices, including gestational length, were unaffected by treatment. **F_1 Mating and Reproductive Effects at F_2 Breed (Table A6.8.2/01-7)** Mating, fertility, and gestational indices, including gestational length, were unaffected by treatment.	
4.1.12 Litter Size & Sex Ratios	**F_1 Offspring (Table A6.8.2/01-3, 5)** There were no treatment-related effects on litter size or sex ratios. **F_2 Offspring (Table A6.8.2/01-8,10)** There were no treatment-related effects on litter size or sex ratios.	
4.1.13 Pup Weights	**F_1 Offspring (Table A6.8.2/01-6)** There were no treatment-related changes in pup body weights through LD 14. From weaning on days 21-28, pup weights were decreased in the 1000 ppm dose group with corresponding decreases in weight gain. Increased pup weights were noted in the 250 ppm group, considered to be related to smaller average litter size in this dose group. **F_2 Offspring (Table A6.8.2/01-11)** There were no treatment-related changes in pup body weights through LD 14. From weaning on days 21-28, pup weights were decreased in the 1000 ppm dose group with corresponding decreases in weight gain.	
4.1.14 Pubertal Onset	Not applicable	
4.1.15 Pup Survival	**F_1 Offspring (Table A6.8.2/01-4)** There were no treatment-related effects on the number of liveborn pups or on F_1 pup viability and survival. Statistical significance associated with the 4-day survival index in the 250 ppm group was not considered to be treatment-related and was likely related to the smaller average litter size of the mid dose group. **F_2 Offspring (Table A6.8.2/01-9)** There were no effects of treatment on the number of liveborn pups or on F_2 pup viability and survival.	
4.2 Other	Not applicable	
	5 APPLICANT'S SUMMARY AND CONCLUSION	
5.1 Materials and methods	Animals were weighed on the first day of treatment, and assigned randomly to groups based on body weights, 28 per sex per dose. The	

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	test material was administered via the drinking water, and the solutions were analyzed for glutaraldehyde concentration by GC. Animals were examined twice daily for mortality. Weekly detailed observations were recorded. Prebreeding body weights were taken weekly, gestation body weights on gestation days 0, 6, 15, 20, and lactation body weights on days 0, 7, 14, and 21 post-partum. Food consumption, and water consumption were recorded weekly. After the 10-week prebreeding exposure period, animals were randomly mated (established via computer randomization protocol) 1:1 within their dose group. They were paired for 7 days. After the 7 days, unmated females and unmated males were repaired for an additional 7 days (and repeated a third time if still unsuccessful). The observation of a dropped vaginal copulation plug was used as evidence of successful mating (checked twice daily). Vaginal smears were made once daily. The day on which a couplation plug or sperm-positive smear was found was considered Gestation Day (GD) 0. Once positive signs of mating were seen, the pair was singly housed. On GD 20, females were transferred to a shoe box cage, and checked three times daily for evidence of littering. Litters were weaned on LD 21, and litters cohoused for an additional 7 days. Twenty-eight male and 28 female rats were randomly chosen as parents of the F_2 generation, and were dosed via the drinking water for 10 weeks prior to their breeding period. An additional subset of pups were examined at necropsy. Non-selected F_1 pups were examined externally only. Parents of the F_2 generation were mated as above, and sister-brother matings were avoided. All pups in both generations were examined as soon as possible on the day of birth to determine the numbers of viable and stillborn males and females. On day 4, the litter sizes were adjusted (by random elimination) to yield 4 males and 4 females. Culled pups were examined externally only. Pup survival indices were calculated at LD 0, 4, 7, 14, and 21, and one week post-weaning on day 28. Pups were examined for physical abnormalities at birth and throughout lactation. All spontaneous deaths were necropsied if possible to determine the cause of death. One week after weaning, parents of the F_2 generation were chosen. Ten pups per sex per dose were examined during necropsy for gross external abnormalities. The vagina, uterus, ovaries, testes with epididymides, seminal vesicles, prostate, and other tissues with gross lesions were evaluated. All pups dying spontaneously were examined, and a complete internal examination was conducted on all pups with external abnormalities. Adult F_0 males and females, and F_1 females were sacrificed after weaning their litters. F_1 males were sacrificed after delivery of their litters. Necropsies of all animals included evaluations of major tissues and organs, grossly and histopathologically. Testes and epididymes were evaluated in the low and mid dose groups for males which did not sire. Fixed uteri from parental females failing to produce a litter were stained for confirmation of pregnancy status.	

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	Reproductive Indices Measured/Calculated: • Mating Index (females) • Mating Index (males) • Fecundity Index (females) • Fecundity Index (males) • Fertility Index (females) • Fertility Index (males) • Gestational Index • Live Birth Index • 4-Day Survival Index • 7-Day Survival Index • 14-Day Survival Index • 21-Day Survival Index • 28-Day Survival Index • Lactation Index	
5.2 Results and discussion	***F_0 Pre-Breeding*** There was no treatment-related mortality or clinical signs of toxicity observed during the prebreeding period (10 weeks). Water consumption was decreased in the 250 and 1000 ppm groups. There was additionally a slight decrease in water consumption for females from the 50 ppm group weeks 2-6. Mean body weight gain was slightly reduced for males at the mid and high dose and females at the high dose sporadically. Food consumption was reduced for males and females at 1000 ppm, though the increase was not consistent through the prebreeding phase. ***F_0 Gestation & Lactation*** Decreases in water consumption were noted in the 250 and 1000 ppm groups, and transiently in the 50 ppm group during the first week of gestation. There were no treatment-related effects on body weights or weight gains in spite of a 5% decrease in food consumption in females from the high dose in late gestation. There were no effects on food consumption during lactation. ***F_0 Littering*** There were no effects on the mating performance or fertility of F_0 adults, nor any effects on the duration of gestation, litter size, litter viability, or pup survival in F_1 offspring. There was no effect on F_1 pup body weights through day 14. Day 21 and 28 litter weights were reduced in the high dose group. There were no treatment-related lesions observed at the necropsy evaluations of a random group of F_1 weanlings, or those that died during lactation. ***F_1 Pre-Breeding*** Similar observations to the F_0 pre-breeding were made for food and water consumption as well as body weights. **F_1 Gestation & Lactation** There were no effects on breeding parameters, and no effects on gestation and lactational body weights. Water consumption continued to be decreased in the mid and high dose groups for gestation and lactation. A slight decrease in water consumption was also noted in the low dose during gestation. A slight decrease in food consumption was	

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	noted for the high dose group late in gestation. No effects on lactational food consumption were noted in any group. **F_1 Littering** There were no effects on litter size, litter viability, or survival in F_2 offspring. There was no effect of treatment on the body weights of F_2 pups through day 7. F_2 litter weights were reduced in the high dose group at days 14, 21, and 28. No treatment-related lesions were observed at necropsy of randomly selected weanlings or pups that died during lactation. There were likewise no treatment-related findings at necropsy of F_0 and F_1 parents, and no findings were observed upon microscopic evaluation of tissues from the control and high-dose F_0 and F1 parents	
5.3 Conclusion	Treatment of rats with glutaraldehyde in the drinking water resulted in aversion to the water. Effects on parental body weights and food consumption, and effects on offspring body weight were observed at 1000 ppm. Minimal parental effects were noted at 250 ppm. There were no adverse effects on reproductive performance at any dose level.	
5.3.1 LOAEL		
5.3.1.1 Parental Toxicity	1000 ppm	
5.3.1.2 F_1 Offspring	1000 ppm	
5.3.1.3 F_2 Offspring	1000 ppm	
5.3.2 NOAEL		
5.3.2.1 Parental Toxicity	250 ppm (based on decreased food, water consumption)	
5.3.2.2 F_1 Offspring	250 ppm (based on decreased food, water consumption)	
5.3.2.3 F_2 Offspring	250 ppm (based on decreased food, water consumption)	
5.3.3 NOEL		
5.3.4 Parental Toxicity	50 ppm	
5.3.5 Offspring Toxicity	250 ppm	
5.3.6 Reproductive Effects	>1000 ppm	
5.3.7 Reliability	1	
5.3.8 Deficiencies	No	
	Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE	
Date	November 9th, 2010	
Materials and Methods	3.1.2.3 Stability. Stability was analytically confirmed. 3.3.8 Total volume applied. The test substance intake is given incorrectly. See comment to 4.1.5 below. 3.4.8 Histopathology P1 and P2. Histopathology was only performed to the organs specifically mentioned starting on line 5, i.e. the sentence should be corrected as follows: "Histopathology on ~~the same organs plus~~ vagina, uterus…" 3.4.9 Histopathology - F1 not selected for mating, F2. Complete necropsy was performed to only 10 pups per sex from each dose group of both F1 and F2.	
Results and discussion	4.1.5 Test material intake. The test substance intake is given incorrectly. The	

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	correct values are as follows (averages, mg/kg bw/day): • F0 ♂: 4.3, 19.5, 69.1 • F0 ♀: 6.7, 28.3, 98.4 (only prebreed period given) • F1♂: 4.5, 21, 71.1 • F1♀: 6.7, 29.6, 99.6 (only prebreed period given) Glutaraldehyde intake can be calculated from these values by multiplying with 0.51. 4.1.7 Gross Pathology. Additionally, there were no effects in F0 males or females. 4.1.8 Histopathology. Additionally, there were no effects in F0 males or females.	
Conclusion	There was no evidence of effects on any reproductive parameters at doses below maternal toxicity.	
Reliability	2	
Acceptability	Acceptable	
Remarks	The detailed assessment of the study is difficult because no summary tables were provided for the necropsy results. The study is in accordance with the OECD guideline 416 (1983) valid at the time of the study. The following deficiencies are noted with regard to the updated guideline of 2001: • No sperm parameters were studied. • Implantation sites were not studied (pre- and postimplantation losses). • Organ weights were not measured. Please note that the tabulated results in the tables below have not been checked in detail by the RMS.	
	COMMENTS FROM ...	
Date	*Give date of comments submitted*	
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*	
Results and discussion	*Discuss if deviating from view of rapporteur member state*	
Conclusion	*Discuss if deviating from view of rapporteur member state*	
Reliability	*Discuss if deviating from view of rapporteur member state*	
Acceptability	*Discuss if deviating from view of rapporteur member state*	
Remarks		

