Annex I to the CLH report

Proposal for Harmonised Classification and Labelling

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

International Chemical Identification:

Chemical name: Potassium chlorate

EC Number: 223-289-7

CAS Number: 3811-04-9

Index Number: 017-004-00-3

Contact details for dossier submitter:

Version number: 2 **Date: 21/11/2019**

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1. ACUTE TOXICITY ORAL

1.1. Study 1 (Study report 1991i)

ENDPOINT_STUDY_RECORD: 7775-09-9, Acute toxicity: oral, 1991i, RS

Reliability
1 (reliable without restriction)

Rationale for reliability incl. deficiencies
other: Study was performed under GLP and according to internationally accpeted guidelines.

Data source

Reference
EPA acute oral toxicity limit test. / study report

Materials and methods
Test guideline
Qualifier
according to **Guideline** EPA OPP 81-1 (Acute Oral Toxicity)
GLP compliance yes
Test type standard acute method
Limit test Yes

Test animals
Species rat common species
Strain Sprague-Dawley rat
Sex male/female

Details on test animals and environmental conditions

TEST ANIMALS
- Source: Hilltop Lab Animals, Scottdale, PA
- Age at study initiation: Young adults
- Weight at study initiation: weighing 222 - 293 grams
- Fasting period before study: Approximately 18 hours prior to selction and test initiation
- Housing: individually in suspended stainless steel caging with mesh floors
- Diet (e.g. ad libitum): Pelleted Purina Rat Chow #5012, ad lib
- Water (e.g. ad libitum): Tap water supplied by automatic water system, ad lib
- Acclimation period: 28 or 30 days

ENVIRONMENTAL CONDITIONS
- Temperature (°C): 20 - 23
- Humidity (%): no info
- Air changes (per hr): no info
- Photoperiod (hrs dark / hrs light): no info
IN-LIFE DATES: Rangefinding: From: November 29, 1990 To: December 7, 1990;
Test: From: December 19, 1990 To: January 11, 1991

Administration / exposure
Route of administration oral: gavage
Vehicle water

Details on oral exposure
VEHICLE
- Concentration in vehicle: 50% w/w solution in distilled water
- Amount of vehicle (if gavage): no info
- Justification for choice of vehicle: no info
- Lot/batch no. (if required): no info
- Purity: no info
MAXIMUM DOSE VOLUME APPLIED: approximately 7 ml/kg
Doses
- range finding study: 300, 600, 1250, 2500 and 5000 mg/kg bw; (one male and one female per dose)
- full acute oral limit test 1: 5000 mg/kg bw
- full acute oral limit test 2: 2000 mg/kg bw

No. of animals per sex per dose full acute oral limit test: 5

Control animals no
Details on study design
- Duration of observation period following administration: 14 days
- Frequency of observations and weighing: rats were observed at 1, 2 and 4 hours post-dosing and at least once daily thereafter for signs of gross toxicity and mortality. Bodyweights were recorded initial ly, on day 7, at termination (day 14) or after death.
- Necropsy of survivors performed: yes, gross necropsies were performed on the animals that died du ring the study and at termination of the study
- Other examinations performed: clinical and behavioural signs, body weight, histopathology
Statistics No.

Results and discussion
Effect levels
Sex male/female
Dose descriptor LD50
Effect level > 5000 mg/kg bw
Mortality
Dose group 5000 mg/kg bw
- one female died
- Time of death: one day after dosing
Dose group 2000 mg/kg bw
- no animal died
Clinical signs
- Dose group 5000 mg/kg bw
Shortly after dosing several animals appeared lethargic (2 females and 2 males) and had a hunched posture (2 females and 2 males). By 24 hours these conditions were no longer evident and sur vivors appeared active and healthy for the remainder of the test period. The female animal that died showed lethargy and ano-genital staining.
- Dose group 2000 mg/kg bw
one male animal, lethargic and hunched posture. By 24 hours these conditions were no longer evident and survivors appeared active and healthy for the remainder of the test period.
Body weight
- Dose group 5000 mg/kg bw
all survivors gained weight between days 0 and 7 and again between days 7 and 14, although the weight gain in most cases was marginal.
- Dose group 200 mg/kg bw
all survivors gained weight between days 0 and 7 and again between days 7 and 14 at a rate generally expected for the strain and age of animals used.
Gross pathology
- Dose group 5000 mg/kg bw
The female that died showed green discoloration of the intestines, a light green fluid in the stomach, pink liquid in the abdominal cavity and dark red lung discoloration. Other animals showed no significant abnormalities at necropsy, moderate redness in the lungs of all animals.
- Dose group 2000 mg/kg bw
Negligible, slight to moderate redness in the lungs of all animals

Applicant's summary and conclusion
Conclusions
LD50 > 5000 mg/kg bw
Executive summary
The study was performed in accordance with EPA Pesticide Assessment Guidelines, Subdivision F, Hazard Evaluation: Human and Domestic Animals, November 1984, Acute Exposure, oral Toxicity Limit Test (equivalent to OECD Guideline 401 - Acute Oral Toxicity -). Furthermore, the study was designed and performed according to Good Laboratory Practice Standards. The test material, Sodium Chlorate Crystal, was evaluated for its acute oral toxicity potential in 30 Sprague Dawley rats. Ten animals were used in a rangefinding study (dose levels: 5, 2.5, 1.25, 0.6 and 0.3 g/kg bw). Thereafter Sodium Chlorate was administered as gavage doses in a first (5.0 g/kg and second (2.0 g/kg) limit test. No mortality occurred in animals dosed at 2.0 g/kg and 1 animal died at dose level 5.0 g/kg. Clinical signs of toxicity at 5.0 g/kg included hunched posture and reduced feces, which were no longer evident on Day 3. At 2.0 g/kg only hunched posture was observed at 2 -4 hours post dosing in one male. There was no meaningful effect on body weight gain in animals surviving to termination. Necropsy findings at 5.0 g/kg showed green discoloration of the intestines, a light green fluid in the stomach, pink liquid in the abdominal cavity and dark red lung discoloration. At 2.0 g/kg only
slight to moderate redness in the lungs of all animals was observed.
Conclusions: The acute oral LD50 of Sodium Chlorate Crystal was determined to be greater than 5000 mg/kg bw.

1.2. Study 2 (Study report 1981)

ENDPOINT_STUDY_RECORD: 7775-09-9, Acute toxicity: oral, Study report, 1981, SS

Reliability
2 (reliable with restrictions)

Rationale for reliability incl. deficiencies
other: Non-guideline study. A well conducted study. No information on test substance purity.

Data source
Reference
Acute oral toxicity study in rats-Sodium chlorate. / study report

Materials and methods
Test guideline
Qualifier equivalent or similar to
Guideline OECD Guideline 401 (Acute Oral Toxicity) before 2002

GLP compliance
yes
Test type standard acute method
Limit test no

Test animals
Species rat common species
Strain other: Charles River CD
Sex male/female

Details on test animals and environmental conditions
TEST ANIMALS
- Source: Charles River Breeding laboratories, Inc., kingston, New York
- Age at study initiation: 8-10 weeks
- Weight at study initiation: 169.9-229.0 g
- Fasting period before study: night before treatment
- Housing: individually in wire bottom cages
- Diet (e.g. ad libitum): Purina Laboratory Chow, ad lib
- Water (e.g. ad libitum): Acidified water (pH 2.5), ad lib
- Acclimation period: 7 days
ENVIRONMENTAL CONDITIONS
- Temperature (°C): controlled
- Humidity (%): no info
- Air changes (per hr): no info
- Photoperiod (hrs dark / hrs light): 12 hour light cycle
IN-LIFE DATES: From: March, 1981 To: May, 1981

Administration / exposure
Route of administration oral: gavage
Vehicle water

Details on oral exposure
VEHICLE
- Concentration in vehicle: test material was dissolved in deionized water at a concentration of 500 mg/ml and diluted to appropiate concentrations (see 'Maximum dose volume applied')
MAXIMUM DOSE VOLUME APPLIED: Volume administered or concentration: 10ml/kg bw at doses of 5000 mg/kg and lower. 13.6 and 20 ml/ kg bw at 6810 and 10000 mg/kg bw, respectively. Due to insolubility of the compound.
Doses
- range finding study: 1000, 1500, 5000 mg/kg bw
- main study: 1470, 2150, 3160, 4640, 6810 mg/kg bw males and 2150, 3160, 4640, 6810, 10000 mg/kg bw females
No. of animals per sex per dose
- Range finding study: 2 rats of each sex per dose
- Main study: 8 rats of each sex per dose, with the exception of the 10000 mg/kg in which 7 females were dosed

Control animals
no
Details on study design
- Duration of observation period following administration: 14 days
- Frequency of observations and weighing: rats were observed frequently on the day of treatment and twice daily for the rest of the study period. The animals were weighed at the beginning of the study and on day 7 and day 14.
- Necropsy of survivors performed: yes, Necropsies were performed on all animals that died during the study and on the surviving animals at the end of the study.
- Other examinations performed:
Statistics Method of Litchfeld Wilcoxon (1949)
Results and discussion
Effect levels
Sex male
Dose descriptor LD50
Effect level ca. 4950 mg/kg bw
95% CL 3960 6188
Sex female
Dose descriptor LD50
Effect level ca. 6250 mg/kg bw
95% CL 5274 7406

Mortality

Range finding		
Males mg/kg bw	Deaths	Day
5000	2/2	1 and 2
1500	0/2	
1000	0/2	
Females mg/kg bw	Deaths	Day
5000	0/2	
1500	0/2	
1000	0/2	

Main study		
Males mg/kg bw	Deaths	Day
6810	7/8	1
4640	3/8	2
3160	0/8	
2150	0/8	
1470	0/8	
Females mg/kg bw		
10000	7/7	0 (5 deaths), 1 (2 deaths)
6810	5/8	1
4640	0/8	
3160	0/8	
2150	0/8	

Clinical signs
At dose levels greater than 2150 (male) and 4640 (female) mg/kg bw ataxia was seen and at dose levels greater than 3160 (male) and 2150 (female) mg/kg bw signs observed included decreased motor activity, yellow semi-solid discharge from the anus and yellow wet fur around the inguinal and perianal regions in male and female rats, respectively.
Gross pathology
The animals that died during the study showed discoloration of the thoracic and abdominal organs. Necropsy findings among surviving animals consisted of one male rat at 4640 mg/kg bw which exhibited a slightly mottled right kidney.**Conclusions**
Conclusions: The test material Sodium Chlorate, was evaluated for its acute oral toxicity potential in rats. The acute oral LD50's of Sodium Chlorate were determined to be ca. 4950 mg/kg in males (95 % Confidence limits: 3960 to 6188) and ca. 6250 mg/kg in females (confidence limits: 5274 to 7406).
Executive summary
The study was equivalent to OECD Guideline 401 (Acute Oral Toxicity). The study wasn't designed and performed according to Good Laboratory Practice Standards, there was, however, a quality control in house. The test material, Sodium Chlorate, was evaluated for its acute oral toxicity potential in Charles River CD rats. Twelve animals were used in a range finding study (dose levels: 5000, 1500 and 1000 mg/kg bw). During the main study Sodium Chlorate was administered as gavage doses at levels of 10000, 6810, 4640, 3160, 2150 and 1470 mg/kg to 8 males and 8 females per dose group, with the exception of 10000 mg/kg dose in which 7 females were dosed and 1470 mg/kg dose in which 8 males were dosed. Mortality occurred in 10 males dosed at the 4640 mg/kg and 6810 mg/kg level. In total 12 females died at the 6810 mg/kg level and the 10000 mg/kg level. Clinical signs of toxicity included ataxia at dose levels greater than 2150 (males) and 4640 (females) mg/ kg bw. At dose levels greater than 3160 (male) and

2150 (female) mg/kg bw signs of decreased motor activity, yellow semi-solid discharge from the anus and yellow wet fur around the inguinal and perianal regions were observed. The animals that died during the study showed discoloration of the thoracic and abdominal organs. Necropsy findings among survivors consisted of one male rat at 4640 mg/kg bw which exhibited a slightly mottled right kidney. There was a small gain in body weight in animals surviving to termination.
Conclusions: The test material Sodium Chlorate, was evaluated for its acute oral toxicity potential in rats. The acute oral LD50's of Sodium Chlorate were determined to be ca. 4950 mg/kg in males (95% Confidence limits: 3960 to 6188) and ca. 6250 mg/kg in females (confidence limits: 5274 to 7406).

1.3. Study 3 (Study report 1971)

ENDPOINT_STUDY_RECORD: 7775-09-9, Acute toxicity: oral, Sheahan, 1971, SS

Reliability
3 (not reliable)
Rationale for reliability incl. deficiencies
other: Reliable with restriction, test was not performed under GLP or according to a standard method.
Only 1 or 2 animals were tested per dose. An LD50 cannot be obtained.

Data source
Reference Experimental sodium chlorate poisoning in dogs. / Sheahan B.J., Pugh, D.M., Winstanley, E.W. /
Publication

Materials and methods
Principles of method if other than guideline Method: other
GLP compliance No

Test animals
Species dog other species
Strain other: collie and boxer
Sex not specified

Details on test animals and environmental conditions
TEST ANIMALS
- Source: no data
- Age at study initiation: collie 1-2 years, boxer 5 years
- Weight at study initiation: collies 11-15 kg, boxer 22 kg

Administration / exposure
Vehicle other: gelatin capsules
Details on oral exposure
MAXIMUM DOSE VOLUME APPLIED: no data
Doses
one dog: 0.5 g/kg bw
two dogs: 1 g/kg bw
one dog: 2 g/kg bw
an additional dog received 0.5 g/kg in a 1.59% solution by slow i.v injection.
No. of animals per sex per dose In total 5 aminals were used
Control animals no
Details on study design
- Frequency of observations: venous blood samples were taken before dosing, at 1, 3, 5 h after dosing and at daily intervals. Routine haematological and blood urea, total plasma bilirubin, plasma sodium, potassium and chloride measurements were taken. Urine was collected. ASAP after death theanimals were examined.