Table A6.8.2/01-1 Animal Husbandry

Parameter	Condition
Species	Rat
Strain	[redacted]
Supplier	[redacted]
Age / Weight at Arrival	28 days of age Weight range (males and females) 65-80 grams
Age / Weight at Study Initiation	7 weeks of age Weight range (males) 236.5-237 grams Weight range (females) 165-166.7 grams
Veterinary Examination	Within 2 days of receipt, examinations of all animals were conducted by a clinical veterinarian. Representative animals were subjected to a pretest health screen (examination for intestinal parasites, histologic examinations of select tissues, clinical pathology evaluations, and serum viral antibody analyses).
Animal Identification	Cage tags and tail tattoos
Housing	Housed 2 per cage for 7 days of acclimation, and singly thereafter except during cohabitation (mating) and lactation. Stainless steel hanging cages (22.5x15.5x18 cm) until gestation day 20. On GD 20, rats were placed in polypropylene shoe box caging with bedding for parturition until weaning.
Feed and Water	Purina Mills Rodent Chow® #5002 *ad libitum* Tap water or distilled water *ad libitum*
Temperature	66-77 °F, recorded continuously
Relative Humidity	40-70% recorded continuously
Photocycle (hours light/dark)	12 hours light / 12 hours dark
Acclimation	3 weeks total acclimation
Assignment to Study Groups	Random assignment using a stratified randomization procedure based on body weight

Table A6.8.2/01-2 Reproductive Parameters for F_0 Parents at F_1 Breed

	0 ppm	50 ppm	250 ppm	1000 ppm
No. F0 pairs at study start	28	28	28	28
No. F0 pairs at start of F1 breed	28	28	28	28
No. males impregnating females[a]	28	28	28	28
No. plug/sperm-positive females	28	28	28	28
No. pregnant[b]	28	25	26	26
No. males siring litter	28	25	26	26
No. live litters on postnatal day 0	28	25[c]	26	26
Gestational length in days (mean ± S.D)	21.9 ± 0.5	22.0 ± 0.5	22.2 ± 0.4	22.0 ± 0.3
INDICES[d]				
Mating Index (females)	100.0	100.0	100.0	100.0
Mating Index (males)	100.0	100.0	100.0	100.0
Fecundity index (females)	100.0	89.3	92.9	92.9
Fecundity index (males)	100.0	89.3	92.9	92.9
Fertility index (females)	100.0	89.3	92.9	92.9
Fertility index (males)	100.0	89.3	92.9	92.9
Gestational index	100.0	100.0	100.0	100.0

[a] Defined as the number of males producing plug- or sperm-positive females.
[b] Determined by delivery of litters/uterine staining.
[c] One female (#11663) died during delivery; litter sacrificed on day 0.
[d] The indices are defined in the text.

Table A6.8.2/01-3 Litter Viability - F_1 Pups

	0 ppm	50 ppm	250 ppm	1000 ppm
Lactation Day 0				
Total Born	414	381	340	371
Total Born Alive	414	378	338	366
# Stillborn	0	3	2	5
Lactation Day 4 (pre-cull)				
# Alive	394	351	333	348
# Dead (Days 0- 4)	20	27	5	18
Lactation Day 4 (post-cull)				
# Alive	224	192	204	208
Lactation Day 7				
# Alive	224	192	204	207
# Dead (Day 4 post-cull - 7)	0	0	0	1
Lactation Day 14				
# Alive	224	192	202	207
# Dead (Day 7- 14)	0	0	2	0
# Dead (Day 4 post-cull - 14)	0	0	2	1
Lactation Day 21[a]				
# Alive	222	192	202	207
# Dead (Days 14 - 21)	2	0	0	0
# Dead (Day 4 post-cull - 21)	2	0	2	1
Lactation Day 28				
# Alive	222	192	202	206
# Dead (Days 21 - 28)	0	0	0	1
# Dead (Day 4 post-cull - 28)	2	0	2	2

[a] Litters were weaned on lactational day 21.

Table A6.8.2/01-4 Pup Survival Indices- F1 Pups

	0 ppm	50 ppm	250 ppm	1000 ppm
LIVE BIRTH INDEX				
MEAN	100.0	99.2	99.4	98.8
S.D.	0.00	2.15	2.26	3.63
N	28	25	26	26
4-DAY SURVIVAL INDEX				
MEAN	95.3	93.6	98.7**	95.4
S.D.	6.13	19.98	4.23	6.89
N	28	25	26	26
7-DAY SURVIVAL INDEX				
MEAN	100.0	100.0	100.0	99.5
S.D.	0.00	0.00	0.00	2.45
N	28	24	26	26
14-DAY SURVIVAL INDEX				
MEAN	100.0	100.0	99.0	100.0
S.D.	0.00	0.00	3.40	0.00
N	28	24	26	26
21-DAY SURVIVAL INDEX				
MEAN	99.1	100.0	100.0	100.0
S.D.	3.28	0.00	0.00	0.00
N	28	24	26	26
28-DAY SURVIVAL INDEX				
MEAN	100.0	100.0	100.0	99.5
S.D.	0.00	0.00	0.00	2.45
N	28	24	26	26
LACTATIONAL INDEX[a]				
MEAN	99.1	100.0	99.0	99.5
S.D.	3.28	0.00	3.40	2.45
N	28	24	26	26

** Significantly different from control group ($p < .01$)
The equations used for calculating pup survival indices are recorded in the protocol.
[a] Litters were weaned on lactational day 21.

Table A6.8.2/01-5 Litter Size and Sex Ratio (% males) - F1 Pups

	0 ppm	50 ppm	250 ppm	1000 ppm
LACTATIONAL DAY 0				
TOTAL BORN/LITTER				
MEAN	14.8	15.2	13.1**	14.3
S.D.	1.52	1.54	2.45	2.49
N	28	25	26	26
TOTAL BORN ALIVE/LITTER				
MEAN	14.8	15.1	13.0**	14.1
S.D.	1.52	1.54	2.50	2.35
N	28	25	26	26
SEX RATIO				
MEAN	50.1	53.2	49.5	51.9
S.D.	15.15	13.78	17.67	14.29
N	28	25	26	26
LACTATIONAL DAY 4 (PRECULL)				
LITTER SIZE				
MEAN	14.1	14.6	12.8*	13.4
S.D.	1.54	1.47	2.42	2.25
N	28	24	26	26
SEX RATIO				
MEAN	50.4	52.8	49.9	52.5
S.D.	15.00	14.44	18.29	13.84
N	28	24	26	26
LACTATIONAL DAY 4 (POSTCULL)				
LITTER SIZE				
MEAN	8.0	8.0	7.8	8.0
S.D.	0.00	0.00	0.78	0.00
N	28	24	26	26
SEX RATIO				
MEAN	50.9	49.5	53.4	52.4
S.D.	7.55	4.48	9.05	7.09
N	28	24	26	26
LACTATIONAL DAY 7				
LITTER SIZE				
MEAN	8.0	8.0	7.8	8.0
S.D.	0.00	0.00	0.78	0.20
N	28	24	26	26
SEX RATIO				
MEAN	50.9	49.5	53.4	52.7
S.D.	7.55	4.48	9.05	7.13
N	28	24	26	26
LACTATIONAL DAY 14				
LITTER SIZE				
MEAN	8.0	8.0	7.8	8.0
S.D.	0.00	0.00	0.82	0.20
N	28	24	26	26
SEX RATIO				
MEAN	50.9	49.5	52.8	52.7
S.D.	7.55	4.48	9.46	7.13
N	28	24	26	26

	0 ppm	50 ppm	250 ppm	1000 ppm
LACTATIONAL DAY 21[a]				
LITTER SIZE				
MEAN	7.9	8.0	7.8	8.0
S.D.	0.26	0.00	0.82	0.20
N	28	24	26	26
SEX RATIO				
MEAN	50.9	49.5	52.8	52.7
S.D.	7.80	4.48	9.46	7.13
N	28	24	26	26
LACTATIONAL DAY 28				
LITTER SIZE				
MEAN	7.9	8.0	7.8	7.9
S.D.	0.26	0.00	0.82	0.27
N	28	24	26	26
SEX RATIO				
MEAN	50.9	49.5	52.8	53.0
S.D.	7.80	4.48	9.46	7.16
N	28	24	26	26

[a] Litters were weaned on lactational day 21.
* Significantly different from control group ($p < .05$)
** Significantly different from control group ($p < .01$)

Table A6.8.1/01-6 Body Weights Per Litter- F1 Pups

	0 ppm	50 ppm	250 ppm	1000 ppm
LACTATIONAL DAY 1				
ENTIRE LITTER				
MEAN	6.22	6.50	7.02**	6.50
S.D.	0.728	0.584	0.723	0.617
N	28	24	26	26
MALE PUPS				
MEAN	6.40	6.69	7.20**	6.67
S.D.	0.829	0.622	0.689	0.633
N	28	24	26	26
FEMALE PUPS				
MEAN	6.05	6.31	6.88**	6.34
S.D.	0.707	0.580	0.766	0.614
N	28	24	26	26
LACTATIONAL DAY 4 (PRE CULL)				
ENTIRE LITTER				
MEAN	8.98	9.36	10.03**	9.30
S.D.	0.936	0.773	1.206	1.106
N	28	24	26	26
MALE PUPS				
MEAN	9.20	9.62	10.26**	9.47
S.D.	0.944	0.799	1.140	1.127
N	28	24	26	26
FEMALE PUPS				
MEAN	8.77	9.08	9.84**	9.10
S.D.	1.014	0.789	1.325	1.093
N	28	24	26	26
LACTATIONAL DAY 4 (POST CULL)				
ENTIRE LITTER				
MEAN	9.05	9.39	10.09**	9.30
S.D.	0.942	0.843	1.206	1.130
N	28	24	26	26
MALE PUPS				
MEAN	9.26	9.66	10.32**	9.51
S.D.	0.934	0.849	1.113	1.111
N	28	24	26	26
FEMALE PUPS				
MEAN	8.84	9.12	9.84**	9.07
S.D.	1.021	0.891	1.357	1.139
N	28	24	26	26
LACTATIONAL DAY 7				
ENTIRE LITTER				
MEAN	14.79	15.27	15.91**	15.04
S.D.	1.368	1.273	1.438	1.420
N	28	24	26	26
MALE PUPS				
MEAN	15.09	15.69	16.24**	15.33
S.D.	1.368	1.272	1.423	1.455
N	28	24	26	26
FEMALE PUPS				
MEAN	14.49	14.86	15.55	14.72
S.D.	1.454	1.368	1.569	1.409
N	28	24	26	26