Results and discussion
Mortality
- Time of death: 12-20 h after dosing
- Number of deaths at each dose: On of the the 1 g/kg bw dogs died (boxer) and the 2 g/kg bw dog died (collie)
Clinical signs
All dogs vomited for about 1 h after dosing. Two dogs became excitable during the first 3 h after exposure. After 5h they became depressed and tachycardia was present. The mucous membranes became cyanotic and turned brown later on.

Other findings
In all dogs moderate degrees of anisocytosis, poikilocytosis, rouleax formation and neutrophilic leucocytosis were seen after 5h. Methamoglobinemia and nephropathy were shown. Peak plasma concentrations were observed between 1 and 3 hours post dosing, and after 7 hours in the animal with nephritis. Chlorate was detected in the urine as early as 1½ hour. No post-mortem lesions were detected in the surviving dogs killed after the observation period. Sodium chlorate converts hemoglobin to methemoglobin slowly,

and together with the probability of a more rapid elimination of the chlorate following i.v. administration, this may explain why a higher methemoglobinemia was not achieved in this dog.

1.4. Study 4 (Study report 1970)

ENDPOINT_STUDY_RECORD: 7775-09-9, Acute toxicity: oral, Ben-Dyke, 1970, SS

Reliability
4 (not assignable)
Rationale for reliability incl. deficiencies
other: No primary source. Data in table without further information

Data source
Reference
Acute toxicity data for pesticides. / Ben-Dyke, R., Sanderson, D.M., Noakes, N. / publication
Materials and methods
GLP compliance No

Results and discussion
Effect levels
Dose descriptor LD50
Effect level 1200 - 7000 mg/kg bw

1.5. Study 5 (Study report 2011)

ENDPOINT_STUDY_RECORD: 7775-09-9, Acute toxicity: oral, AFSSA - IN FRENCH, 2011, SS

Administrative data
Reliability
other: review
Rationale for reliability incl. deficiencies
other: Review of poison control center data in France in relation to poisoning incidents with sodium chlorate and toxicity in humans
Data source
Reference
Expositions au Chlorate de sodium enregistrées dans la BNCI Analyse des données des Centres antipois... / AFFSA / other: AFFSA review
Applicant's summary and conclusion
Conclusions
1> The classification for acute toxicity: according to the test in rats the LD50 is greater than 2000 mg / kg and the product is not classified. The interpretation of these human data in terms of death is very difficult indeed reflect certain death at doses much lower than others that have led to few symptoms and occurred in some cases with very low methaemoglobinaemia (other associated toxicity?). If one considers that 50% methemoglobinemia starts to induce signs that may lead to death in the absence of treatment, the dose would be about 20 g / kg for the whole population and 4.5 g / kg for the most sensitive part is the same range as rats.
2> Limit values:
a. Acceptable daily intake (ADI) is proposed for 0045 mg / kg / day or 4200 times less than the dose inducing 3% Methemoglobinemia in the general population. The occurrence of methemoglobinemia in these circumstances, even in cases of repeated dose seems unlikely.
b. The acceptable dose for the operator proposed is 0.35 mg / kg / day or 420 times less than the dose inducing 3% ethemoglobinemia in the general population. The occurrence of methemoglobinemia in these circumstances, repeated dose seems unlikely.

2. ACUTE TOXICITY INHALATION

2.1. Study 1 (Study report 2010a)

ENDPOINT_STUDY_RECORD: 3811-09-4, Acute toxicity: inhalation, Study report, 2010a, RS

Reliability
1 (reliable without restriction)
Rationale for reliability incl. deficiencies
other: performed accoding to OECD test guidelines and GLP compliant

Data source
Reference Assessment of acute inhalation toxicity with potassium chlorate in the rat (acute toxic class method... /
/ study report

Materials and methods
Test guideline Qualifier according to

Guideline OECD Guideline 436 (Acute Inhalation Toxicity: Acute Toxic Class Method)
Deviations no
GLP compliance yes (incl. certificate)
Test type acute toxic class method
Limit test Yes
Test animals

Species rat common species
Strain Wistar rat
Sex male/female
Details on test animals and environmental conditions
Species Rat: Crl:WI(Han) (outbred, SPF-Quality)
The rat is recognised by international guidelines as the recommended test system (e.g. OECD, EC).
Source: Charles River Deutschland, Sulzfeld, Germany.
Number of animals 3 males and 3 females (females were nulliparous and non-pregnant).
Age and body weight Young adult animals were selected (approximately 11 weeks old).
Animals used within the study were of approximately the same age and body weight variation did not exceed +/- 20% of the sex mean.
Identification Earmark
Health inspection A health inspection was performed prior to commencement of treatment to ensure that the animals were in a good state of health.
Animal husbandry
Conditions
Animals were housed in a controlled environment, in which optimal conditions were considered to be approximately 15 air changes per hour, a temperature of 21.0 ± 3.0˚C (actual range: 19.9 – 21.4C), a relative humidity of 30-70% (actual range: 31 - 77%) and 12 hours artificial fluorescent light and 12 h ours darkness per day.
Cleaning procedures in the room might have caused the temporary fluctuations above the optimal maximum level of 70% for relative humidity. Based on laboratory historical data, these fluctuations were considered not to have affected the study integrity. During exposure the temperature and relative humidity were 19.8 to 21.1C (mean 20.8 ± 0.4 C) and 21 to 50% (mean 29 ± 9%) respectively.
Accommodation
Animals were housed in groups of 3 animals per sex per cage in labelled Macrolon cages (type IV; height 18 cm) containing sterilised sawdust as bedding material (Litalabo, S.P.P.S., Argenteuil, France) and paper as cage-enrichment (Enviro-dri, Wm. Lillico & Son (Wonham Mill Ltd), Surrey, Un ited Kingdom). After exposure, animals were placed in a cage with a stainless steel grid and the bedding and cage enrichment were withheld . At the end of the Day of exposure animals were housed as described above.
Acclimatisation period was at least 5 days before start of treatment under laboratory conditions. Animals were housed with maximally 5 animals per cage per sex as described above.
Diet: Free access to pelleted rodent diet (SM R/M-Z from SSNIFF® Spezialdiäten GmbH, Soest, Germany) except during exposure to the test substance.
Water: Free access to tap water except during exposure to the test substance. Results of analysis for each batch of diet (nutrients and contaminants), sawdust, paper and water were assessed and did not reveal any findings that were considered to have affected the study integrity. All certificates and results of analysis are retained in the NOTOX archives.
Administration / exposure
Route of administration inhalation: dust
Type of inhalation exposure nose only
Vehicle other: unchanged (no vehicle)
Details on inhalation exposure
Animals were exposed to the test substance via the inhalatory route. For this purpose the animals were placed in restraining tubes, connected to the exposure chamber. The design of the exposure chamber was based on the flow past nose-only inhalation chamber (Am.Ind. Hyg Assoc. J. 44(12): 923-928, 1983). The chamber consisted of 3 animal sections with 8 animal ports each. The number of animal sections and number of open animal ports were adapted to the air flow in such a way that at each animal port the theoretical air flow was on average 1.4 L/min, which ensures an adequate oxygen supply to the test animals. The inlet of the test atmosphere was located at the top section and the outlet was located at the bottom section. The direction of the flow of the test atmosphere guaranteed a freshly generated atmosphere for each individual animal. The placement of the individual animals in the inhalation chamber is shown in figure 2. All components of the exposure chamber, which could come in contact with the test material, were made of stainless steel, glass, rubber or plastic. To avoid exposure of the personnel and contamination of the laboratory the exposure chamber was placed in a fume hood, which was maintained at a slightly negative pressure.
Analytical verification of test atmosphere concentrations no
Duration of exposure 4 h
Concentrations 5.1 mg/L
No. of animals per sex per dose 3 males / 3 females
Control animals no
Details on study design
Potassium chlorate was administered as an aerosol by inhalation for a single but interrupted exposure lasting 4 hours and 8 minutes in total to one group of three male and three female Wistar rats. Animals were subjected to daily observations and weekly determination of body weight. Macroscopic examination was performed after terminal sacrifice (day 15).
Any other information on materials and methods incl. tables

The mean time-weighed actual concentration was 5.1 ± 0.3 mg/L. The nominal concentration was 144 mg/L. The generation efficiency (ratio of actual and nominal concentration) was 3.5%. The Mass Median Aerodynamic Diameter (MMAD) and geometric standard deviation (gsd) were determined twice during exposure. The MMAD was 4.0 mm and 4.4 mm respectively and the gsd was 1.9 and 1.8 respectively. Agglomeration of the aerosol at this high concentration might have caused the MMAD for the second measurement to fall outside the recommended range of 1 – 4µm. Since the MMAD was at or close to the upper limit of 4µm and since the gsd was sufficiently large, it was considered that adequate deposition in the lower respiratory tract occurred.

Results and discussion
Sex male/female
Dose descriptor LC50
Effect level > 5.1 mg/L air
Exp. Duration 4 h
Mortality No mortality occurred.
Clinical signs no clinical signs were noted during the study.
Body weight Overall body weight gain was within the range expected for rats of this strain and age used in this type of study.
Gross pathology No abnormalities were found at macroscopic post mortem examination of the animals.

Applicant's summary and conclusion
Conclusions
The inhalatory LC50, 4h value of potassium chlorate in Wistar rats was established to exceed 5.1 mg/L.
Executive summary
Assessment of acute inhalation toxicity withpotassium chloratein the rat (acute toxic class method) The study was carried out based on the guideline described in Organisation for Economic Co-operation and Development (OECD), OECD Guidelines for Testing of Chemicals, Section 4, Health Effects. No.436, "Acute Inhalation Toxicity-Acute Toxic Class Method", September 2009. Potassium chloratewas administered as an aerosol by inhalation for a single but interrupted exposure lasting 4 hours and 8 minutes in total to one group of three male and three female Wistar rats. Animals were subjected to daily observations and weekly determination of body weight.Macroscopic examination was performed after terminal sacrifice (day 15). The mean time-weighed actual concentration was 5.1 ± 0.3 mg/L. The nominal concentration was 144 mg/L. The generation efficiency (ratio of actual and nominal concentration) was 3.5%. The Mass Median Aerodynamic Diameter (MMAD) and geometric standard deviation (gsd) were determined twice during exposure. The MMAD was 4.0 mm and 4.4 mm respectively and the gsd was 1.9 and 1.8 respectively. Agglomeration of aerosol particles at this high concentration might have resulted in these higher MMAD values, causing the second measurement to fall outside the recommended range of 1 – 4µm. Since the MMAD values were at or close to the upper limit of 4µm and since the gsd was appropriate (i.e. between 1.5 and 3), it can be assumed that deposition of particles in the lower respiratory tract had occurred. No mortality occurred andno clinical signs were noted during the study. Overall body weight gain was within the range expected for rats of this strain and age used in this type of study. No abnormalities were found at macroscopic post mortem examination of the animals. The inhalatory LC50, 4hvalue ofpotassium chloratein Wistar rats was established to exceed 5.1 mg/L.

3. SHORT-TERM TOXICITY TO FISH

3.1. Study 1 (Study report 1991a)

ENDPOINT_STUDY_RECORD: 7775-09-9, Short-term toxicity to fish, Study report,1991a,RS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies

other: see 'Remark' Study generated according to valid and internationally accepted testing guideline and performed under GLP.

No chemical analyses were performed on the test solution, only the stock solution could be analyzed. Because of analytical results from chronic tests it can be assumed that the test concentrations were stable and the fish were exposed properly.

Data source

Reference: Acute flow-through toxicity of sodium chlorate to the rainbow trout, Oncorhynchus mykiss. / study report

Materials and methods

Test performed according to Guideline EPA OPP 72-1 (Fish Acute Toxicity Test)

GLP compliance: yes

Test material:

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis:

Analytical monitoring, no

Details on sampling:

Analysis conducted on stock only.

Details on analytical methods:

Conductivity detector. The test concentrations could not be analyzed because of too much interference of compounds in the dilution water. Only stock solutions were analyzed. Interfering compounds, chlorate and nitrate present in the natural groundwater used as dilution water, interfered with the analysis by eluting near the sodium chlorate peak. The concentration of sodium chlorate in test media could therefore not be determined because of naturally occurring interference in the dilution water. The available analytical methods were effective in deionized water used to prepare toxicant stock solutions but not in the dilution water. Analysis of the stock only was conducted.

Test solutions

Vehicle: no

Details on test solutions:

Preparation: 600,000 mg/L stock solution was prepared by adding 1200.0 g test substance to 2000 ml deionized water. Appropriate amounts of stock were added directly to dilution water by a proportional diluter. The dilution water was filtered natural groundwater collected at Hampton, New Hampshire.

Test organisms

Test organisms (species): Oncorhynchus mykiss (previous name: Salmo gairdneri)

Details on test organisms:

Supplier: Aquatic Research Organisms, Hampton, New Hampshire

- Wild caught: No

- Age/weight/loading: juvenile, 0.57 g on average, 0.38 g/L

- Feeding: fish food (EnviroSystems lot number TM02) once or twice daily

- Pretreatment: acclimatized for 14 days under test conditions.

- Feeding during test: no, feeding stopped 48 hours prior to the test

Study design

Test type: flow-through

Water media type: freshwater

Limit test: no

Total exposure duration: 96 h

Post exposure observation period

Pretreatment: acclimatized for 14 days under test conditions

Test conditions

Test temperature: 11.0-11.6 °C

pH: 6.8-7.3

Dissolved oxygen: 9.0-9.8 mg/L

Nominal and measured concentrations: 0 mg/L (control), 150 1 240, 380, 600, and 1,000 mg/L. (Nominal)

Details on test conditions

- Test type: flow-through, 9.0 media exchanges per 24 hours in each test vessel.

- Exposure vessel type: 20 L glass aquaria that contained 15 L of test solution

- Number of replicates, fish per replicate: 2, 10

- Conductivity: 490-1200 µmhos/cm (equal to µS/cm) at the start of the test and 530-680 µmhos/cm

(equal to µS/cm) at the end

- Intensity of irradiation: cool white fluorescent lights with an intensity of 12 µEs/m2

- Photoperiod: 16 hours photoperiod daily

- Nominal test concentrations: 0, 150, 240, 380, 600, 1000 mg/L. Measured concentration of the

stock solution was 630,000 mg/L

Dilution water:

– Source: groundwater collected from wells in Hampton, New Hampshire

- Aeration: Yes

- Hardness: 48 mg CaCO3/L

- pH: 6.8

- Conductance: 490 µmhos/cm (equal to µS/cm)

- Holding water: same as dilution water

Reference substance (positive control): no

Any other information on materials and methods incl. tables

Test animals were acclimated at EnviroSystems under test conditions for more than 14 days. After 96 hours of exposure the control organisms had an average wet weight (blotted dry) of 0.57 g. All animals were in good condition at the beginning of the study.

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.	Nominal / measured	Basis for effect
96 h	NOEC	600 mg/L	nominal	mortality
96 h	LC50	> 1000 mg/L		

Details on results

mortality and loss of equilibrium, erratic swimming, loss of reflex, excitability, discoloration or change in behavior.

- Mortality: At 72 hours 1 fish was dead in 1000 mg/l

- Affected: At 72 hours 2 and 4 fish were lethargic in the 2 replicates of 1000 mg/l. At 96 hours only 1

and 2 fish respectively showed these effects in the same concentration.

- No effects were observed in the control, all fish were normal.

Results with reference substance (positive control)

Not Reported

Reported statistics and error estimates

No statistics could be performed because greater than 50% survival occurred in all test concentrations.

Applicant's summary and conclusion

Validity criteria fulfilled: no

The test concentrations could not be monitored, only the stock solution was analyzed.

Conclusions

Study performed to EPA guidelines under flow through conditions with GLP accreditation. Stocks were analyzed but analysis in the test media was not possible to interference. The stock recovery was 105%. Due to the test substance stability and the fact that the solution is continually renewed and providing the automatic diluting system used was working accurately the nominal concentrations can be considered reliable. The LC50 of >1000mg/l is considered reliable without major restrictions.

Executive summary

Exposure of rainbow trout to the test substance resulted in a 96 hour LC50 greater than 1,000 mg/L sodium chlorate. The 96 hour no observed effect concentration was 600 mg/L sodium chlorate.

3.2. Study 2 (study report 1991c)

ENDPOINT_STUDY_RECORD: 7775-09-9,Short-term toxicity to fish, Study report,1991c,RS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies

Study generated according to generally valid and internationally accepted testing guideline and performed under GLP. No analytical monitoring was performed, only the stock solution was analyzed, but based on chronic tests it can be assumed that the test substance was stable during the test and the fish were properly exposed.

Data source

Reference:

Acute flow-through toxicity of sodium chlorate to the sheepshead minnow, Cyprinodon variegatus / study report

Materials and methods

Test guideline:

Test performed according to Guideline EPA-FIFRA, guideline 72-3

Deviations: no

GLP compliance: yes

Test material

Test material information: sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: no

Details on analytical methods:

Conductivity detector. The test concentrations could not be analyzed because of too much interference of compounds in the dilution water. Only stock solutions were analyzed at the beginning of the test.

Test solutions

Vehicle: no

Details on test solutions

- Preparation: 600,000 mg/L stock solution was prepared by adding 1200.0 g test substance to 2000 ml deionized water. Appropriate amounts of stock were added directly to dilution water by a proportional diluter.

Test organisms

Test organisms (species): Cyprinodon variegatus

Details on test organisms:

- Supplier: The aquatic research organisms division of resource analysts, Inc., Hampton, New Hampshire

- Wild caught: No

- Age/weight/loading: juvenile

- Feeding: fish food (Enviro Systems lot number TM02) once or twice daily

- Pretreatment: acclimatized for more than 7 days under test conditions.

- Feeding during test: no, feeding stopped 48 hours prior to the test

Study design

Test type: flow-through

Water media type: saltwater

Limit test: no

Total exposure duration: 96 h

Test conditions

Test temperature: 21.8-22.9 °C

pH: 7.6-7.8

Dissolved oxygen: 7.4-8.1 mg/L

Salinity: 15-17 ppt

Nominal and measured concentrations: Nominal concentrations: 0, 140, 240, 380, 600, 1000 mg/L

Details on test conditions

DILUTION WATER:

- Source: unfiltered sea water collected from Atlantic ocean in Hampton, New Hampshire
- Aeration: Yes
- Salinity: 16 ppt (parts per thousand)
- pH: 7.6
- Holding water: same as dilution water

TEST SYSTEM:

- Test type: 7.8 media exchanges per 24 hours in each test vessel.
- Exposure vessel type: 20 L glass aquaria that contained 15 L of test solution
- Number of replicates, fish per replicate: 2, 10
- Intensity of irradiation: cool white fluorescent lights with an intensity of 12 µEs/m2
- Photoperiod: 16 hours photoperiod daily

Reference substance (positive control); no

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.		
96 h	NOEC	1000 mg/L		
96 h	LC50	> 1000 mg/L		

Details on results

Measured concentration of the stock solution was 600,000 mg/L

Effect data (Mortality): In the control and in all test concentrations no mortality and no sublethal effects were observed.

Reported statistics and error estimates

No statistics could be performed because greater than 50% survival occurred in all test concentrations.

Applicant's summary and conclusion

Validity criteria fulfilled: no, the test substance concentration could not be monitored

Conclusions

only the stock solution could be analyzed, but based on chronic tests for which chemical analyses were performed it can be assumed that the test concentrations were stable and that the fish were exposed properly. No fish died during the test, therefore the 96h-LC50 is greater than 1000 mg/l and the NOEC is 1000 mg/l.

Executive summary

The acute toxicity of sodium chlorate to the sheepshead minnow, Cyprinodon variegatus, is described in this final report. The test was conducted for Albright and Wilson Americas for 96 hours during February 19 to 23, 1991, at the EnviroSystems Division of Resource Analysts, Inc. in Hampton, New Hampshire. It was conducted by Peter Kowalski, Ellen Stanford, Jeanne Magazu, Robert Boeri, and Timothy Ward according to the protocol developed for EnviroSystems Study Number 90115-DE. The analytical portion of this study was conducted under the supervision of Gloria Switalski. Sodium chlorate (reported purity >99% active ingredient) was supplied by the sponsor.

The test was performed under flow-through conditions with five concentrations of test substance and a dilution water control at a temperature of 22 ± 1°C. The dilution water was unfiltered natural seawater collected at Hampton, New Hampshire. Mean nominal concentrations of sodium chlorate were: 0 mg/L (control), 140, 240, 380, 600, and 1,000 mg/L. Nominal concentrations were used for all calculations.

Organism used in the test were procured from a commercial supplier (Aquatic Research Organisms, Hampton, New Hampshire) and acclimated at EnviroSystems under test conditions for more than 7 days. After 96 hours of exposure the control organisms had an average wet weight (blotted dry) of 0.70 g. All animals were in good condition at the beginning of the study.

Exposure of sheepshead minnow to the test substance resulted in a 96 hour LC50 greater than 1,000 mg/L sodium chlorate. The 96 hour no observed effect concentration was 1,000 mg/L sodium chlorate.

3.3. Study 3 (study report 1991h)

ENDPOINT_STUDY_RECORD: 7775-09-9,Short-term toxicity to fish, Study report,1991h,SS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies:

other: Study generated according to generally valid and internationally accepted testing guideline and performed under GLP. No chemical analyses were performed, but the test substance is considered to be stable.

Data source

Reference:

Acute toxicity of sodium chlorate to Brachydanio rerio. / study report

Materials and methods

Test guideline:

Test performed according to Guideline OECD Guideline 203 (Fish, Acute Toxicity Test)

GLP compliance: yes

Test material:

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: no

Test organisms

Test organisms (species): Danio rerio (previous name: Brachydanio rerio)

Study design

Test type: semi-static

Limit test: yes

Any other information on materials and methods incl. tables

A limit test was performed at 1000 mg/l. Two times 7 fish were tested at a concentration of 1000 mg/l.

Results and discussion

Effect concentrations:

Duration	Dose descriptor	Effect conc.

96 h	LC50	> 1000 mg/L

Any other information on results incl. tables

No fish died and no abnormalities in behavior were observed.

3.4. Study 4 (Study report 1991b)

ENDPOINT_STUDY_RECORD: 7775-09-9,Short-term toxicity to fish, Study report,1991b,SS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies: other: Study generated according to generally valid and internationally accepted testing guideline and performed under GLP.

Data source

Reference: Acute flow-through toxicity of sodium chlorate to the bluegill sunfish Lepomis macrochirus. / study report

Materials and methods

Test guideline:

Test performed according to Guideline EPA OPP 72-1 (Fish Acute Toxicity Test)

GLP compliance: yes

Test material:

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: no

Test organisms:

Test organisms (species): Lepomis macrochirus

Study design

Test type: flow-through

Any other information on materials and methods incl. tables

ANALYTICAL METHODS: Conductivity detector. The test concentrations could not be analyzed because of too much interference of compounds in the dilution water. Only stock solutions were analyzed.

TEST ORGANISMS: Lepomis macrochirus

- Supplier: Aquatic Research Organisms, Hampton, New Hampshire

- Wild caught: No

- Age/weight/loading: juvenile

- Feeding: fish food (EnviroSystems lot number TM01) once or twice daily

- Pretreatment: acclimatized for more than 14 days under test conditions.

- Feeding during test: no, feeding stopped 48 hours prior to the test.

STOCK AND TEST SOLUTION:

- Preparation: 600,000 mg/L stock solution was prepared by adding 1200.0 g test substance to 2000 ml deionized water. Appropriate amounts of stock were added directly to dilution water by a proportional diluter.

DILUTION WATER:

- Source: groundwater collected from wells in Hampton, New Hampshire

- Aeration: Yes

- Hardness: 48 mg CaCO3/L
- pH: 8.7
- Conductance: 1500 µmhos/cm (equal to µS/cm)
- Holding water: same as dilution water

TEST SYSTEM:
- Test type: flow-through, 6.3 media exchanges per 24 hours in each test vessel.
- Exposure vessel type: 20 L glass aquaria that contained 15 L of test solution
- Number of replicates, fish per replicate: 2, 10
- Test temperature: 21.1-22.9 °C
- Dissolved oxygen: 8.3-9.4 mg/L
- pH: 8.0-8.7
- Conductivity: 900-2000 µmhos/cm (equal to µS/cm)
- Intensity of irradiation: cool white fluorescent lights with an intensity of 10 µEs/m2
- Photoperiod: 16 hours photoperiod daily

STATISTICAL METHODS: No statistics could be performed because greater than 50% survival occurred in all test concentrations.

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.		
96 h	NOEC	1000 mg/L		
96 h	LC50	> 1000 mg/L		

Any other information on results incl. tables

EXPOSED
- Nominal concentrations: 0, 150, 240, 380, 600, 1000 mg/L. Measured concentration of the stock solution was 620,000 mg/L

CONTROL
- Number/percentage of animals showing adverse effects: No mortality or other effects were observed in the control.

3.5. Study report 5 (Study report 1993)

ENDPOINT_STUDY_RECORD: 7775-09-9,Short-term toxicity to fish, Study report,1993, SS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies:

other: Study generated according to generally valid and internationally accepted testing guideline and performed under GLP. No chemical analyses were performed, but the test substance is considered to be stable.

Data source

Reference:

Acute toxicity of sodium chlorate to Pimephales promelas. / study report

Materials and methods

Test guideline:

Test performed according to OECD Guideline 203 (Fish, Acute Toxicity Test)

GLP compliance: yes

Test material:

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: no

Test organisms

Test organisms (species): Pimephales promelas

Study design

Test type: semi-static

Limit test: yes

Any other information on materials and methods incl. tables

A limit test was performed at 1000 mg/l. Two times 7 fish were tested at a concentration of 1000 mg/l.

Results and discussion

Effect concentrations:

Duration	Dose descriptor	Effect conc.
96 h	LC50	> 1000 mg/L

Any other information on results incl. tables:

No fish died and no abnormalities in behavior were observed.

3.6. Study 6 (Toussaint et al. (2001)

ENDPOINT_STUDY_RECORD: 7775-09-9,Short-term toxicity to fish, Toussaint-Brennan, 2001, SS

Reliability

2 (reliable with restrictions)

Rationale for reliability incl. deficiencies:

other: No standard test and no GLP, but test described in enough details.

Data source

Reference: Acute toxicity of four|drinking water disinfection by-products to Japanese medaka fish. / Toussaint, M.W.,Brennan, L.M., Rosencrance, A.B., Dennis, W.E., Hoffmann, F.J., Gardner, H.S. Jr. / publication

Materials and methods

Test material:

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: yes

Test organisms

Test organisms (species): Oryzias latipes

Any other information on materials and methods incl. tables

ANALYTICAL METHODS: Dionex 500 series Ion Chromatograph (IC) equipped with conductivity detector, autosampler, anion self generating suppressor ASRS-1 4 mm and Dionex Peak Net Data system. A Dionex IonPac(R) AS14 4x250 mm column with an IonPac(R) AG14 guard column was used for the separation.

Dionex method 5.9 Isocratic analysis of selected oxyanions was used to determine chlorate concentrations. Briefly, the mobile phase consisted of 2.7 mM sodium carbonate/1 mM sodium bicarbonate buffer in reagent grade water. A flow rate of 1.2 milliliters / minute was used. The injection volume was 10 microliters. Samples (10 mL each) were collected in Nalgene plastic 60 mL bottles. The detection limit was 10 mg/L . The average percent recovery for chlorate was 99%.