	0 ppm	50 ppm	250 ppm	1000 ppm
LACTATIONAL DAY 14				
ENTIRE LITTER				
MEAN	31.75	33.16	33.13	32.12
S.D.	2.474	2.190	2.705	2.581
N	28	24	26	26
MALE PUPS				
MEAN	32.27	33.86	33.77	32.64
S.D.	2.781	2.095	2.684	2.554
N	28	24	26	26
FEMALE PUPS				
MEAN	31.25	32.48	32.48	31.52
S.D.	2.358	2.412	2.907	2.609
N	28	24	26	26
LACTATIONAL DAY 21				
ENTIRE LITTER				
MEAN	50.60	51.29	51.50	48.25*
S.D.	3.645	3.771	3.874	3.754
N	28	24	26	26
MALE PUPS				
MEAN	51.53	52.55	52.61	49.16*
S.D.	3.807	3.665	4.038	3.605
N	28	24	26	26
FEMALE PUPS				
MEAN	49.72	50.03	50.29	47.20*
S.D.	3.775	4.132	3.914	3.930
N	28	24	26	26
LACTATIONAL DAY 28				
ENTIRE LITTER				
MEAN	84.75	85.40	87.91	76.39**
S.D.	7.195	7.163	7.231	8.817
N	28	24	26	26
MALE PUPS				
MEAN	88.24	89.65	91.83	79.72**
S.D.	7.370	7.311	7.278	9.554
N	28	24	26	26
FEMALE PUPS				
MEAN	81.23	81.24	83.49	72.60**
S.D.	7.285	7.240	6.889	8.172
N	28	24	26	26

* Significantly different from control group ($p < .05$)
** Significantly different from control group ($p < .01$)

Table A6.8.2/01-7 Reproductive Parameters for F_1 Parents at F_2 Breed

	0 ppm	50 ppm	250 ppm	1000 ppm
No. F1 pairs at study start	28	28	28	28
No. F1 pairs at start of F2 breed	28	28	28	28
No. males impregnating females[a]	26	26	26	27
No. plug/sperm-positive females	26	26	27	28
No. pregnant[b]	25	22	27	28
No. males siring litter	25	23	26	27
No. live litters on postnatal day 0	25	22	27	28
Gestational length in days (mean ± S.D)	22.1 ± 0.4	22.0 ± 0.4	22.0 ± 0.4	22.0 ± 0.5
INDICES[c]				
Mating Index (females)	92.9	92.9	96.4	100.0
Mating Index (males)	92.9	92.9	92.9	96.4
Fecundity index (females)	96.2	84.6	100.0	100.0
Fecundity index (males)	96.2	88.5	100.0	100.0
Fertility index (females)	89.3	78.6	96.4	100.0
Fertility index (males)	89.3	82.1	92.9	96.4
Gestational index	100.0	100.0	100.0	100.0

[a] Defined as the number of males producing plug- or sperm-positive females.
[b] Determined by delivery of litters/uterine staining.
[c] The indices are defined in the text.

Table A6.8.2/01-8 Litter Viability- F2

	0 ppm	50 ppm	250 ppm	1000 ppm
Lactation Day 0				
Total Born	303	308	339	381
Total Born Alive	299	306	337	378
# Stillborn	4	2	2	3
Lactation Day 4 (pre-cull)				
# Alive	295	302	331	368
# Dead (Days 0- 4)	4	4	6	10
Lactation Day 4 (post-cull)				
# Alive	192	176	204	224
Lactation Day 7				
# Alive	192	176	204	224
# Dead (Day 4 post-cull - 7)	0	0	0	0
Lactation Day 14				
# Alive	190	176	203	223
# Dead (Day 7- 14)	2	0	1	1
# Dead (Day 4 post-cull - 14)	2	0	1	1
Lactation Day 21[a]				
# Alive	190	176	202	223
# Dead (Days 14 - 21)	0	0	1	0
# Dead (Day 4 post-cull - 21)	2	0	2	1
Lactation Day 28				
# Alive	190	176	202	223
# Dead (Days 21 - 28)	0	0	0	0
# Dead (Day 4 post-cull - 28)	2	0	2	1

[a] Litters were weaned on lactational day 21.

Table A6.8.2/01-9 Pup Survival Indices- F2 Pups

	0 ppm	50 ppm	250 ppm	1000 ppm
LIVE BIRTH INDEX				
MEAN	98.7	99.4	99.5	99.2
S.D.	3.69	1.96	1.79	2.37
N	25	22	27	28
4-DAY SURVIVAL INDEX				
MEAN	99.0	98.8	98.6	97.4
S.D.	2.39	2.63	3.23	5.39
N	25	22	27	28
7-DAY SURVIVAL INDEX				
MEAN	100.0	100.0	100.0	100.0
S.D.	0.00	0.00	0.00	0.00
N	25	22	27	28
14-DAY SURVIVAL INDEX				
MEAN	99.0	100.0	99.5	99.6
S.D.	5.00	0.00	2.41	2.36
N	25	22	27	28
21-DAY SURVIVAL INDEX				
MEAN	100.0	100.0	99.5	100.0
S.D.	0.00	0.00	2.41	0.00
N	25	22	27	28
28-DAY SURVIVAL INDEX				
MEAN	100.0	100.0	100.0	100.0
S.D.	0.00	0.00	0.00	0.00
N	25	22	27	28
LACTATIONAL INDEX[a]				
MEAN	99.0	100.0	99.1	99.6
S.D.	5.00	0.00	3.34	2.36
N	25	22	27	28

None significantly different from control group
The equations used for calculating pup survival indices are recorded in the protocol.
[a] Litters were weaned on lactational day 21.

Table A6.8.2/01-10 Litter Size and Sex Ratio (% males) - F2 Pups

	0 ppm	50 ppm	250 ppm	1000 ppm
LACTATIONAL DAY 0				
TOTAL BORN/LITTER				
MEAN	12.1	14.0	12.6	13.6
S.D.	3.47	2.39	3.70	2.44
N	25	22	27	28
TOTAL BORN ALIVE/LITTER				
MEAN	12.0	13.9	12.5	13.5
S.D.	3.45	2.37	3.66	2.47
N	25	22	27	28
SEX RATIO				
MEAN	50.5	51.1	51.5	51.5
S.D.	11.92	14.79	17.25	16.15
N	25	22	27	28
LACTATIONAL DAY 4 (PRECULL)				
LITTER SIZE				
MEAN	11.8	13.7	12.3	13.1
S.D.	3.28	2.29	3.50	2.51
N	25	22	27	28
SEX RATIO				
MEAN	50.1	51.0	51.6	51.8
S.D.	11.97	14.36	17.22	16.18
N	25	22	27	28
LACTATIONAL DAY 4 (POSTCULL)				
LITTER SIZE				
MEAN	7.7	8.0	7.6	8.0
S.D.	0.95	0.00	1.28	0.00
N	25	22	27	28
SEX RATIO				
MEAN	48.8	50.0	53.2	51.8
S.D.	7.47	6.68	11.28	11.64
N	25	22	27	28
LACTATIONAL DAY 7				
LITTER SIZE				
MEAN	7.7	8.0	7.6	8.0
S.D.	0.95	0.00	1.28	0.00
N	25	22	27	28
SEX RATIO				
MEAN	48.8	50.0	53.2	51.8
S.D.	7.47	6.68	11.28	11.64
N	25	22	27	28
LACTATIONAL DAY 14				
LITTER SIZE				
MEAN	7.6	8.0	7.5	8.0
S.D.	1.00	0.00	1.28	0.19
N	25	22	27	28
SEX RATIO				
MEAN	49.5	50.0	53.0	52.0
S.D.	8.27	6.68	11.44	11.68
N	25	22	27	28

	0 ppm	50 ppm	250 ppm	1000 ppm
LACTATIONAL DAY 21[a]				
LITTER SIZE				
MEAN	7.6	8.0	7.5	8.0
S.D.	1.00	0.00	1.28	0.19
N	25	22	27	28
SEX RATIO				
MEAN	49.5	50.0	53.2	52.0
S.D.	8.27	6.68	11.46	11.68
N	25	22	27	28
LACTATIONAL DAY 28				
LITTER SIZE				
MEAN	7.6	8.0	7.5	8.0
S.D.	1.00	0.00	1.28	0.19
N	25	22	27	28
SEX RATIO				
MEAN	49.5	50.0	53.2	52.0
S.D.	8.27	6.68	11.46	11.68
N	25	22	27	28

None significantly different from control group
[a] Litters were weaned on lactational day 21.