TEST ORGANISMS: Medaka fish

- Supplier: inhouse culture facilities

- Wild caught: no

- Age/weight/loading: 14 +/- l day old

- Feeding: -

- Pretreatment: -

- Feeding during test: -

STOCK AND TEST SOLUTION:

- Preparation: Stock solutions of 64 g/L chlorate were prepared in a glass carboy by stirring for 24 h prior to use.

DILUTION WATER:

- Source: A hard groundwater was processed through a softener, reverse osmosis, blended with raw groundwater, carbon filtered, particle filtered, and sterilized with ultraviolet light before being used as the dilution water

- Aeration: -

- Hardness: -

- pH:-

- Conductance: -

- Holding water: -

TEST SYSTEM:

- Test type: flow through;Test solution was replenished in the aquaria by the proportional diluter every 6 rnin 30 sec +/-30 sec at a rate of 300 * 15 mL per cycle throughout the 96 h test.

- Exposure vessel type: The tests were conducted within mesh sided polypropylene cylinders (water column = 9 cm x 17 cm; mesh = 32 x 32 linedih) immersed in 5 gallon glass aquaria that were sealed with glass hinged tops.

- Number of replicates, fish per replicate: 2, 10

- Test temperature: 25 +/- 1 Degrees Celcius.

- Dissolved oxygen: 7.0 mg/L

- pH: 7.9-8.0

- Conductivity: 10,300 (microhos/cm)

- Intensity of irradiation: Light intensities averaged 733 lux

- Photoperiod: Iight/dark cycle of 16/8 h

STATISTICAL METHODS: median lethal concentration was estimated using probit analysis.

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc
96 h	LC50	2585 mg/L

Any other information on results incl. tables

EXPOSED

- Nominal concentrations: from a range finding study 0 1037 1728 2880 4880 8000 mg/L were selected. Corresponding to <10 1263 1738 2675 4776 8309 mg/L as measured concentrations

Effect data (Mortality): 96h LC50 2585 (95% CI 1925-3487) mg/L

CONTROL

- Number/percentage of animals showing adverse effects: -

4. LONG-TERM TOXICITY TO FISH

1.1 2.1 Study 1 (study report 2004a)

ENDPOINT_STUDY_RECORD: 7775-09-9,Long-term toxicity to fish, Study report, 2004a,RS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies:

other: Study generated according to valid and internationally accepted testing guideline and was performed under GLP.

Data source

Reference: Chronic toxicity of sodium chlorate to Danio rerio in an early-life stage toxicity test under flow-t... / study report

Materials and methods

Test guideline:

Test performed according to OECD Guideline 210 (Fish, Early-Life Stage Toxicity Test)

GLP compliance: yes

Test material:

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: yes

Details on sampling

first at preparation, then weekly and at the end of the test, for all test concentrations in duplicate. The samples were filter sterilised over a 0.22 µm filter. Details on analytical methods Water samples were analysed for chlorate, chlorite and chloride by ion chromatography using (suppressed) conductivity detection. => Remark: Only chlorate was analysed because the Daphnia magna 21 d reproduction test showed that chlorate was completely stable during the test. A Dionex DX-120 ion chromatograph equipped with an ASS-HC 4 mm analytical column, an AG9-HC 4 mm guard column, a 50 pL loop, an ASRS-ultra 4 mrn and a CDM-3 flow through conductivity cell with a DS4 detection stabilizer was used to detect and quantify chlorate.

The DX-120 was operated at a column temperature of 20 "C, a detector temperature of 35 "C and an eluent flow rate of 1.0 m/lmin. The eluent composition was 9.0 mM Na2C03. Data was acquired and integrated using a Thermo Labsystems Chromatography Server and Atlas 2002 version 6.18. Samples were loaded using a Dionex AS40 automated sampler with 0.5 rnl vials.

Test solutions

Vehicle: no

Details on test solutions

Procedure: 1.28, 3.2 and 20 g of test substance were weighed and dissolved in 5, 5 and 2 L respectively of DSW.

The stock solutions were agitated for 1 to 5 hours to dissolve the test substance.

Test solutions were prepared by further diluting the stock solutions.

Test organisms:

Test organisms (species): Danio rerio (previous name: Brachydanio rerio)

Details on test organisms

- Supplier: Dierenspeciaalzaak Engelen Arnhem, The Netherlands.

-The broodstock is maintained in Akzo Nobel Environmental Chemistry laboratory.

-Eggs, Akzo Nobel Environmental Chemistry laboratory

- Wild caught: No

- Post-hatch transfer time: about 45 minutes

- Age/loading: between zygote and blastodisc cleavage stage /

- maximum 1 g of biomass per liter of test solution per 24 hours and not exceeding 2 g per liter of test solution any time.

-Finally 20 larvae per replicate.

- Pretreatment: no

-Feeding during test: 4 days after hatching with Paramecia species, 3 times per day to provide in total 2490 Paramecia per fish per day. From day 28 on the fish were fed with brine shrimp nauplii.

Study design

Test type: flow-through

Water media type: freshwater

Limit test: no

Total exposure duration:36 d

Test conditions

Total hardness: 8.43-13.1 mg/L

Test temperature: 24.25-26.7 ºC, not more than +/- 1.5 ºC between successive days, except for two occasions during the test when maximum difference over a 24h time period was 1.85 and 2.05 ºC. This was not considered to have an impact on the validity of the test.

pH: 7.8-8.1

Adjustment of pH: No

Dissolved oxygen: 7.9-8.4 mg O2-/L

Nominal and measured concentrations:

- Nominal/measured test concentrations: 0, 12.8, 32, 80, 200 and 500 mg/L

- Nominal is equal to measured concentrations.

Details on test conditions

Dutch Standard Water:

- Hardness: approx. 97.5 mg/L as CaCO3

- pH: 6.0-8.5

- Oxygen content: >= 60% of the air saturation value

- Holding water: Dutch Standard Water

- Renewal of test solution: continuously; at least 5 volumes of test solution per day with a peristaltic pump.

- Exposure vessel type: 1.5 L glass aquaria

- Number of replicates, individuals per replicate: 2, test was started with 40 eggs per replicate, at day 1 it was reduced to 30 eggs and at day 6 the number was reduced to 20 individuals per replicate.

- Conductivity: control 507-630 µS/cm; 12.8 mg/L 517-639 µS/cm; 32 mg/L 537-658 µS/cm; 80 mg/L 574-713 µS/cm; 200 mg/L 685-814 µS/cm; 500 mg/L 976-1113 µS/cm.

- Intensity of irradiation: No information

- Photoperiod: 14 h of ambient light per day

Reference substance (positive control): no

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.	Nominal / measured	Conc. based on test mat.	Basis for effect
36 d	NOEC	>= 500 mg/L	nominal	other: all endpoints	

Details on results

Chemical analyses:

All test solutions were found to be stable over the test period. Concentrations were observed to be between 100 and 111% of the nominals.

- Hatching: hatching started on day 3 except for one fish at 12.8 mg/L that hatched on day 2. Hatching was complete by day 4. All fish hatched at all concentrations.

- Mortality: No concentration related mortality was observed at any concentration at any of the stages during the test. In the test concentrations less fish died compared to the control.

- Morphology and behaviour: No abnormalities were observed.

CONTROL

- Number/percentage of animals surviving: 36 out of 40 survived. The post-hatch success (until the end of the test) was greater than 70% in the control.

- Length: 0.62-1.39 cm

Reported statistics and error estimates:

Chi-square test and Shapiro Wilks test for normality. Hartley's and Bartlett's tests for homogeneity of variance. Bonferroni t-test and Dunnett's test was performed on weight and length data.

Overall remarks, attachments

Applicant's summary and conclusion

Validity criteria fulfilled: yes

Conclusions:

Test conducted under GLP with analysis, analysis certificate and carried out to relevant guideline. This study can be considered reliable without restrictions.

The NOEC is greater than or equal to 500 mg/l for all endpoints.

Executive summary

The purpose of this study was to assess the toxicity of the test substance dissolved in fresh water, on the early life stages of Danio rerio, in a 36-day flow-through test complying with the OECD Guideline No. 210,17 July 1992. The test criterion of toxicity used was the effects on hatching, larvae mortality, morphological abnormalities and growth of Danio rerio exposed to the test substance over the test period.

The nominal concentrations used in the study were as follows: 0, 12.8, 32.0, 80.0, 200 and 500 mg/l. Analytical determinations of the test solutions were made on six occasions during the test. The concentrations were found to remain stable to within 20% of the nominals. The nominal concentrations were used to calculate the effect concentrations.

The validity criteria were respected:

• the dissolved oxygen concentration was between 60 and 100% of the air saturation value throughout the test,

• water temperature remained between 23 and 27°C over the test period and did not differ more than ±1.5°C between successive days except on 2 occasions during the test when maximum difference over a 24 h period was 1.85 and 2.05°C respectively. This was not thought to have any impact on the validity of the study.

• The post-hatch success (until the end of the test) was greater than 70% in the control.

The No Observed Effect Concentration (NOEC) is determined as the concentration used in the study that is immediately below the Lowest Observed Effect Concentration (LOEC), the latter derived statistically from the data using the appropriate statistical test.

However, as all embryos hatched at the highest concentration tested of 500 mg/l as well as in the control and post-hatch mortality was less than that of the control, a statistical test was not used for this parameter and the NOEC was considered to be at or greater than the highest concentration tested. No teratogenic malformations were noted for any larvae at any concentration.

Based on results from weight and length, the Lowest Observed Effect Concentration (LOEC) cannot be calculated and the No Observed Effect Concentration (NOEC) was determined as greater than or equal to the highest concentration tested, 500 mg/l.

5. SHORT-TERM TOXICITY TO AQUATIC INVERTEBRATES

5.1. Study 1 (Study report 1991d)

Endpoint study record: 7775-09-9,Short-term toxicity to aquatic invertebrates, Study report,1991d,RS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies

other: see 'Remark' Study generated according to valid, internationally accepted testing guideline and was performed under GLP. No chemical analyses was performed on the test concentrations, only the stock solution could be analyzed. From chronic tests with chemical analyses it can be assumed that the test concentrations were stable and the animals were exposed properly.

Data source

Reference: Acute flow-through toxicity of sodium chlorate to the daphnid, Daphnia magna. /study report

Materials and methods

Test guideline

Test performed according to Guideline EPA OPP 72-2 (Aquatic Invertebrate Acute Toxicity Test)

GLP compliance: yes

Test material:

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: no

Details on analytical methods:

Conductivity detector. Conductivity detector. The test concentrations could not be analyzed because of too much interference of compounds in the dilution water. Only stock solutions were analyzed. Interfering compounds, chlorate and nitrate present in the natural groundwater used as dilution water, interfered with the analysis by eluting near the sodium chlorate peak. The concentration of sodium chlorate in test media could therefore not be determined because of naturally occurring interference in the dilution water. The available analytical methods were effective in deionized water used to prepare toxicant stock solutions but not in the dilution water. Analysis of the stock only was conducted.

Test solutions

Vehicle: no

Details on test solutions

- Preparation: 600.0 mg/L stock solution was prepared by adding 1200.0 g test substance to 2000 ml deionized water.

Appropriate amounts of stock were added directly to dilution water by a proportional diluter.

Test organisms

Test organisms (species): Daphnia magna

Details on test organisms:

- Supplier: Aquatic Research Organisms, Hampton, New Hampshire
- Age: less than 24 hours
- Feeding: yeast/trout chow, and/or Selenastrum capricornutum daily
- Pretreatment: acclimatized for 14 days under test conditions.
- Feeding during test: No

Study design

Test type: flow-through

Water media type: freshwater

Limit test: no

Total exposure duration: 48 h

Test conditions

Test temperature: 19.5-20.9 °C

pH: 7.3-7.7

Dissolved oxygen: 8.5-9.0 mg/L

Nominal and measured concentrations

Nominal test concentrations: 0, 150, 240, 380, 600, 1000 mg/L

Details on test conditions

DILUTION WATER:

- Source: groundwater collected from wells in Hampton, New Hampshire
- Aeration: Yes
- pH: 7.6
- Hardness: 180 mg CaCO3/L
- Conductance: 670 µmhos/cm (equal to µS/cm)
- Holding water: same as dilution water
- flow-through, 8.8 media exchanges per 24 hours in each test vessel
- Exposure vessel: 20 liter glass aquaria that contained 15 liter of test solution.
- 20 daphnids were confined in two groups of 10 in cages consisting of glass and Nitex screen.
- Number of replicates, individuals per replicate: 2, 10
- Conductance: 620-1500 µmhos/cm (equal to µS/cm)
- Intensity of irradiation: cool white fluorescent lights with an intensity of 13 µEs/m2
- Photoperiod: 16 hours photoperiod daily
- TEST PARAMETER: immobilization and sublethal effects

Reference substance (positive control): no

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.

48 h	NOEC	1000 mg/L
48 h	EC50	> 1000 mg/L

Details on results

- 24 hours: 1 organism died in 1000 mg/l

- 48 hours: 1 organism died in 150 mg/l and 1 in 240 mg/l

- No other sublethal effects were observed.

- Control: No affected or immobile organisms were observed

- The replicates mentioned in the report are no true replicates because the two cages were placed in the same aquarium.

Reported statistics and error estimates:

No statistics could be performed because greater than 50% survival occurred in all test concentrations.

Applicant's summary and conclusion

Validity criteria fulfilled: no, the test concentrations were not analyzed, only for the stock concentration chemical analyses was performed.

Conclusions

Study performed to EPA guidelines under flow through conditions with GLP accreditation. Stocks were analysed but analysis in the test media was not possible to interference. The stock recovery was within specified limits. Due to the test substance stability and the fact that the solution is continually renewed and providing the automatic diluting system used was working accurately the nominal concentrations can be considered reliable. The LC50 of >1000mg/l is considered reliable without major restrictions.