Table A6.8.2/01-11 Body Weights Per Litter- F2 Pups

	0 ppm	50 ppm	250 ppm	1000 ppm
LACTATIONAL DAY 1				
ENTIRE LITTER				
MEAN	6.92	6.99	7.10	6.86
S.D.	0.814	0.664	0.659	0.737
N	25	22	27	28
MALE PUPS				
MEAN	7.08	7.21	7.30	7.07
S.D.	0.832	0.683	0.718	0.736
N	25	22	27	28
FEMALE PUPS				
MEAN	6.76	6.78	6.89	6.57
S.D.	0.826	0.645	0.642	0.618
N	25	22	26	27
LACTATIONAL DAY 4 (PRE CULL)				
ENTIRE LITTER				
MEAN	10.26	10.13	10.33	9.94
S.D.	1.616	0.941	1.197	1.292
N	25	22	27	28
MALE PUPS				
MEAN	10.48	10.36	10.54	10.17
S.D.	1.600	0.996	1.240	1.302
N	25	22	27	28
FEMALE PUPS				
MEAN	10.04	9.89	10.06	9.57
S.D.	1.661	0.906	1.179	1.082
N	25	22	26	27
LACTATIONAL DAY 4 (POST CULL)				
ENTIRE LITTER				
MEAN	10.25	10.16	10.40	9.97
S.D.	1.625	0.891	1.144	1.287
N	25	22	27	28
MALE PUPS				
MEAN	10.50	10.39	10.55	10.28
S.D.	1.582	1.014	1.198	1.291
N	25	22	27	28
FEMALE PUPS				
MEAN	10.00	9.95	10.17	9.52
S.D.	1.702	0.864	1.118	1.121
N	25	22	26	27
LACTATIONAL DAY 7				
ENTIRE LITTER				
MEAN	16.33	16.52	16.49	15.99
S.D.	2.230	1.157	1.271	1.662
N	25	22	27	28
MALE PUPS				
MEAN	16.73	16.90	16.70	16.47
S.D.	2.063	1.301	1.333	1.704
N	25	22	26	27
FEMALE PUPS				
MEAN	15.94	16.16	16.20	15.34
S.D.	2.460	1.150	1.330	1.383
N	25	22	26	27

	0 ppm	50 ppm	250 ppm	1000 ppm
LACTATIONAL DAY 14				
ENTIRE LITTER				
MEAN	33.09	34.08	33.81	32.56
S.D.	2.896	1.884	1.951	2.857
N	25	22	27	28
MALE PUPS				
MEAN	33.62	34.71	34.21	33.26
S.D.	2.792	1.846	2.397	2.576
N	25	22	26	27
FEMALE PUPS				
MEAN	32.54	33.44	33.28	31.61
S.D.	2.792	1.846	2.397	2.576
N	25	22	26	27
LACTATIONAL DAY 21				
ENTIRE LITTER				
MEAN	52.77	53.94	52.99	48.96**
S.D.	4.575	3.096	2.716	4.187
N	25	22	27	28
MALE PUPS				
MEAN	53.69	55.22	53.50	49.95**
S.D.	4.919	3.501	3.040	4.269
N	25	22	27	28
FEMALE PUPS				
MEAN	51.79	52.64	52.21	47.57**
S.D.	4.397	2.878	2.940	3.640
N	25	22	26	27
LACTATIONAL DAY 28				
ENTIRE LITTER				
MEAN	88.72	88.27	87.38	78.08**
S.D.	6.188	6.004	4.463	8.043
N	25	22	27	28
MALE PUPS				
MEAN	92.56	92.38	90.39	81.56**
S.D.	7.096	6.566	5.443	8.316
N	25	22	27	28
FEMALE PUPS				
MEAN	84.98	84.11	84.39	73.75**
S.D.	5.684	5.727	4.563	6.457
N	25	22	26	27

* Significantly different from control group ($p < .05$)
** Significantly different from control group ($p < .01$)

Section IIIA 6.12.3 **Annex Point IIA** **VI.6.9.3**	**Health records, both from industry and any other available sources**	
	JUSTIFICATION FOR NON-SUBMISSION OF DATA	Official use only
Other existing data [ ] **Limited exposure [ ]**	**Technically not feasible [ ]** **Scientifically unjustified [ ]** **Other justification [X]**	
Detailed justification:	[redacted] evaluated worker exposure when glutaraldehyde was used as a disinfecting agent in hospital settings. Breathing air samples and area air samples were collected to determine potential exposure. A medical questionnaire was used to determine the prevalence of acute symptoms possibly attributed to glutaraldehyde. Personal air samples ranged from non-detectable to 1.6mg/m^3, 2 of the 5 samples were above the then ACGIH-TLV ceiling of 0.7mg/m^3 (0.2ppm). Area air samples ranged from not detectable to 1.0mg/m^3. Inadequate ventilation, absence of exhaust ventilation and varying work practices were the main contributors to exposure. From 44 completed questionnaires, 28 reported eye and nose irritation, 18 reported throat irritation, 14 reported skin irritation, 7 reported sore throat and 12 reported headache. With adequate ventilation and an improvement and standardisation in work practices the level of exposure can be drastically reduced.	
Undertaking of intended data submission [ ]		
	Evaluation by Competent Authorities	
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted	
	EVALUATION BY RAPPORTEUR MEMBER STATE	
Date	February 14th, 2011	
Evaluation of applicant's justification	The RMS considers that the applicant has provided sufficient information on exposure levels and health records.	
Conclusion	No further information is requested.	
Remarks		
	COMMENTS FROM OTHER MEMBER STATE *(specify)*	
Date	*Give date of comments submitted*	
Evaluation of applicant's justification	*Discuss if deviating from view of rapporteur member state*	
Conclusion	*Discuss if deviating from view of rapporteur member state*	
Remarks		

Section A6.12.4(1) Epidemiological Study

Annex Point IIA6.12.4

IUCLID 5.10/21

Epidemiological studies on the general population, if available
Cohort study

			Official use only
		1 REFERENCE	
1.1	**Reference**	[redacted] (2006), Odour and Chemesthesis from Exposures to Glutaraldehyde Vapour, [redacted] Cain S.W., Schmidt R., Jalowayski A.A. (2007), Odour and Chemesthesis from Exposures to Glutaraldehyde Vapour, Chemosensory Perception Laboratory, Department of Surgery (Otolaryngology), University of California, San Diego, La Jolla, CA 92093-0957, USA, Int. Arch Occup Environ Health, 1 March 2007. [redacted], 2006, Occupational Exposure Limits for Glutaraldehdye: A Comparision to Exposures in the Cain Glutaraldehyde Study, [redacted], 30 November 2006	X
1.2	**Data protection**	Yes	
1.2.1	Data owner	The Dow Chemical Company	
1.2.2	Companies with letter of access	[redacted]	
1.2.3	Criteria for data protection	Data submitted to the MS after 13 May 2000 on existing a.s. for the purpose of its entry into Annex I.	
		2 GUIDELINES AND QUALITY ASSURANCE	
		Not applicable	
		3 MATERIALS AND METHODS	
3.1	**Test material**	Glutaraldehyde 26.1% or 25.7% in water [redacted]	
3.1.1	Lot/Batch number	[redacted]	
3.1.2	Specification	Deviating from specification given in section 2 as follows:	
3.1.2.1	Purity	[redacted]	
3.2	**Type of study**	Cohort study	
3.3	**Method of data collection**	Telephone interview followed by a laboratory screening.	
3.4	**Test Persons / Study Population**	Non-entry field	
3.4.1	Selection criteria	Suitable volunteers	
3.4.2	Number of test persons per group/cohort size	50 subjects [redacted]	
3.4.3	Sex	Female	
3.4.4	Age	18-35 years	
3.4.5	Diseases	Healthy	

Section A6.12.4(1)	Epidemiological Study	
Annex Point IIA6.12.4 IUCLID 5.10/21	Epidemiological studies on the general population, if available Cohort study	
3.4.6	Smoking status	Non-smokers
3.5	**Controls**	Yes
3.5.1	Type of control	The subjects detailed above were used for test experiments and control experiments.
3.6	**Administration/ Exposure**	No Entry field
3.6.1	Exposure Route	Inhalation
3.6.2	Exposure Situation	Laboratory tests using vapour delivery equipment presented controlled amounts of glutaraldehyde to the subjects.
3.6.3	Exposure concentration(s)	Information available, see Section 4.1.1.2 Measured
3.6.4	Method(s) to determine exposure	Area air sampling OSHA Method 64 was employed. Glutaraldehyde is sampled from air using 2 glass fibre filters each of which is coated with 2,4-dinitrophenylhydrazine and phosphoric acid. A known volume of air is drawn through an open face air monitoring cassette which contains the coated filters. The sample filters are extracted separately with 2mL of acetonitrile for 1h on a tube rotator at 60 rpm. The extracts are then analysed by HPLC with uv detection. The concentration in µg per sample is determined by comparing its detector response to the calibration curve. See TNG Summary 4.2(b) for a full description of this method.
3.6.5	Postexposure period	Not applicable
3.7	**Examinations**	No Entry field
3.7.1	Type of disease	Not applicable
3.7.2	Parameters	Not applicable
3.8	**Further remarks**	None

4 RESULTS AND DISCUSSION

4.1	**Exposure**	No Entry field
4.1.1.1	Number of measurements	**Experiment 1**: samples for analysis were taken 2/day during odour testing at the highest level (*ca* 5ppb for odour detection) and 4/day during chemesthesis (chemically stimulated feel) testing at the lowest and highest concentrations tested on that day. **Experiment 2**: samples were taken from each chamber at each exposure level on each day of testing.
4.1.1.2	Average concentrations	**Experiment 1**: See Figure 6.12.4(1)-3 (Range 67.6-112.2% expressed as % of target) **Experiment 2**:

Nominal ppm	Mean Measured ppm	Standard deviation	Coefficient of variation	Measured as a % of nominal
35	35.78	5.39	15.07	102

Section A6.12.4(1) **Epidemiological Study**

Annex Point IIA6.12.4

IUCLID 5.10/21

Epidemiological studies on the general population, if available

Cohort study

		50	52.94	6.82	12.88	106
		75	78.97	9.34	11.82	105
		100	103.45	7.93	7.67	103

4.1.1.3	Standard deviation	See Section 4.1.1.2
4.1.1.4	Date(s) of measurement(s)	**Experiment 1**: carried out daily during testing. **Experiment 2**: carried out daily during the 27 days of testing.
4.1.2	Other	None
4.2	**Number of cases for each disease / parameter under consideration**	Not applicable
4.3	**SMR (Standard mortality ratio), RR (relative risk), OR (Odds ratio)**	Not applicable
4.4	**Other Observations**	None

5 APPLICANT'S SUMMARY AND CONCLUSION

5.1 **Materials and methods**

Experiment 1 (brief exposures, up to 5 sec for the nose and up to 25 sec for the eye): On a given day subjects performed one of the following 3 tasks; odour detection, ocular detection or nasal localisation of feel. An 8 station vapour delivery device (VDD) presented controlled amounts of glutaraldehyde to the subjects. Glutaraldehyde in water was metered to a heated chamber where the liquid was vaporised. A flow of nitrogen carried the vapour to rotameters that led to 8 stations each of which ended in 3 glass cones from which the subjects sampled the vapour. Testing of odour progressed from lowest to highest concentrations (range: 0.039 to 4.95ppb) in 2-fold increments. Testing of chemesthesis progressed from 229 to 772 ppb in 1.5-fold increments. Samples were collected for analysis and analysed as per Section 3.6.4. The subjects sampled from 3 cones per station in which one cone was active and the other 2 were blanks. The subject marked their choice of active cone on a data sheet along with a confidence rating in their choice.