Executive summary

The acute toxicity of sodium chlorate to the daphnid, Daphnia magna, is described in this final report. The test was conducted for Albright and Wilson Americas for 48 hours during March 5 to March 7, 1991, at the EnviroSystems Division of Resource Analysts, Inc. in Hampton, New Hampshire. It was conducted by Peter Kowalski, Ellen Stanford, Jeanne Magazu, Robert Boeri, and Timothy Ward according to the protocol developed for EnviroSystems Study Number 90144-AW. The analytical portion of this study was conducted under the supervision of Gloria Switalski. Sodium chlorate (reported purity >99\ active ingredient) was supplied by the sponsor.

The test was performed under flow-through conditions with five concentrations of test substance and a dilution water control at a temperature of 20 ± 1°C. The dilution water was filtered natural groundwater collected at Hampton, New Hampshire. Nominal concentrations of sodium chlorate were: 0 mg/L (control), iSO, 240, 380, 600, and 1,000 mg/L. Nominal concentrations were used for all calculations.

Organism used in the test were procured from a commercial supplier (Aquatic Research Organisms, Hampton, New Hampshire) and acclimated at EnviroSystems under test conditions for more than 14 days. After 48 hours of exposure the control organisms had an average wet weight (blotted dry) of 0.0006 g. All animals were in good condition at the beginning of the study.Exposure of daphnids LC50 greater than 1,000 concentration. The 48 hour no observed effect concentration was 1,000 mg/L sodium chlorate.

5.2. Study 2 (Study report 1991f)

ENDPOINT_STUDY_RECORD: 7775-09-9,Short-term toxicity to aquatic invertebrates, Study report, 1991f, RS

Reliability

2 (reliable with restrictions)

Rationale for reliability incl. deficiencies:

No standard protocol was used, but the test method was described in enough detail. No chemical analyses on the test concentrations were perfored, only the stock solution was analyzed. From results in chronic tests it can be assumed that the test substance was stable during the test and animals were exposed properly.

Data source

Reference: Acute Flow-through Toxicity of Sodium Chlorate To The Mysid, Mysidopsis bahia / study report

Materials and methods:

Test guideline:

Test performed according to other: protocol developed for EnviroSystems Study Number 90117-DE.

Principles of method if other than guideline: It is not known if the protocol used is equal to a standard guideline. Twenty mysids were randomly and equally distributed among two replicates of each treatment. The test was performed in 19.6 liter glass aquaria that contained 15 liters of test solution (water depth was approximately 20 cm). Test vessels were randomly arranged in a water bath during the 96 h test (a random numbers table was used to select the location of each vessel). The test substance was supplied to the test vessels under flow through conditions by an intermittent flow proportional diluter. The diluter, which was constructed at EnviroSystems, allowed test media to contact only glass or Teflon coated surfaces. The diluter was calibrated before and after the test. During the test the diluter was activated 812 times, resulting in an average of 6.8 media exchanges per 24 hours in each test vessel. The number of surviving organisms and the occurrence of sublethal effects (loss of equilibrium, erratic swimming, loss of reflex, excitability, discoloration, or change in behavior) were determined visually and recorded after 24, 48, 72, and 96 hours.

GLP compliance: yes

Test material:

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: yes

Details on sampling

The concentration of sodium chlorate in test media could not be determined because of naturally occurring interference in the dilution water. The available analytical methods were effective in deionized water used to prepare toxicant stock solutions but not in the dilution water. A subsample of the 600,000 mg/L primary stock solution (prepared in deionized water) was withdrawn from the toxicant reservoir placed into 40 ml VOA vial, and stored in 2-4°C prior to analysis.

Details on analytical methods

HPLC method:

The analytical method was validated in deionized water at 1 to 25 mg/L. Several attempts were made to validate the method in dilution water without success. Interfering compounds, chlorate and nitrate present in the natural groundwater used as dilution water, interfere with the analysis by eluting near the sodium chlorate peak. Analytical samples (including standards and blanks) were filtered through a 0.5 micron filter into an HPLC autosampler vial and analyzed using a high performance liquid chromatograph (Waters Model 510 pump [2] with 680 controller, 431 conductivity detector, 712 Wisp, and HP 3350 data system or equivalent).

Test solutions:

Vehicle: no

Details on test solutions

- Dilution water: Water used for acclimation of test organisms and for all toxicity testing was unfiltered seawater collected from the Atlantic Ocean at EnviroSystems in Hampton, New Hampshire. Water, which had a salinity of 11 to 17 ppt (parts per thousand) and a pH of 7.7, was stored in 500-gallon polyethylene tanks where it was aerated.

- Stock solution: An initial 600,000 mg/L stock solution was prepared by combining 1,200.0 g of test substance and deionized water and adjusting the volume to approximately 1,800 ml in a 2 liter glass class A volumetric flask. The stock solution was mixed on a magnetic stir plate until the test substance dissolved and the total volume was adjusted to 2.0 L with deionized water. This procedure was repeated twice and all stock solution was transferred to a 6.0 L Erlenmeyer flask.

Test solutions: Appropriate amounts of the stock solution were added directly to dilution water by a proportional diluter (5.0 ml of test substance was combined with 3,000 ml of water during each diluter cycle) and this diluter toxicant cell solution was mixed by a high shear pump equipped with a Teflon head.

Test organisms

Test organisms (species): Americamysis bahia (previous name: Mysidopsis bahia)

Details on test organisms

Juvenile mysids (less than 24 hours old) were identified using an appropriate taxonomic key. They were produced from in-house cultures at EnviroSystems in Hampton, New Hampshire. Mysids were not treated for disease and they were free of apparent sickness, injuries,and abnormalities at the beginning of the test. Mysids were fed newly hatched Artemia salina nauplii once or twice daily before the test.

Study design

Test type: flow-through

Water media type:saltwater

Limit test: no

Total exposure duration: 96 h

Test conditions

Test temperature: 21.4 to 23.0 °C

pH: 7.6 to 7.8

Dissolved oxygen: 7.4 to 8.9 mg/L

Salinity: 16 to 17 parts per thousand

Nominal and measured concentrations: Mean nominal concentrations: 0 (control), 130, 220, 360, 590, and 1,000 mg/L sodium chlorate. The 600,000 mg/L (nominal) stock solution had a measured concentration of 650,000 mg/L sodium chlorate.

Details on test conditions

- Photoperiod: A 16 hour light and 8 hour dark photoperiod was automatically maintained

- Light intensity: cool-white fluorescent lights that provided a light intensity of 10 pEs-1/m2

- Aeration: Aeration was not required to maintain dissolved oxygen concentrations above acceptable levels.

- Feeding: Mysids were fed newly hatched Artemia salina nauplii once per day during the test.

Reference substance (positive control): no

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.	Nominal / measured	Conc. based on	Basis for effect
96 h	LC50	> 1000 mg/ L	nominal	test mat.	mortality
96 h	NOEC	>= 1000 mg/L	nominal	test mat.	mortality

Details on results:

One hundred percent survival occurred in the control and no sublethal effects were noted during the exposure period. Control mysids had an average wet weight (blotted dry) of 0.0001 g, resulting in a loading rate during the toxicity test of approximaJely 0.0001 g/L. One animal died in 590 mg/l and 3 died in 100 mg/l, but this was not significant.

Reported statistics and error estimates:

Results of the toxicity test could not be interpreted by standard statistical techniques (Stephan, 1983) because greater than 50% survival occurred at all tested concentrations.

Applicant's summary and conclusion

Validity criteria fulfilled: not specified

Conclusions

The test substance did not give significant effects at the highest test concentration of 1000 mg/l, therefore LC50 is greater than 1000 mg/l.

Sodium chlorate is not toxic to Mysid shrimp.

Executive summary

The acute toxicity of sodium chlorate to the mysid Mysidopsis bahia, is described in this final report. The test was conducted for Albright and Wilson Americas for 96 hours during February 15 to 19, 1991, at the EnviroSystems Division of Resource Analysts Inc. in Hampton, New Hampshire. It was conducted by Peter Kowalski, Ellen Stanford, Jeanne Magazu, Robert Boeri and Timothy Ward according to the protocol developed for EnviroSystems Study Number 90117-DE. The analytical portion of this study was conducted under the supervision of Gloria Switalski. Sodium chlorate (reported purity >99\ active ingredient) was supplied by the sponsor. The test was performed under flow-through conditions with five concentrations of test substance and a dilution water control at a temperature of 22 ± 1°C. The dilution water was unfiltered natural seawater collected at Hampton, New Hampshire. Nominal concentrations of sodium Chlorate were: 0 mg/L (control), 130, 220, 360, 590, and 1,000 mg/L. Nominal concentrations were used

for all calculations. Mysids used in the test were less than 24 hours old at the start of the test. They were produced from in-house cultures at EnviroSystems in Hampton, New Hampshire. After 96 hours of exposure the control organisms had an average wet weight (blotted dry) of 0.0001 g. All animals were in good condition at the beginning of the study. Exposure of mysids to the test substance resulted in a 96 hour LC50 greater than 1,000 mg/L sodium chlorate. The 96 hour no observed effect concentration was 1,000 mg/L sodium chlorate, the highest tested concentration.

5.3. Study 3 (Study report 1995)

ENDPOINT_STUDY_RECORD: 7775-09-9,Short-term toxicity to aquatic invertebrates, Study report,1995, SS

Reliability

2 (reliable with restrictions)

Rationale for reliability incl. deficiencies

other: see 'Remark' Study generated according to generally valid and internationally accepted testing guideline and performed under GLP, but some results were found which were not concentration related and could not be explaned. Therefore the study is considered to be valid with restrictions.

Data source

Reference: Acute toxicity of Sodium chlorate to the water flea, Daphnia magna, under static test conditions. / study report

Materials and methods

Test guideline

Test performed according to Guideline EPA OPP 72-2 (Aquatic Invertebrate Acute Toxicity Test)

GLP compliance: yes

Test material:

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: yes

Test organisms

Test organisms (species): Daphnia magna

Study design

Test type: static

Any other information on materials and methods incl. tables

Analytical methods: Ion chromatography with conductivity detection

TEST ORGANISM

- Source/supplier: In house culture, stock culture organisms were obtained from Aquatic Research Organisms, Hampton, NH

- Age: less than 24 hours

- Feeding: Selenastrum capricornutum and YTC (a mixture of yeast, trout chow and cereal leaves)

- Pretreatment: no

- Feeding during test: no

TEST CONDITIONS

- Stock solutions preparation: 2.554 g of sodium chlorate was dissolved in 2 l dilution water, to get a stock solution with a nominal concentration of 1000 mg chlorate/l (is equal to 1228 mg sodium chlorate/l)

- Dilution water

- Source: from a deep well located near the test site. The water was passed through a mixed bed deionized resin and then reconstituted with 192 mg/l sodium bicarbonate, 120 mg/l calcium sulfate, 120 mg/l magnesium sulfate and 8 mg/l potassium chloride.

- Aeration: No

- Alkalinity: 118 mg/l as CaCO3

- Hardness: 163 mg/l CaCO3

- pH: no information

- Oxygen content: no information

- Conductivity: 490 µmhos/cm (equal to µS/cm)

- Holding water: same as dilution water

TEST SYSTEM

- Concentrations:

nominal: 0, 62.5, 125, 250, 500 and 1000 mg chlorate/l

measured: 0, 51.8, 103.3, 208.1, 405.0 and 1018.5 mg chlorate/l

All measured concentrations were above 80% of the nominals.

- Exposure vessel type: 0.34 l cylindrical glass vessels containing 200 ml of test or control solution.

- Number of replicates, individuals per replicate: 4, 5

- Test temperature: 20.4 - 21.0 degree C

- Dissolved oxygen: 8.3-8.7 mg/l

- pH: 8.2-8.4

- Adjustment of pH: No

- Intensity of irradiation: 1098 +/- 146 lux

- Photoperiod: 16 hours of light and 8 hours of darkness with a 15 to 30 minutes transition period.

- Test parameter: Mortality

- Sampling: samples were taken immediately prior to the test and after 48 hours of exposure.

STATISTICS: Trimmed Spearman Karber

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.
48 h	EC50	1172 mg/L

Any other information on results incl. tables

- EC50: 919.3 mg chlorate/l (95% c.i. 612.28-1380.30 mg chlorate/l)

The concentrations above are expressed in mg chlorate ion, the concentrations of the substance are as follows: EC50 =1172 mg/l (95% c.i. 780.7-1759.9 mg/l). EC50 is a measured concentration. In the report a NOEC is given, but this is not a clear value. The reviewers consider that a NOEC could not be determined because effects were seen which were not concentration related. At 24 hours no organisms were found dead. At 48 hours 3 animals died in 103.3 mg/l, no dead were found in 208.1 mg/l and 2 in 405.0 mg/l and 11 in 1018.8 mg/l.

Applicant's summary and conclusion

Executive summary

Study Title: Acute Toxicity of Sodium Chlorate to the Water Flea, Daphnia magna Under Static Test Conditions

Data Requirement: Section 72-2 of the EPA Pesticide Assessment Guidelines, Subdivision E

Sponsor: Sodium Chlorate Reregistration Task Force c/o Delta Analytical Corporation, 79 10 Woodmont Avenue, Suite 1O00, Bethesda, Maryland 20814 Tel. No. (301) 652-5495

Location of Study: Environmental Science & Engineering, Inc.WE), P.O. Box 1703 Gainesville, FL 32602-1703

Study Director: Joe Owusu-Yaw, Ph.D. (ESE)

Study Initiation Date: June5, 1995

Experimental Start Date: July 6, 1995

Experimental Termination Date: July 8, 1995

Test Substance: Sodium Chlorate, CAS Number 7775-09-9, Lot No. DL 2, purity 99.95 percent

Test Organism and Source: Daphnia mgna, 24 hours old, obtained from laboratory culture maintained at ESE

Test Conditions: Dilution water-reconstituted hard water with a hardness of 163 mgL and alkalinity of 118 mgk, both as CaCO,, photoperiod--16 -h light and 8 -h dark with 15 to 30 -minute dawddusk transition, temperature-20.4 to 21.0 "C

Test Results: 48-hour EC,: 919.3 mg chloratek; NOEC: 405 mg chlorate/L

Location of Raw Data and Final Report: Environmental Science & Engineering, Inc. (ESE) P.O. Box

1703 Gainesville, FL 32604 -1703

6. LONG-TERM TOXICITY TO AQUATIC INVERTEBRATES

6.1. Study 1. (Study report 2004b)

ENDPOINT_STUDY_RECORD: 7775-09-9, Long-term toxicity to aquatic invertebrates, Study report,2004b,RS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies:

other: Study generated according to a valid and internationally accepted testing guideline and was performed under GLP.

Data source

Reference: Chronic toxicity of sodium chlorate to Daphnia magna in a 21 day reproduction test under semi-static... / study report

Materials and methods

Test guideline

Test performed according to OECD Guideline 211 (Daphnia magna Reproduction Test)

GLP compliance: yes

Test material:

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: yes

Details on sampling:

- Samples were taken on the first day of the study on preparation and just before changing.

- Further samples were taken weekly.

- Samples were taken in duplicate and were frozen until analysis.

Details on analytical methods

A Dionex DX-120 ion chromatograph equipped with an ASS-HC 4 mm analytical column, an AG9-HC 4 mm guard column, a 50 pL loop, an ASRS-ultra 4 mrn and a CDM-3 flow through conductivity cell with a DS4 detection stabiliser was used to detect and quantify chlorate.

The DX-120 was operated at a column temperature of 20 ºC, a detector temperature of 35 ºC and an eluent flow rate of 1.0 ml/min.

The eluent composition was 9.0 mM Na2C03. Data was acquired and integrated using a ThermoLabsystems Chromatography Server and Atlas 2002 version 6.18. Samples were loaded using a Dionex AS40 automated sampler with 0.5 ml vials.

Test solutions

Vehicle: no

Details on test solutions:

- Procedures: To prepare the stock solutions, approximately 5 or 10 g of test substance was weighed and dissolved directly in 500 or 1000 ml, respectively of M4 medium.

- The obtained preparations were agitated mechanically for 24 h in an attempt to completely dissolve the test substance.

- The pH of each stock solution was between 6.8 and 8.3.

- The pH was adjusted to between 7.6 and 7.8 with 1 M HCI or NaOH (reagent grade) when necessary.

- A fresh stock solution was prepared for each solution change.

Test organisms

Test organisms (species): Daphnia magna

Details on test organisms:

Daphnia magna STRAUS-clone 4

- Source/supplier: AquaSense, Amsterdam, The Netherlands

- Breeding method: According to the relevant SOP

- Age: less than 24 hours

- Feeding: Chlorella vulgaris

- Feeding during test: yes, between 0.1 and 0.2 mg of carbon per daphnid per day, from day 8

Study design

Test type: semi-static

Water media type: freshwater

Limit test: no

Total exposure duration: 21 d

Test conditions

Hardness: 133-138 °dH

Test temperature:20.2-21.4 °C

pH:7.6-8.2

Dissolved oxygen:8.0-9.1 mg/l

Nominal and measured concentrations

- Nominal: 0, 12.8, 32, 80, 200 and 500 mg/L

- Measured: The measured concentrations were within 20% of the nominal concentrations and therefore the nominals were used for all statistical evaluation.

Details on test conditions

DILUTION WATER:

- Elendt M4 medium

- Aeration: yes

- Ca/Mg ratio: 4:1

- Na/K ratio: 10:1

- pH: 8.0 ± 0.5

- Holding water: same as dilution water

- Renewal of test solution: every two to three days

- Exposure vessel type: 50 ml glass beakers

- Number of replicates, individuals per replicate: 10, 1

- Conductance: in control 609-663 µs/cm, in 500 mg/L 1099-1145 µs/cm

- Adjustment of pH: no

- Intensity of irradiation: ambient light provided by fluorescent tubes

- Photoperiod: 16 hours photoperiod daily

Reference substance (positive control): no

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc	Nominal / measured	Conc. based on	Basis for effect
21 d	NOEC	>= 500 mg/L	nominal	test mat.	other: reproduction, weight and length

Details on results

- Unhatched eggs: None found

- Length and weight of surviving animals: No significant difference between the concentrations.

- Numbers of dead young: At all concentrations except the control at least one and at most five neonates were found to be dead.

The mortality appeared to be concentration related but was insignificant compared to the living neonat

es. Immobilization: one daphnid died in 80 mg/L on day 15.

- EC50 for mortality of adults and for reproduction could not be determined due to insufficient mortality

in the test concentrations.

CONTROL:

- Number/percentage of animals showing adverse effects: No mortality occurred

- Average number of juveniles per parent: 103.3 after 21 days

Reported statistics and error estimates

Weight data were tested for normality using Chi-square test and Barlett's test for homogeneity of variance.

ANOVA was performed on the number of living neonates per parent using Bonferroni t-test and verified with the Dunnett's test.

All other parameters were not tested because the result in the control group was lower than in the test concentrations.

Any other information on results incl. tables

Time to first brood: Number of animals having the first brood on a given day.

Conc (mg/l)	Day of first brood					
	8	9	10	11	12	13
0	7	3				
12.8	9		1			
32	6	3	1			
80	6	3		1		
200	9	1				
500	3	2	4			1

Number of juveniles produced: No significant difference was found in total and mean number of juveniles between the concentrations.

Average total number of living juveniles produced per parent per concentration:

Conc (mg/l)	no. juveniles/parent
0	103.3
12.8	109.8
32	120.7
80	121.7
200	126.5
500	108.6

Applicant's summary and conclusion

Validity criteria fulfilled: yes

Conclusions

Test conducted under GLP with analysis, analysis certificate was present and to relevant guideline was used. This study can be considered reliable without restrictions. The NOEC for reproduction, weight and length is equal to or greater than 500 mg/l.

Executive summary

The purpose of this study was to assess the toxicity of the test substance dissolved in fresh water, on the reproductive efficacy of Daphnia magna STRAUS - clone 4, in a 21-day semi-static test complying with the OECD Guideline No. 211 , 21st September 1998.

The test criterion of toxicity used was reproductive capacity expressed as the number of neonates per daphnid per day.

The nominal concentrations used in the study were as follows: 0, 12.8, 32.0, 80.0, 200 and 500 mg/l

Analytical determinations of the test solutions were made on six occasions during the test. The concentrations were found to remain stable to within ±20% of the nominals. The nominal concentrations were used to calculate the effect concentrations.

The validity criteria were respected: No mortality occurred in the control group over the test period.

The average number of juveniles per parent in the control was 103.3 after 21 days The No Observed Effect Concentration (NOEC) is determined as the concentration used in the study that is immediately below the Lowest Observed Effect Concentration (LOEC), the latter derived statistically from the data, where possible, using the appropriate statistical test.

Reproductive output and length of adults at the end of the study were lower in the control than in any other concentration tested and was therefore not evaluated statistically. Weight data were found to be normally distributed and homogeneous. Using Dunnett’s and Bonferroni-t tests, the lowest Observed Effect Concentration (LOEC) based on weight was found to be greater than 500 mg/l. Based on these statistical results the NOEC for reproduction, weight and length is 500 mg/l.

The EC50 for adult mortality and for reproduction could not be determined due to insufficient mortality in any of the test concentrations.

7. TOXICITY TO AQUATIC ALGAE AND CYANOBACTERIA

7.1. Study 1 (Study report 1991 e)

ENDPOINT_STUDY_RECORD: 7775-09-9,Toxicity to aquatic algae and cyanobacteria, Study report,1991e,RS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies

other: Study generated according to generally valid and internationally accepted testing guideline and performed under GLP. Only the concentration the stock solution was analysed, but the test substance is considered to be stable.

Data source

Reference: Static acute toxicity of sodium chlorate to the freshwater algae, Selenastrumcapricornutum. / study report

Materials and methods

Test guideline

Qualifier: according to Guideline EPA OPP 122-2 (Algal Toxicity, Tiers I and II)

GLP compliance: yes

Test material

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: no

Details on sampling

Only a stock solution of 1000 mg/l was measured because of naturally occurring interference of the dilution water.

Details on analytical methods:

HPLC, analytical determination was carried out on an anion exchange column using an external standard. Analysis was performed in isocratic mode with conductivity detection.

Test solutions

Vehicle: no

Test organisms

Test organisms (species): Pseudokirchneriella subcapitata (previous names: Raphidocelis subcapitata, Selenastrum capricornutum)

Details on test organisms:

- Source/supplier: Culture Collection of Algae at the University of Texas at Austin

- Laboratory culture: Yes - Method of cultivation: In sterile enriched media (U.S. EPA, 1978)

- Initial cell concentration: 10000 cells/ml.

- No information given about the growth stage the algae culture was in.

Study design

Test type: static

Water media type: freshwater

Limit test: no

Total exposure duration: 96 h

Post exposure observation period

Pretreatment: maintained for at least 14 days

Test conditions

Test temperature: 23.5-25.1 °C

pH: 7.2-7.6

Nominal and measured concentrations:

- Nominal concentrations: 0, 62.5, 125, 250, 500, 1000 mg/L

- Measured concentration: The stock solution had a measured concentration of 1100 mg/l.

This is within 80% of the nominal concentration of 1000 mg/l.

Details on test conditions

DILUTION WATER:

- Sterile enriched media (U.S. EPA, 1978)

- pH: 7.5

- Intensity of irradiation: 40 µEs/m2

- Photoperiod: continuous light

- Endpoint: number of cells

- Exposure vessel type: 250 ml glass erlenmeyer flasks

- Number of replicates: 3

GROWTH/TEST MEDIUM:

- Sterile enriched media (U.S. EPA, 1978)

Reference substance (positive control): no

Results and discussion

Details on results

- EbC50 (72h) = 129 mg/L (95% c.i. 117-141- mg/L)

- EbC50 (96h) = 133 mg/L (95% c.i. 122-144- mg/L)

- NOEC = 62.5 mg/l

Reported statistics and error estimates

Standard method according to Stephan, 1983

Applicant's summary and conclusion

Validity criteria fulfilled: no

No chemical analyses was performed on the test concentrations, only the stock solution was analyzed.

7.2. Study 2 (study report 2004c)

ENDPOINT_STUDY_RECORD: 7775-09-9,Toxicity to aquatic algae and cyanobacteria, Study report,2004c,SS

Reliability

2 (reliable with restrictions)

Rationale for reliability incl. deficiencies

other: see 'Remark' Study generated according to generally valid and internationally accepted testing guideline and performed under GLP, but the results obtained were not consistent at the highest concentration tested and Fe2O3 had an influence on the spectroscopic measurements.

Data source

Reference: Effects of Sodium chlorate on the growth of the freshwater green alga, Scenedesmus subspicatus. / study report

Materials and methods

Test guideline:

Qualifier according to OECD Guideline 201 (Alga, Growth Inhibition Test) before 23 March 2006

Deviations: yes; the NaHCO3 concentration of the test medium was 150 mg/L instead of 50 mg/L, as recommended by the OECD/EEC guidelines, in order to maintain a more constant pH during the test. The pH should not deviate more than 1.5 units during the test (EEC). The growth rate at the highest test concentration is calculated using only the extinction at the beginning and the end of the test. This deviation is considered to have no impact on the integrity and quality of the study.

GLP compliance: yes

Test material

Test material information, sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: yes

Details on analytical methods

The test item concentrations were analysed by ion chromatography for the control, the lowest, the middle and the highest test concentration.

Test solutions

Vehicle: no

Test organisms

Test organisms (species): Desmodesmus subspicatus (previous name: Scenedesmus subspicatus)

Details on test organisms

- source: Culture Collection of Algae and Protozoa, Institute of Freshwater Ecology, Ambleside, United Kingdom.

Study design

Test type: static

Water media type: freshwater

Limit test: no

Total exposure duration: 72 h

Test conditions

Test temperature: 23.0-23.7°C

pH: 8.0-8.4

Nominal and measured concentrations

Test concentrations: 0, 97.4, 202.2, 396.9, 793.8 and 1592.3 mg a.i./L (nominal).

Measured concentrations were higher than 80% of the nominals, therefore nominal concentrations were used for the effect calculations.

Details on test conditions

- Replicates: 5 test concentrations with 3 replicates, plus a control with 6 replicates

- Glassware: 100 mL glass Erlenmeyer flasks, continuous shaking

- initial cell densities of 10.000 cells/mL

- continuous light at about 91-93 µmol.s-1m-2.

- Cell concentrations were determined photometrically with a UV/VIS spectrophotometer.

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.	Nominal / measured	Conc. based on	Basis for effect
72 h	NOEC	1592.3 mg/L	nominal	test mat.	growth rate

Details on results

- The cell density of the controls increased at least a factor 16 within 72 hours, the quality criteria have been met.

- The measured concentrations are higher than 80% of the nominal. The results are based on the nominal concentrations.

- The extinction at the beginning of the test is higher in the test solutions than in the control. The Fe2O3 present in the test substance is most likely the cause of this increase. At the highest test concentration, a small decrease of the extinction was observed after 24 h. This is probably caused by the precipitation of Fe2O3. This phenomenon is considered to have a negligible impact on the final result. However, the growth rate at this concentration was calculated using only the extinctions at the beginning and at the end of the test.

There is no significant effect on the growth rate of Scenedesmus subspicatus.