Experiment 2 (15 min exposures): including 2 exposures to air, 2 exposures to 10ppm heptane (as an odour control) and 1 exposure to each of 35, 50, 75 and 100ppb. Exposures were carried out in a chamber ($6.2m^3$) which had been equilibrated prior to the subject entering the chamber. Ratings were given for feel in the nose, in the eyes and in the throat. The level of confidence at which chemesthetic sensations were induced was reported during 15 min exposures of glutaraldehyde at concentrations of occupational interest. Samples were collected for analysis and analysed as per Section 3.6.4. Subjects scored whether the level of glutaraldehyde stimulated feel in the eyes, in the nose and in the throat.

5.2 **Results and discussion**

Experiment 1: detection of odour occurred over the range 0.04ppb (near chance detection) to 4ppb (virtually perfect detection) with a median of 0.3ppb. Detection of ocular feel occurred at 390ppb and

Section A6.12.4(1)		**Epidemiological Study**
Annex Point IIA6.12.4		**Epidemiological studies on the general population, if available**
IUCLID 5.10/21		**Cohort study**
		nasal feel at 470ppb. **Experiment 2**: levels of glutaraldehyde in the range 35 to 100ppb were tested. These levels were discernible by the odour but did not induce chemesthesis over exposure durations of 15 min. It appears from the results that the order of sensitivity is in the order eyes>nose>throat.
5.3	**Conclusion**	**Experiment 1**: The median level for odour detection is approximately 150 times lower than the current maximum exposure limit (MEL) of 50ppb for glutaraldehyde. **Experiment 2**: levels of glutaraldehyde in the range 35 to 100ppb were tested. These levels were discernible by the odour but did not induce chemesthesis over exposure durations of 15 min.
5.3.1	Reliability	2
5.3.2	Validity	The study is considered to be valid as reported by Ripple S., 2006. 'Applicability of chemesthetic threshold studies to setting guidelines • The chemesthetic thresholds for GA as determined by Cain *et al* in a controlled laboratory environment should be viewed as the key study in establishing guidelines to prevent irritation. The odour threshold for GA was established by Cain *et al* at less than 0.001 ppm. An odour threshold as low as the one for glutaraldehyde serves as a good warning signal that exposure has occurred. It also contributes to altering an individual's responsivity to GA by changing their attitude or expectations about their exposure. This study reduces the uncertainty associated with data previously used to establish OELs because the protocol is designed to significantly reduce confounders of reported irritation such as odour and perceived risk of glutaraldehyde exposure, which likely contributed to reports of irritation at lower levels in previous studies. • The Cain study provides a quantitative measure for a conservative and transient threshold for sensory irritation in the naïve worker population. Thus, the ocular and nasal chemesthetic thresholds can be considered the no-observable adverse effect level (NOAEL) for sensory irritation given that this threshold is based on the perception of 'feeling' the chemical, which is considered to be the first and most sensitive step in the cascade of physiological events leading to chemical induced-irritation. • OSHA and NIOSH were provided the opportunity to review the study protocol during the design-phase since they would ultimately be using the study data to set safe exposure levels or OELs. NIOSH, the research support group for OSHA, gave positive feedback on the value of the study and recommended minor protocol enhancements. OSHA's comments were also favorable, and the agency was kept apprised periodically of the status of the testing . Both groups acknowledged the value of conducting the study and its utility for establishing relevant, health-protective OELs.'
5.3.3	Deficiencies	No
5.4	**Other**	No

Section A6.12.4(1) Epidemiological Study

Annex Point IIA6.12.4

IUCLID 5.10/21

Epidemiological studies on the general population, if available
Cohort study

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	February 9th, 2011
Materials and Methods	Reference. 1.1 Reference. The reference for the published article is as follows: Cain WS, Schmidt R, Jalowayski AA (2007) Odor and chemesthesis from exposures to glutaraldehyde vapor. Int. Arch. Occup. Environ. Health, 80:8, 721-731.
Results and discussion	Agree with applicant's version.
Conclusion	In an experimental setup, glutaraldehyde odour can be detected at 0.3 ppb. The concentration around OEL (35-100 ppb) should be recognisable but not irritating. Irritation will occur at concentrations above the detection levels for ocular (390 ppb) and nasal (470 ppb) feel.
Reliability	2
Acceptability	Acceptable
Remarks	
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Figure 6.12.4(1)-1 Graphical Representation of Odour Detection, Ocular Detection and Nasal Localisation from Experiment 1

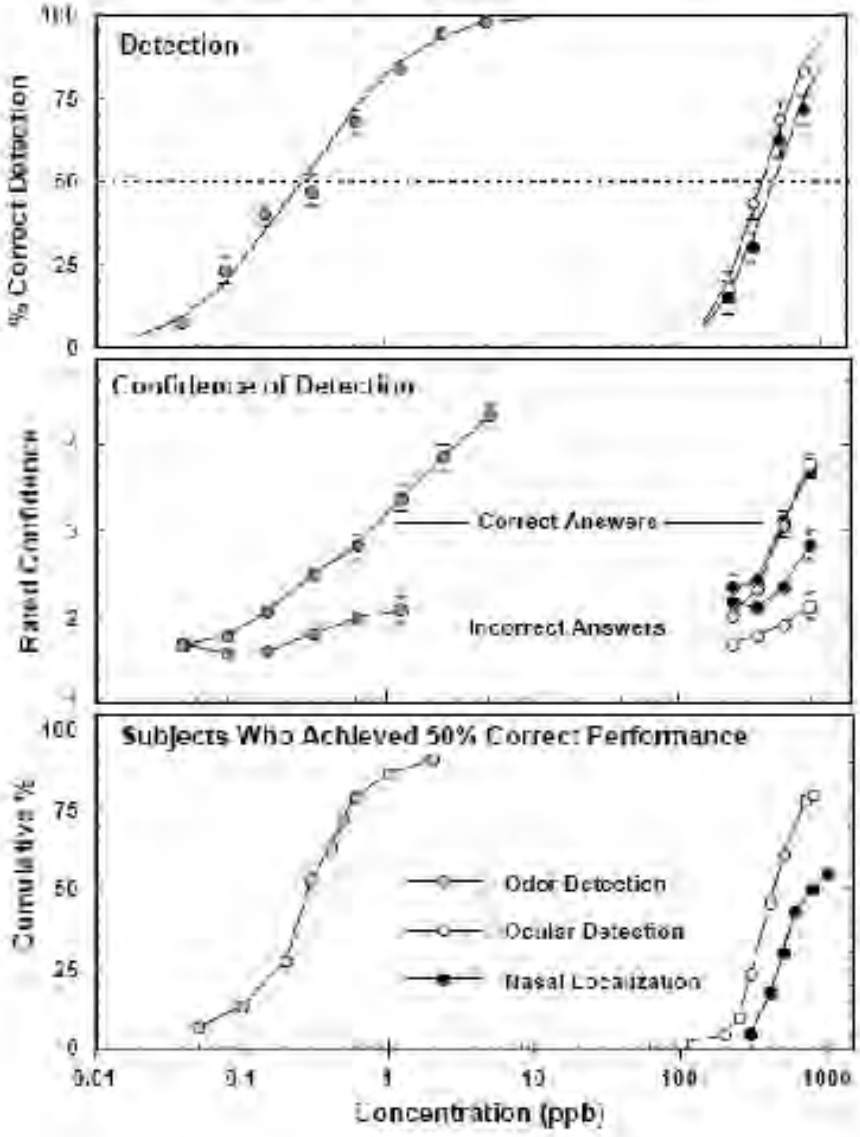


Figure 6.12.4(1)-2 Graphical Representation of Ocular Chemesthesis, Nasal Chemesthsis and Throat Chemesthesis from Experiment 2