For the biomass a 9% response was found at a test concentration of 794 mg/L and a 22% response was found at the highest test concentration of 1592.3 mg/L. An EbC50 of 3665 mg/L is calculated after extrapolation. NOEC (biomass) = 396.9 mg/l

Reported statistics and error estimates

Results of the toxicity test were interpreted by probit analysis. All computations were performed using the TOXCALCTM 5.0 program.

Any other information on results incl. tables

Remark => The reviewer finds it more accurate to say that the EbC50 is higher than 1592.3 mg/l, the highest concentration tested.

An ErC50 is not given in the report, but it can also be stated that this is higher than 1592.3 mg/l.

The NOEC (growth rate) = 1592.3 mg/l

The increase in extinction most likely caused by Fe2O3, is not considered to have a significant impact on the results of this test. In the calculation of the biomass a correction is made for the higher extinctions measured at t=0.

The higher values at t=0 do not have an influence on the slope of the growth curve, which is used for the calculation of the growth rate. Therefore, these results are considered to be accurate.

Applicant's summary and conclusion

Validity criteria fulfilled: yes

Conclusions

Sodium chlorate is not very toxic to Scenedesmus subspicatus. The highest test concentration of 1595 mg/l gave only a 22% biomass inhibition response. It was therefore not possible to calculate reliable EC50 values for both the inhibition based on biomass and growth rate.

The NOEC determined from the results is 396.9 mg/l, the LOEC is 793.8 mg/l.

Executive summary

In order to predict effects of Sodium chlorate in an aquatic environment, the toxicity of this chemical to freshwater algae was assessed. The algal toxicity was determined in the Algal Growth Inhibition test in accordance with OECD, EEC and IS0 test guidelines and with the OECD Principles of Good Laboratory Practice. The guidelines were slightly modified to ensure good growth and pH control of the cultures. The green algae Scenedesmus subspicatus was exposed to the following final nominal test concentrations: 97.4 - 202.2 - 396.9 - 793.8 and 1592.3 mg/l of sodium chlorate.

The toxicity of sodium chlorate to an exponentially growing culture of Scenedesmus subspicatus was determined over an exposure period of 72 hours. Sodium chlorate is not very toxic to Scenedesmus subspicatus. The highest test concentration of 1595 mg/l gave only a 22% biomass inhibition response. It was therefore not possible to calculate reliable EC50 values for both the inhibition based on biomass and growth rate. The NOEC determined from the results is 396.9 mg/l, the LOEC is 793.8 mg/l. The test was conducted in a mineral salts medium in a climatized illuminated orbital incubator. The maximum variation in pH in the test media was 0.4 pH unit. The definitive test is valid as shown by the increase of the extinction of the control over 72 h by a factor of 26.

The chemical analyses carried out showed that the concentration of the test compound during the test remained within 80% of the concentration at the beginning of the test. The results are based on nominal concentrations.

7.3. Study 3 (Study report 1998a)

ENDPOINT_STUDY_RECORD: 7775-09-9,Toxicity to aquatic algae and cyanobacteria, Stauber,1998,RS(A)

Reliability

2 (reliable with restrictions)

Rationale for reliability incl. deficiencies:

other: No GLP and no standard test protocol, but test described in enough detail.

Data source

Reference: Toxicity of chlorate to marine microalgae. / Stauber, J.L. / publication

Materials and methods

Test guideline: Guideline; other: Standard 72h growth inhibition bioassay (Stauber etal., 1994)

Principles of method if other than guideline:

Toxicity of chlorate at 3 nitrate concentrations (<0.005, 1 and 15 mg/l NO3) was studies using a standard 72 hours growth inhibtion bioassay (Stauber et al., 1994). At a nitrate concentration of <0.005 mg/l the toxicity of chlorate at 5 concentrations (0.5, 1, 5, 10 and 20 mg/l) was determined. At higher nitrate concentration (1 and 15 mg/l), 5 concentrations of chlorate were tested ranging from 1-1000 mg/l depending on the alga and nitrate concentration in each experiment. Cell densities were determined daily using a Coulter Multisizer IIE with 70 µm aperture.

GLP compliance: no

Test material

Test material information: potassium chlorate / 3811-04-9 / 223-289-7

Sampling and analysis

Analytical monitoring: yes

Details on sampling

See details in analytical methods.

Details on analytical methods

Chlorate concentrations at the beginning and the end of the test (in the presence and absence of algal cells) were analysed according to a modified method (Ceba et al., 1978). In this method, chlorate is reduced to chlorine in hte presence of chloride and perchloric acid, and the chlorine then reacts with reduced 1,3-cyclohexanedione bisthiosemicarbazone hydrochloride to form a stable yellow oxidation product. An aliquot from each bioassay flask (0.3 ml) was added to 3 ml of 1 M NaCl in a 25 ml volumetric flask.NaBH4 (1 ml of 0.5%) was added to one of the two replicates and the flasks were allowed to stand for 5 minutes for complete reduction of chlorite. To each flask, 1 ml of a 1,3-cyclohexanedione bisthiosemicarbazone. HCl stock (0.25 g dissolved in 100 ml 0f 0.1 M HCl) and 11 ml of concentrated HClO4 was added. After standing for 10 minutes, flasks were made up to 25 ml volume and the absorbance at 402 nm was measured versus a blank. Chlorate standards (0-100 µg ClO3- 25 ml-1) were included in each assay. The molar absorptivity of the semicarbazone was 1.71 x 10^4 l/mol/cm and the LOD was 100 µg/l ClO3-. Chlorate analytical recovery was always >90%. The presence of algal cells/cell debris had no effect on the determination of chlorate using this method.

Test solutions

Vehicle: no

Details on test solutions:

Three nitrate levels were used: <0.005, 1 and 15 mg/l NO3. At nitrate concentrations of <0.005 mg/l the toxicity of chlorate was tested at 0.5, 1, 5, 10 and 20 mg/l ClO3-. At nitrate concentrations of 1 and 15 mg/l the test concentrations were ranging from 1 to 1000 mg/l ClO3-, depending on the alga and nitrate concentration in each experiment.

Test organisms

Test organisms (species)

other: other algae: Nitzschia closterium

Details on test organisms

Nitzschia closterium (Ehrenberg) W. Smith, originally isolated from Port Hacking, NSW, Strain CS-5c.

Culture medium: Medium f (Guillard and Ryther, 1962) with the iron and trace element concentrations halved.

Study design

Test type: static

Water media type: saltwater

Limit test: no

Total exposure duration: 72 h

Test conditions

Nominal and measured concentrations:

At nitrate concentrations of <0.005 mg/l the toxicity of chlorate was tested at 0.5, 1, 5, 10 and 20 mg/l ClO3-. At nitrate concentrations of 1 and 15 mg/l the test concentrations were ranging from 1 to 1000 mg/l ClO3-, depending on the alga and nitrate

concentration in each experiment. Chlorate was measured in each bioassay flask at the beginning and end of the test and was always within 10% of the nominal chlorate concentration.

Details on test conditions

Seawater for the preparation of the test medium was collected 0-1 km offshore from Port Hacking, NSW. The seawater was filtered through a 0.45 µm membrane filter and stored at 4 °C.

Reference substance (positive control): no

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.	Nominal / measured	Conc. based on	Basis for effect	Remarks on result
72 h	EC50	1.9 mg/L	nominal	other: chlorate ion	growth rate	other: 95% CL: 1.6 - 2.3 mg/l Nitrate conc: < 0.005 mg/l
72 h	EC50	10 mg/L	nominal	other: chlorate ion	growth rate	other: Nitrate conc: 1 mg/l
72 h	EC50	> 500 mg/L	nominal	other: chlorate ion	growth rate	other: Nitrate conc: 15 mg/l
72 h	NOEC	100 mg/L	nominal	other: chlorate ion	growth rate	other: Nitrate conc: 15 mg/l

Details on results

- Analytical results: Chlorate was measured in each bioassay flask at the beginning and end of the test and was always within 10% of the nominal chlorate concentration. Chlorate concentrations remained stable over the 3 days and chlorate was not reduced to chlorite or chloride in the light either in the presence or absence of algal cells.

Reported statistics and error estimates

Data analyses were performed using Toxcalc version 5.0 (Tidepool scientific software).

Any other information on results incl. tables

Recalculation of the results from mg/l ClO3- to KClO3. EC50:

- <0.005 mg nitrate/l: 1.9 mg ClO3-/l (95% c.i. 1.6-2.3 mg/l) corresponds to 2.8 mg KClO3/l (95% c.i. 2.3-3.4 mg/l)

- 1 mg nitrate/l: 10 mg ClO3-/l corresponds to 15 mg KClO3/l

- 15 mg nitrate/l: >500 mg ClO3-/l corresponds to >735 mg KClO3/l

NOEC:- 15 mg nitrate/l: 100 mg ClO3-/l corresponds to 147 mg KClO3/l

Control cell division rate (doublings/day)

- <0.005 mg nitrate/l: 0.8

- 1 mg nitrate/l: 1.1

- 15 mg nitrate/l: 1.3

Remark from reviewer: According to OECD guideline 201 Freshwater Alga and Cyanobacteria, Growth Inhibition Test the he biomass in the control cultures should have increased exponentially by a factor of at least 16 within the 72-hour test period. This corresponds to a specific growth rate of 0.92 day-1. For the test performed at <0.005 mg nitrate/l this was not the case and therefore this test result is not valid and can not be used.

Applicant's summary and conclusion

Validity criteria fulfilled: not specified

Conclusions

EC50:

- 1 mg nitrate/l: 10 mg ClO3-/l corresponds to 15 mg KClO3/l

- 15 mg nitrate/l: >500 mg ClO3-/l corresponds to >734 mg KClO3/l

NOEC:- 15 mg nitrate/l: 100 mg ClO3-/l corresponds to 147 mg KClO3/l

The results obtained at a nitrate concentration of <0.005 mg/l were not considered to be valid due to a low doubling rate in the control vessels and can not be used, therefore.

Executive summary

Toxicity of chlorate at 3 nitrate concentrations (<0.005, 1 and 15 mg/l NO3) was studies using a standard 72 hours growth inhibtion bioassay (Stauber et al., 1994). At a nitrate concentration of <0.005 mg/l the toxicity of chlorate at 5 concentrations (0.5, 1, 5, 10 and 20 mg/l) was determined. At higher nitrate concentration (1 and 15 mg/l), 5 concentrations of chlorate were tested ranging from 1-1000 mg/l depending on the alga and nitrate concentration in each experiment.

Cell densities were determined daily using a Coulter Multisizer IIE with 70 µm aperture.

Nitzschia closterium was used as test organism.

EC50:

- <0.005 mg nitrate/l: 1.9 mg ClO3-/l (95% c.i. 1.6-2.3 mg/l) corresponds to 2.8 mg KClO3/l (95% c.i. 2.3-3.4 mg/l)

- 1 mg nitrate/l: 10 mg ClO3-/l corresponds to 15 mg KClO3/l

- 15 mg nitrate/l: >500 mg ClO3-/l corresponds to >734 mg KClO3/l

NOEC:- 15 mg nitrate/l: 100 mg ClO3-/l corresponds to 147 mg KClO3/l

The results obtained at a nitrate concentration of <0.005 mg/l were not considered to be valid due to a low doubling rate in the control vessels and cannot be used, therefore.

7.4. Study 4 (Study report 1998 b)

ENDPOINT_STUDY_RECORD: 7775-09-9,Toxicity to aquatic algae and cyanobacteria, Stauber,1998,SS(B)

Reliability

2 (reliable with restrictions)

Rationale for reliability incl. deficiencies

other: No GLP and no standard test protocol, but test described in enough detail.

Data source

Reference: Toxicity of chlorate to marine microalgae. / Stauber, J.L. / publication

Materials and methods

Test guideline:

Guideline, other: Standard 72h growth inhibition bioassay (Stauber etal., 1994)

Principles of method if other than guideline:

Toxicity of chlorate at 3 nitrate concentrations (<0.005, 1 and 15 mg/l NO3) was studies using a standard 72 hours growth inhibtion bioassay (Stauber et al., 1994). At a nitrate concentration of <0.005 mg/l the toxicity of chlorate at 5 concentrations (0.5, 1, 5, 10 and 20 mg/l) was determined. At higher nitrate concentration (1 and 15 mg/l), 5 concentrations of chlorate were tested ranging from 1-1000 mg/l depending on the alga and nitrate concentration in each experiment. Cell densities were determined daily using a Coulter Multisizer IIE with 70 µm aperture.

GLP compliance: no

Test material

Test material information: potassium chlorate / 3811-04-9 / 223-289-7

Sampling and analysis

Analytical monitoring: yes

Details on sampling:

See details in analytical methods.

Details on analytical methods

Chlorate concentrations at the beginning and the end of the test (in the presence and absence of algal cells) were analysed according to a modified method (Ceba et al., 1978). In this method, chlorate is reduced to chlorine in hte presence of chloride and perchloric acid, and the chlorine then reacts with reduced 1,3-cyclohexanedione bisthiosemicarbazone hydrochloride to form a stable yellow oxidation product. An aliquot from each bioassay flask (0.3 ml) was added to 3 ml of 1 M NaCl in a 25 ml volumetric flask.NaBH4 (1 ml of 0.5%) was added to one of the two replicates and the flasks were allowed to stand for 5 minutes for complete reduction of chlorite. To each flask, 1 ml of a 1,3-cyclohexanedione bisthiosemicarbazone. HCl stock (0.25 g dissolved in 100 ml 0f 0.1 M HCl) and 11 ml of concentrated HClO4 was added. After standing for 10 minutes, flasks were made up to 25 ml volume and the absorbance at 402 nm was measured versus a blank. Chlorate standards (0-100 µg ClO3- 25 ml-1) were included in each assay. The molar absorptivity of the semicarbazone was 1.71 x 10^4 l/mol/cm and the LOD was 100 µg/l ClO3-. Chlorate analytical recovery was always >90%. The presence of algal cells/cell debris had no effect on the determination of chlorate using this method.