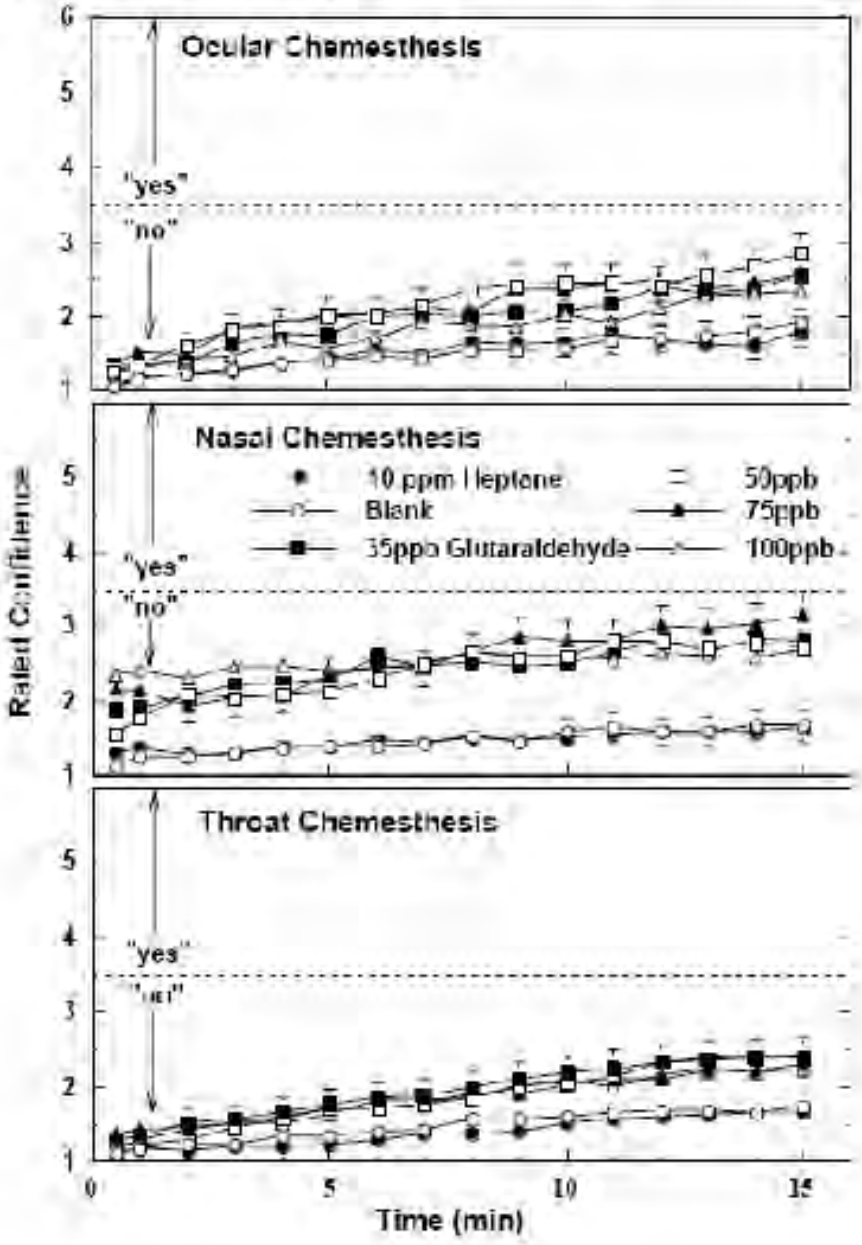
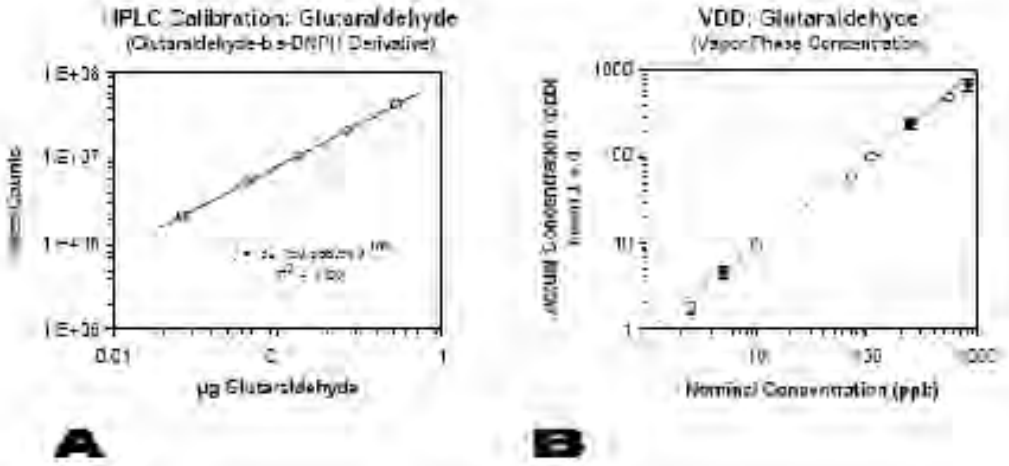


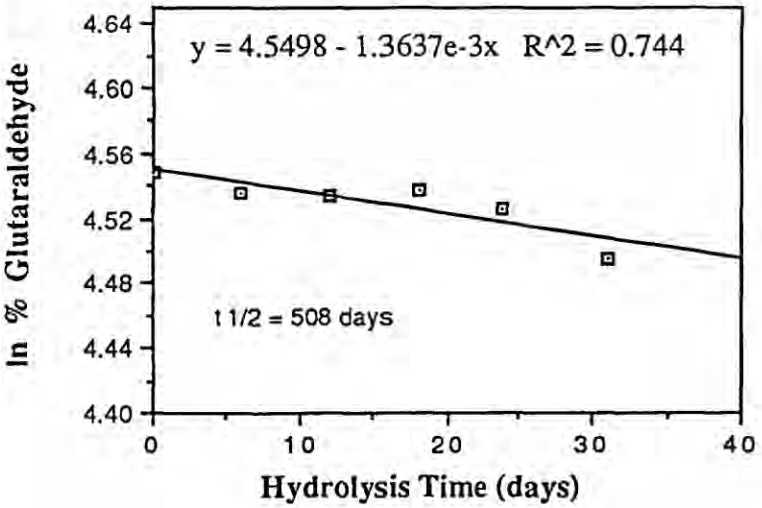
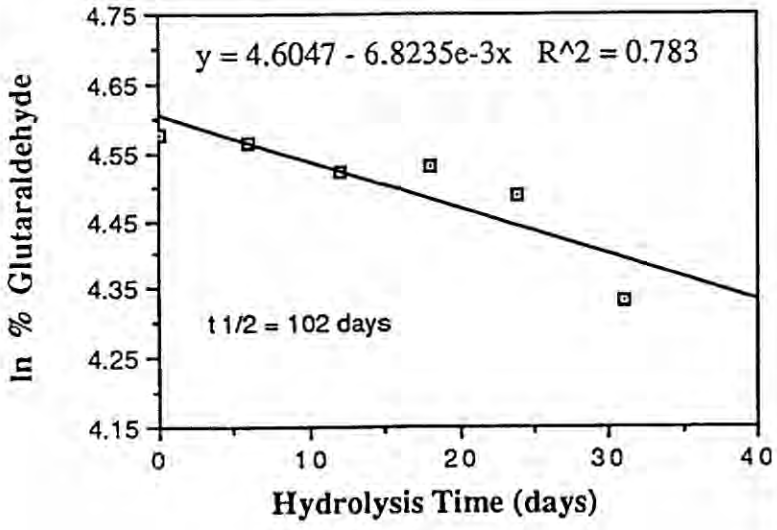
Figure 6.12.4(1)-3

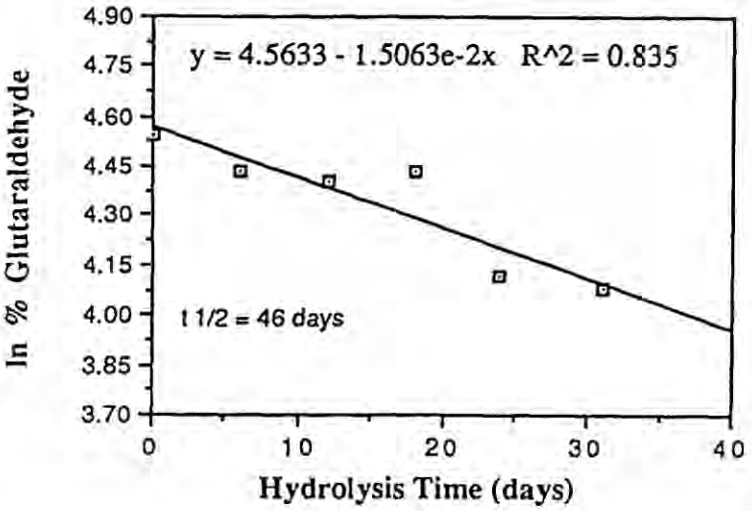
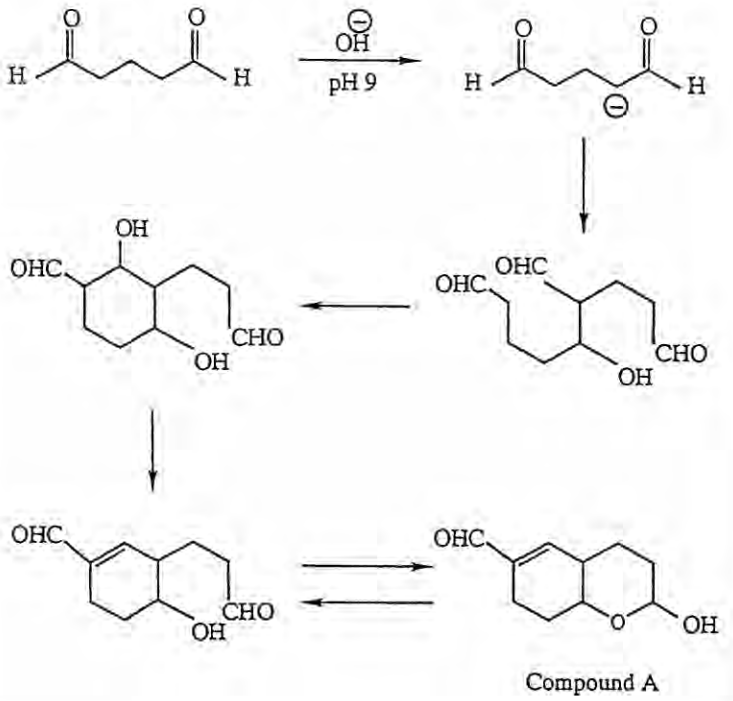
Text Figure 2. Calibration (A) and Analytical Confirmation of Glutaraldehyde Vapor Concentration (B)



Section A7.1.1.1.1 **Annex Point IIA, II.7.6.2.1** **IUCLID 3.1.2/01**		**Hydrolysis as a function of pH and identification of breakdown products**	
		1 REFERENCE	**Official use only**
1.1	**Reference**	[redacted] (1992a) Hydrolysis of [1,5-14C] glutaraldehyde at pH 5, 7 and 9, [redacted], Unpublished, 23 December 1992	
1.2	**Data protection**	Yes	
1.2.1	Data owner	The Dow Chemical Company	
1.2.2	Companies with letter of access	[redacted]	
1.2.3	Criteria for data protection	Data submitted to the MS after 13 May 2000 on an existing a.s. for the purpose of its entry to Annex I	
		2 GUIDELINES AND QUALITY ASSURANCE	
2.1	**Guideline study**	Yes US EPA FIFRA N 161-1	
2.2	**GLP**	Yes	
2.3	**Deviations**	No	
		3 MATERIALS AND METHODS	
3.1	**Test material**	Radiolabeled glutaraldehyde, specific activity 13.6 mCi/mmole  [1,5-^{14}C]-Glutaraldehyde ** denotes position of the radio-label ^{14}C*	
3.1.1	Lot/Batch number	[redacted]	
3.1.2	Specification	Not reported	
3.1.3	Purity	[redacted]	
3.1.4	Further relevant properties	none	
3.2	**Reference substances**	Glutaraldehyde Glutaric acid Glutaric anhydride	
3.2.1	Initial concentration of reference substance	For use as chromatographic reference standards only.	
3.3	**Test solution**	The preparation of the buffer solutions is described in **Table A7.1.1.1.1/01-1.**	

Section A7.1.1.1.1 **Annex Point IIA, II.7.6.2.1** **IUCLID 3.1.2/01**		**Hydrolysis as a function of pH and identification of breakdown products**	
		The experimental procedures, including replications and experimental conditions are provided in **Table A7.1.1.1.1/01-2**.	
3.4	**Testing procedure**		
3.4.1	Test system	The test system is described in **Table A7.1.1.1.1/01-3**. This table also includes a list of the equipment used in the study.	
3.4.2	Temperature	24.5-25.5 °C	
3.4.3	pH	5, 7, and 9	
3.4.4	Duration of the test	31 days	
3.4.5	Number of replicates	2 (duplicates) per sampling point	
3.4.6	Sampling	Duplicate (2) samples of vessels treated with radiolabeled material were collected at 0, 6, 12, 18, 24, and 31 days after treatment (DAT). See also **Table A7.1.1.1.1/02-2**.	
3.4.7	Analytical methods	Samples were analyzed directly by HPLC, LSC, and TLC co-chromatography with a ^{14}C glutaraldehyde analytical standard provided by the Sponsor. After sampling, all samples were frozen, and reanalyzed after 7 days storage to confirm that there was no change in the HPLC profile. Thin layer chromatography and/or gas chromatography-mass spectroscopy were also used to confirm the concentration of glutaraldehyde or degradation products at the various timepoints. GC/MS was used on selected samples in order to characterize unknown degradates and confirm glutaraldehyde concentrations. LC-MS was used to characterize the pH 9 unknown degradate. Chromatographic methods were validated with the authentic standards achieving the necessary resolution and sensitivity. Products with yields as low as 0.2% could be reliably quantified and 0.1% yields detected.	
3.5	**Preliminary test**	Yes Solution stability was examined at nominal concentration of 10 ppm in the three buffer solutions to determine the possibility of physical or chemical sorption to test containers. Results confirmed that the material remains in solution in the test system for 48 hours. A pilot application of 1,5-^{14}C-glutaraldehyde was conducted on single replicates for each pH at nominal concentrations of 10 ppm. Samples were analyzed at one and seven days to estimate the hydrolysis rate and determine the appropriate sampling schedules for the definitive study.	
		4 RESULTS	
4.1	**Concentration and hydrolysis values**	Data are reported in ***Table A7.1.1.1.1/01-4, 5, 6***	
4.2	**Hydrolysis rate constant (k_h)**	The half lives were 508 days at pH 5, 102 days at pH 7, and 46 days at pH 9 with a starting concentration of 10 ppm.	x
4.3	**Dissipation time**	The extrapolated half-lives: pH $t_{1/2}$ R^2 5 508 0.74	

Section A7.1.1.1.1 **Annex Point IIA, II.7.6.2.1** **IUCLID 3.1.2/01**	**Hydrolysis as a function of pH and identification of breakdown products**	
	7 102 0.78 9 46 0.84	
4.4 Concentration – time data	*Hydrolysis of 1,5-^{14}C-Glutaraldehyde at pH 5*  *Hydrolysis of 1,5-^{14}C-Glutaraldehyde at pH 7*  	

Section A7.1.1.1.1 **Annex Point IIA, II.7.6.2.1** **IUCLID 3.1.2/01**	**Hydrolysis as a function of pH and identification of breakdown products**	
	Hydrolysis of 1,5-^{14}C-Glutaraldehyde at pH 9  	
4.5 Specification of the transformation products	At pH 9, one major hydrolysis product was observed, referred to as Compound A. Compound A was less polar than the parent, with a longer HPLC retention time. In 31 days, it accounted for >30% of the radioactivity in the pH 9 samples. It was also found in the pH 7 samples, but at less than 10% of the radioactivity. Compound A was proposed to be a bicyclic dimer of glutaraldehyde based on mass spectroscopy results. *Proposed Hydrolysis Pathway for 1,5-^{14}C-Glutaraldehyde*  	

Section A7.1.1.1.1 **Annex Point IIA, II.7.6.2.1** **IUCLID 3.1.2/01**	**Hydrolysis as a function of pH and identification of breakdown products**	
	5 APPLICANT'S SUMMARY AND CONCLUSION	
5.1 Materials and methods	The test material was evaluated in purified water (meeting ASTM-D1193 standards for reagent grade water) at 10 ppm, verified by radio-analysis. Samples were kept in the dark in sterilized glassware. Cultures of samples showed no growth after 48 hours when plated. The temperature range for the test was 24.5-25.5 °C. Buffer solutions were acetate (pH 5), phosphate (pH 7), and borate (pH 9), with no cosolvent used. The pH of each solution was measured with a pH meter at test day 31, and at all other time points with pH paper. Samples were taken from each solution at test days 0, 6, 12, 18, 24, and 31. Samples were analyzed directly by HPLC, LSC, and TLC co-chromatography with a ^{14}C glutaraldehyde analytical standard provided by the Sponsor. After sampling, all samples were frozen, and reanalyzed after 7 days storage to confirm that there was no change in the HPLC profile. Thin layer chromatography and/or gas chromatography-mass spectroscopy were also used to confirm the concentration of glutaraldehyde or degradation products at the various timepoints. LC-MS was used to characterize the pH 9 unknown degradate. Chromatographic methods were validated with the authentic standards achieving the necessary resolution and sensitivity. Products with yields as low as 0.2% could be reliably quantified and 0.1% yields detected. The hydrolysis rate constant and half-lives were calculated using pseudo-first order kinetics. Degradation rate constants were calculated from a standard equation.	
5.2 Results and discussion	***Table A7.1.1.1.1/01-4, 5, 6*** Mass-balance of the radiotracer was determined, and recoveries averaged greater than 95% for all three solutions. The pH of the buffer solutions did not change significantly during the study period. Sterility tests confirmed that the buffer solutions were not contaminated. Glutaraldehyde did not appear to degrade appreciably in buffer systems at pH 5 and pH 7. At pH 9, however, the test material degraded appreciably during the study period. By test day 31, less than 60% of the radiocarbon in the hydrolysate remained as glutaraldehyde. One major transformation product in the pH 9 system was observed: 3-formyl-6-hydroxy-2-cyclohexene-1-propanal. The metabolite is a cyclized dimer of glutaraldehyde. The proportions of other metabolites were less than 10% of the applied radioactivity. The extrapolated half-lives: pH t½ R^2 5 508 0.74 7 102 0.78 9 46 0.84 After 31 days 95%, 78% and 63% glutaraldehyde was detected at pH 5, 7 and 9, respectively. The half-lives were calculated assuming pseudo-first order kinetics.	

Section A7.1.1.1.1 **Annex Point IIA, II.7.6.2.1** **IUCLID 3.1.2/01**	**Hydrolysis as a function of pH and identification of breakdown products**	
5.2.1 k_H	Not determined	
5.2.2 DT_{50}	pH $t_{1/2}$ R^2 5 508 0.74 7 102 0.78 9 46 0.84	
5.2.3 r^2	0.74, 0.78, 0.84 for pH 5, 7, and 9, respectively	
5.3 Conclusion	Glutaraldehyde is stable to hydrolysis at pH 5, and is somewhat stable at pH 7. At pH 9, one major hydrolysis product was observed, referred to as Compound A. Compound A was less polar than the parent, with a longer HPLC retention time. In 31 days, it accounted for >30% of the radioactivity in the pH 9 samples. It was also found in the pH 7 samples, but at less than 10% of the radioactivity. Compound A was proposed to be a bicyclic dimer of glutaraldehyde based on mass spectroscopy results.	
5.3.1 Reliability	1	
5.3.2 Deficiencies	No	
	Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE	
Date	22.8.2008	
Materials and Methods	The applicant's description of materials and methods is correct. A few deviations from the current OECD 111 guideline were identified: a preliminary test at 50 °C was not done, bubbling with nitrogen or argon in order to avoid oxygen is not mentioned in the test report, hydrolysis has been tested at pH 5 instead of 4, confidence intervals have not been reported for half lives. 4.2: Half lives are given instead of rate constants, rate constants have not been reported.	
Results and discussion	The applicant's version is correct. The hydrolytic half-lives of glutaraldehyde were 508 d, 102 d and 46 d at pH 5, 7 and 9, respectively. The corresponding rate constants calculated from the half lives with eq. $k=\ln 2/t_{1/2}$ were $1.36E^{-3}$, $6.79E^{-3}$ and $1.51E^{-2}$. Average radioactivity recovery ranged from 94.5 to 99.4% fulfilling the quality criteria of 90-110% recovery for labelled chemicals. The analytical method was reported to be sufficient to quantify test substance down to less than 10% of initial concentration.	
Conclusion	Glutaraldehyde is stable to hydrolysis at pH 5, and is somewhat stable at pH 7. No hydrolysis products exceeded 10% at pH 5 and 7. Glutaraldehyde hydrolyses at pH 9 and one transformation product was formed: 59.2% of radioactivity accounted for glutaraldehyde and 30.8% accounted for Compound A at the end of the test (31 d). Compound A was proposed to be a bicyclic dimer of glutaraldehyde based on mass spectroscopy results.	
Reliability	2	
Acceptability	Acceptable	

Section A7.1.1.1.1 Annex Point IIA, II.7.6.2.1 IUCLID 3.1.2/01	**Hydrolysis as a function of pH and identification of breakdown products**	
Remarks		
	COMMENTS FROM ...	
Date	*Give date of comments submitted*	
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*	
Results and discussion	*Discuss if deviating from view of rapporteur member state*	
Conclusion	*Discuss if deviating from view of rapporteur member state*	
Reliability	*Discuss if deviating from view of rapporteur member state*	
Acceptability	*Discuss if deviating from view of rapporteur member state*	
Remarks		

Table A7.1.1.1.1/01-1 Composition of buffer solutions

pH	Composition
5	146 mL of 0.1M acetic acid, added to 100 mL of 0.1M NaOH, and distilled water to make a final volume of 1 liter
7	22.4 mL of 0.1M KH_2PO_4, added to 25.8 mL of 0.1M Na_2HPO_4, and distilled water to make a final volume of 1 liter
9	46 mL of 0.04M HCl, added to 500 mL of 0.01M $Na_2B_4O_7$*$10H_2O$, and distilled water to make a final volume of 1 liter

Table A7.1.1.1.1/01-2 Description of test solution

Criteria	Details
Purity of water	Distilled, sterilized water
Preparation of test medium	The test material was added to distilled water (9.57×10^8 dpm) to make a 10 mL stock solution. It was added to the sterilized buffers to make test solutions.
Test concentrations (mg a.i./L)	Using sterile technique, test material was added to buffer solution in an individual pyrex sample tube. The resulting initial concentration (C_0) was measured at 10 ppm for each replicate. Samples were maintained in the dark. Glassware was autoclaved prior to use.

Temperature (°C)	25 ± 0.2 °C
Controls	Sterility was verified.
Identity of co-solvent	The dosing solution was prepared in water.
Replicates	Duplicate samples were tested on days 0, 6, 12, 18, 24, and 31

Table A7.1.1.1.1/01-3 Description of test system

Glassware	The test flasks used in this study were pyrex tubes with Teflon® screw caps.
Other equipment	Equipment used in this study included a temperature-monitored incubator (dark), HPLC,TLC system, LSC, GC-MS, LC-MS, Chemical Ionization mass spectroscopy. A pH meter was also required.
Method of sterilization	The buffers were sterilized by filtration, and all glassware was sterilized by autoclaving.

Table A7.1.1.1.1/01-4 Degradation of 1,5-^{14}C-Glutaraldehyde in Buffered Solutions (pH 5)

Sample	% Glutaraldehyde	% Unknowns	% Compound A	Recovery
Day 0				
A	94.4	2.2	*	96.6
B	94.5	0.7	*	95.2
Day 6				
A	93.9	1.7	*	95.6
B	92.6	1.3	*	93.9
Day 12				
A	92.4	2.4	*	94.8
B	93.8	0.5	*	94.3
Day 18				
A	95.2	1.0	*	96.2
B	91.5	2.6	*	94.1
Day 24				
A	91.1	4.1	*	95.2
B	93.7	4.6	*	98.3
Day 31				
A	87.1	7.3	*	94.4
B	92.1	3.1	*	95.2

*No single peak of any other degradate exceeded 10% of the applied radiocarbon. Compound A was not identified in these samples.

Table A7.1.1.1.1/01-5 Degradation of 1,5-^{14}C-Glutaraldehyde in Buffered Solutions (pH 7)

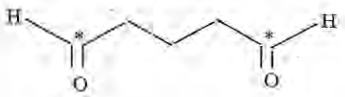
Sample	% Glutaraldehyde	% Unknowns	% Compound A	Recovery
Day 0				
A	97.0	2.3	*	99.3
B	97.3	1.6	*	98.9
Day 6				
A	96.2	1.5	*	97.7
B	95.9	3.8	*	99.7
Day 12				
A	92.2	3.5	*	95.7
B	91.5	5.2	*	96.7
Day 18				
A	94.5	1.1	*	95.6
B	90.7	6.6	*	97.3
Day 24				
A	92.2	7.1	*	99.3
B	85.2	14.3	*	99.5
Day 31				
A	77.4	21.3	*	98.7
B	74.9	22.0	*	96.9

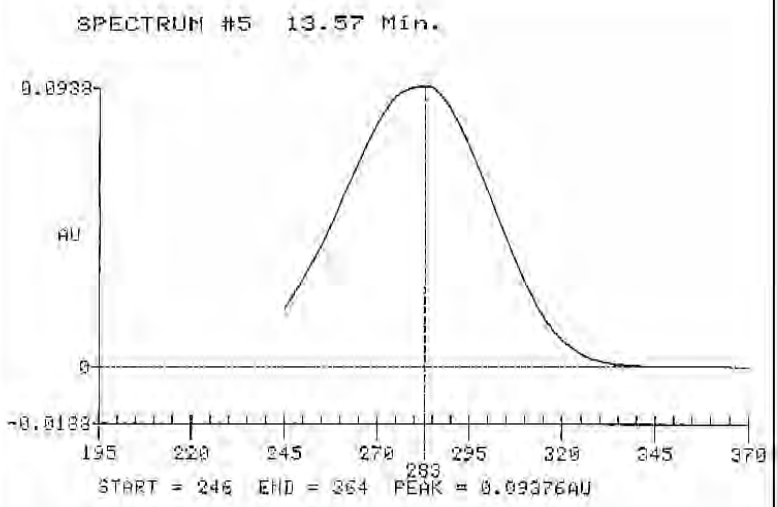
*No single peak of any other degradate exceeded 10% of the applied radiocarbon. Compound A was not identified in these samples >10%.

Table A7.1.1.1.1/01-6 Degradation of 1,5-^{14}C-Glutaraldehyde in Buffered Solutions (pH 9)

Sample	% Glutaraldehyde	% Unknowns	% Compound A	Recovery
Day 0				
A	95.6	0.2	0.0	95.8
B	92.1	2.8	0.5	95.4
Day 6				
A	84.4	4.8	5.7	94.9
B	84.3	1.0	9.9	95.2
Day 12				
A	77.2	2.8	15.3	95.3
B	86.0	1.0	10.1	97.1
Day 18				
A	78.6	2.1	15.4	96.1
B	89.7	0.0	6.0	95.7
Day 24				
A	61.8	10.0	28.1	9.99
B	60.9	5.9	30.9	97.7
Day 31				
A	57.4	7.4	31.7	96.5
B	60.9	5.2	30.0	96.1

*No single peak of any other degradate exceeded 10% of the applied radiocarbon.

Section A7.1.1.1.2 **Annex Point IIA, II.7.6.2.2** **IUCLID 3.1.1/01**		**Phototransformation in water including identity of transformation products**	
		1 REFERENCE	**Official use only**
1.1	**Reference**	[redacted] (1992b) Sunlight photodegradation of [1,5-^{14}C] glutaraldehyde in a buffered aqueous solution at pH 5, [redacted], Unpublished, 17 August 1992	
1.2	**Data protection**	Yes	
1.2.1	Data owner	The Dow Chemical Company	
1.2.2	Companies with letter of access	[redacted]	
1.2.3	Criteria for data protection	Data submitted to the MS after 13 May 2000 on existing a.s. for the purpose of its entry into Annex I	
		2 GUIDELINES AND QUALITY ASSURANCE	
2.1	**Guideline study**	Yes US EPA FIFRA 161-2	
2.2	**GLP**	Yes	
2.3	**Deviations**	No	
		3 MATERIALS AND METHODS	
3.1	**Test material**	Radiolabeled glutaraldehyde, specific activity 13.6 mCi/mmole  [1,5-^{14}C]-Glutaraldehyde * denotes position of the radio-label ^{14}C	
3.1.1	Lot/Batch number	[redacted]	
3.1.2	Specification	Not reported	
3.1.3	Purity	[redacted]	

Section A7.1.1.1.2 Annex Point IIA, II.7.6.2.2 IUCLID 3.1.1/01		Phototransformation in water including identity of transformation products	
3.1.4	UV/VIS absorption spectra and absorbance value		
3.1.5	Further relevant properties	None	
3.2	**Reference substances**	***Chromatographic Standards*** Glutaric acid Glutaric anhydride Glutaraldehyde (50% a.i.)	
3.3	**Test solution**	Water for buffer solutions was purified to meet ASTM-D1193 standards for reagent grade water. There was no cosolvent used. Buffer solutions were sterilized prior to use. The pH 5 buffer system was prepared with 146 mL of 0.10M acetic acid solution, added to 100 mL of 0.10M NaOH. The final volume was brought to 1 liter with distilled water. An application solution of 7.3 mL aqueous radiolabeled glutaraldehyde containing 817,123,348 dpm (2707 ug) to 253 mL sterilized buffer. Solutions were transferred to sample tubes using aseptic technique. The final concentration of the radiolabeled glutaraldehyde was 10.4 ppm. **Table A7.1.1.1.2/01-1**	
3.4	**Testing procedure**		
3.4.1	Test system	Solutions were mixed and irradiated in quartz sample tubes; dark controls were in pyrex containers, wrapped in foil during the study period. All samples (except the controls) were irradiated and collected in duplicate at time 0 and five subsequent time points. The sampling intervals were 0, 3, 7, 15, 23 and 30 days. **Table A7.1.1.1.2/01-2**	
3.4.2	Properties of light source	Natural sunlight was used as the light source.	
3.4.3	Determination of irradiance	Irradiance was measured with an International Light #1490 Radiometer (wavelength interval of integration was 250-700 nm). Average total light energy (daily) over the 30-day exposure period was 5.68 $\pm$ 2.07	