Test solutions

Vehicle: no

Details on test solutions:

Three nitrate levels were used: <0.005, 1 and 15 mg/l NO3. At nitrate concentrations of <0.005 mg/l the toxicity of chlorate was tested at 0.5, 1, 5, 10 and 20 mg/l ClO3-. At nitrate concentrations of 1 and 15 mg/l the test concentrations were ranging from 1 to 1000 mg/l ClO3-, depending on the alga and nitrate concentration in each experiment.

Test organisms

Test organisms (species): Dunaliella tertiolecta

Details on test organisms:

Dunaliella tertiolecta Butcher was obtained from the CSIRO Division of Fisheries Microlagal Culture Collection (Strain CS-175)

Culture medium: Medium f (Guillard and Ryther, 1962) with the iron and trace element concentrations halved.

Study design

Test type: static

Water media type: saltwater

Limit test: no

Total exposure duration: 72 h

Test conditions

Nominal and measured concentrations:

At nitrate concentrations of <0.005 mg/l the toxicity of chlorate was tested at 0.5, 1, 5, 10 and 20 mg/l ClO3-.

At nitrate concentrations of 1 and 15 mg/l the test concentrations were ranging from 1 to 1000 mg/l ClO3-, depending on the alga and nitrate concentration in each experiment. Chlorate was measured in each bioassay flask at the beginning and end of the test and was always within 10% of the nominal chlorate concentration.

Details on test conditions

Seawater for the preparation of the test medium was collected 0-1 km offshore from Port Hacking, NSW. The seawater was filtered through a 0.45 µm membrane filter and stored at 4 °C.

Reference substance (positive control): no

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.	Nominal / measured	Conc. based on	Basis for effect	Remarks on result
72 h	EC50	11 mg/L	nominal	other: chlorate ion	growth rate	other: 95% CL: 9 - 12 mg/l Nitrate conc: <

						0.005 mg/l
72 h	EC50	13 mg/L	nominal	other: chlorate ion	growth rate	other: 95% CL: 10 - 16 mg/l Nitrate conc: 1 mg/l
72 h	EC50	> 1000 mg/L	nominal	other: chlorate ion	growth rate	other: Nitrate conc: 15 mg/l
72 h	NOEC	500 mg/L	nominal	other: chlorate ion	growth rate	other: Nitrate conc: 15 mg/l

Details on results

- Analytical results: Chlorate was measured in each bioassay flask at the beginning and end of the test and was always within 10% of the nominal chlorate concentration. Chlorate concentrations remained stable over the 3 days and chlorate was not reduced to chlorite or chloride in the light either in the presence or absence of algal cells.

Reported statistics and error estimates

Data analyses were performed using Toxcalc version 5.0 (Tidepool scientific software).

Any other information on results incl. tables

Recalculation of the results from mg/l ClO3- to KClO3.

EC50:

- <0.005 mg nitrate/l: 11 mg ClO3-/l (95% c.i. 9 -12 mg/l) corresponds to 16 mg KClO3/l (95% c.i. 13 -18 mg/l)

- 1 mg nitrate/l: 13 mg ClO3-/l corresponds to 19 mg KClO3/l (95% c.i. 15-23 mg/l)

- 15 mg nitrate/l: >1000 mg ClO3-/l corresponds to >1469 mg KClO3/l

NOEC:- 15 mg nitrate/l: 500 mg ClO3-/l corresponds to 735 mg KClO3/l

Control cell division rate (doublings/day)

- <0.005 mg nitrate/l: 0.5

- 1 mg nitrate/l: 0.7

- 15 mg nitrate/l: 1.1

Remark from reviewer: According to OECD guideline 201 Freshwater Alga and Cyanobacteria, Growth Inhibition Test the biomass in the control cultures should have increased exponentially by a factor of at least 16 within the 72-hour test period. This corresponds to a specific growth rate of 0.92 day-1. For the test performed at <0.005 and 1 mg nitrate/l this was not the case and therefore these test results are not considered to be valid and cannot be used.

Applicant's summary and conclusion

Validity criteria fulfilled: not specified

Conclusions

EC50 at 15 mg nitrate/l: >1000 mg ClO3-/l corresponds to >1469 mg KClO3/l

NOEC at 15 mg nitrate/l: 500 mg ClO3-/l corresponds to 735 mg KClO3/l

The results obtained at a nitrate concentration of <0.005 and 1 mg/l were not considered to be valid due to a low doubling rate in the control vessels and cannot be used, therefore.

Executive summary

Toxicity of chlorate at 3 nitrate concentrations (<0.005, 1 and 15 mg/l NO3) was studies using a standard 72 hours growth inhibtion bioassay (Stauber et al., 1994). At a nitrate concentration of <0.005 mg/l the toxicity of chlorate at 5 concentrations (0.5, 1, 5, 10 and 20 mg/l) was determined. At higher nitrate concentration (1 and 15 mg/l), 5 concentrations of chlorate were tested ranging from 1-1000 mg/l depending on the alga and nitrate concentration in each experiment.

Cell densities were determined daily using a Coulter Multisizer IIE with 70 µm aperture. Dunaliella tertiolecta was used as test organism.

EC50:

- <0.005 mg nitrate/l: 11 mg ClO3-/l (95% c.i. 9 -12 mg/l) corresponds to 16 mg KClO3/l (95% c.i. 13 -18 mg/l)

- 1 mg nitrate/l: 13 mg ClO3-/l corresponds to19 mg KClO3/l (95% c.i. 15-23 mg/l)

-15 mg nitrate/l: >1000 mg ClO3-/l corresponds to >1469 mg KClO3/l

NOEC:

- 15 mg nitrate/l: 500 mg ClO3-/l corresponds to 735 mg KClO3/l

The results obtained at a nitrate concentration of <0.005 and 1 mg/l were not considered to be valid due to a low doubling rate in the control vessels and cannot be used, therefore.

7.5. Study 5 (study report 2010 a)

ENDPOINT_STUDY_RECORD: 7775-09-9, Toxicity to aquatic algae and cyanobacteria, Study report, 2010, SS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies:

other: Well conducted study according to standard guideline and GLP.

Data source

Reference: Sodium Chlorate Growth inhibition of the marine alga Skeletonema costatum / study report

Materials and methods

Test guideline:

Qualifier according to ISO 10253 (Water quality - Marine Algal Growth Inhibition Test with Skeletonema costatum and Phaeodactylum tricornutum)

Deviations: no

GLP compliance: yes

Test material

Test material information: sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: yes

Details on sampling:

Samples were taken at the start of the test before addition of the algae and at the end of the test. Samples were filtered through 0.2 µm disposable cellulose acetat filters and stored at 4°C in 20 ml scintillations vials. Storage time was less than 15 days for all samples.

Details on analytical methods

Preparation of a 1000 mg/l stock of the test substance by weighing 1 g of test compound and dissolving it in 1 liter of aged seawater. The stock solution was stored at 4°C and was less than 1 week old when used for establishing a calibration curve. (The stock solution was in fact only one day old: personal communication with SD). From this stock a series of standard solutions were made to cover the test concentrations of 1 to 1000 mg/l of Sodium Chlorate. Because of the high amount of chlorine in seawater both standards and test solutions were diluted 100x before analysis. A simultaneous calibration with SO4 gave that the response factor for Chlorate relative to SO4 was 0.5365. The linearity of the calibration curve had correlation coefficient of 99.948 % with an intercept of -0.001 and slope value of 1.349 on the day samples were analysed.

DX 320 Dionex Ionchrometograph including : 2 modules of IC25, an Dionex Ionpac CS16 separation column for cations, an Dionex Ionpac AS15 separation column for anions, an Dionex Ionpac CG16 precolumn for cations, Dionex Ionpac AG15 precolumn for anions, Dionex Ionpac ATC-HC trap column for anions and 2 x EG40 Eluent generators.

Ion Cromatograph

- Column: Dionex IonPack AS15 3mm (id) x 150 mm

- Eluent Cartrigde: KOH Eluent cartridge from Dionex art no 058900

- Gard column: Dionex IonPac AG15 3mm(id) x 150 mm

- Gas: 99.999 % pure N2

- Injection sample volume: 200 µl

- Eluent flow: 0.7 ml/min

- Electrical Current: 100 mA

- Detector temperature: 27 °C

- Eluent: KOH

Test solutions

Vehicle: no

Details on test solutions:

A solution of the test material in seawater (1000 mg/l) was prepared by dissolving overnight with a magnet stirrer. The solution was further diluted in test medium to obtain the selected exposure concentrations.

Test organisms

Test organisms (species): Skeletonema costatum

Details on test organisms:

Strain: NIVA strain BAC 1

Source: NIVA culture collection

Stock culture: Cultured in natural seawater with 10 % Z8 medium on reciprocating shaker and continuous light at approximately 20 °C.

Inoculation culture: The inoculum was taken from a culture in ISO 10253 growth medium prepared from natural sea water with salinity 33 PSU.

Study design

Test type: static

Water media type: saltwater

Limit test: no

Total exposure duration: 72 h

Test conditions

Test temperature: 20.6 - 21.4 °C

pH: 7.85 - 8.92

Nominal and measured concentrations

Sample nominal	Measured Sodium Chlorate	% of nominal
10 mg/l initial	5.1	51
10 mg/l end	4.3	43
22 mg/l initial	16.8	77
22 mg/l end	15.6	71
46 mg/l initial	37	81
46 mg/l end	36	78
100 mg/l initial	98	98
100 mg/l end	96	96
220 mg/l initial	211	96
220 mg/l end	198	90
460 mg/l initial	455	99
460 mg/l end	430	93
1000 mg/l initial	995	100
1000 mg/l end	944	94

Details on test conditions

TEST SYSTEM

- Test vessel: 30 ml glass vials, covered with plastic film. The culture volume was approximately 12 ml.

- Initial cells density: 5*10E6 cells/l

- Control end cells density: 1.23*10E9 cells/l

- No. of vessels per concentration (replicates): 3

- No. of vessels per control (replicates): 6

GROWTH MEDIUM

- Standard medium used: yes, ISO 10253 medium, prepared from filtered natural seawater

TEST MEDIUM / WATER PARAMETERS

- Source/preparation of dilution water: collected from 60 m depth in the Oslofjord

- Culture medium different from test medium: No

OTHER TEST CONDITIONS

- Sterile test conditions: yes

- Adjustment of pH: no

- Photoperiod: continuous light

- Light intensity and quality: 68 µM m-2 s-1.

- Salinity (for marine algae):

EFFECT PARAMETERS MEASURED (with observation intervals if applicable) :

- Determination of cell concentrations: Coulter Multisizer electronic particle counter with a 100 µ orifice tube

Reference substance (positive control): yes

Results and discussion

Effect concentrations:

Duration	Dose descriptor	Effect conc.	Nominal / measured	Conc. based on	Basis for effect
72 h	NOEC	>= 1000 mg/L	nominal	test mat.	growth rate
72 h	EC10	> 1000 mg/L	nominal	test mat.	growth rate

Details on results:

The results of the chemical analyses were not always within 80 to 120% of the nominals. The highest test concentration gave measured concentrations of 100 and 94% of the nominal concentration and no effects were seen upto and including the highest test concentration, therefore it is not considered to have an impact on the outcome of the study.

Reported statistics and error estimates:

Dunnett's test

Any other information on results incl. tables:

Mean cell density (10E6 cells/l) and calculated growth rates (d-1) after 72 hours

Concentration SODIUM CHLORATE	start	24 hours	49 hours	72 hours	0-72 h µ (d-1)

Control	5	45	322	1234	1.8
10 mg/l	5	50	367	1467	1.9
22 mg/l	5	47	370	1190	1.8
46 mg/l	5	46	358	1490	1.9
100 mg/l	5	45	321	1317	1.9
220 mg/l	5	45	334	1491	1.9
460 mg/l	5	45	340	1507	1.9
1000 mg/l	5	40	292	1215	1.8

Applicant's summary and conclusion

Validity criteria fulfilled: yes

Conclusions

The study is well conducted according to standard guideline, with chemical ananlysis and GLP.Sodium chlorate is not toxic to the marine algae Skeletonema costatum. The NOEC is equal to or greater than 1000 mg/l.

Executive summary

The inhibitory effect of Sodium Chlorate on the growth of the marine diatom Skeletonema costatum, strain NIVA BAC1, has been investigated. The test was performed according to ISO 10253: Marine algal growth inhibition test with Skeletonema costatum and Phaeodactylum tricornutum. A series of concentrations (10, 22, 46, 100, 220, 460 and 1000 mg/l) of the test material were prepared by dilution of a dispersion of the test material in algal growth medium. The solutions were inoculated with 5*10E6 cells/l of an exponentially growing culture of S. costatum. Three replicates of each concentration were incubated in 30 ml glass vials with 12 ml culture volume on a shaking table at 20 ±1 °C, under continuous illumination. Six replicate cultures in growth medium were used as controls. Growth was monitored by daily counting of cell numbers using a Coulter Multisizer M3. Test concentrations were verified by chemical analysis of chlorate in the test medium at start and end of the test. The test material caused no significant inhibition of the growth of S. costatum at concentrations up to 1000 mg/l. Therefore, NOEC is equal to or greater than 1000 mg/l.

7.6. Study 6 (study report 1994a)

ENDPOINT_STUDY_RECORD: 7775-09-9,Toxicity to aquatic algae and cyanobacteria, Study report,1994a, SS

Reliability

2 (reliable with restrictions)

Rationale for reliability incl. deficiencies

other: Test was not performed under GLP, some details missing on test condition, but described in enough detail.

Data source

Reference: Sodium chlorate: toxicity to the green alga Scenedesmus subspicatus. / study report

Materials and methods

Test guideline:

Qualifier according to OECD Guideline 201 (Alga, Growth Inhibition Test), before 23 March 2006

GLP compliance: no

Test material

Test material information: sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: no

Test organisms

Test organisms (species): Desmodesmus subspicatus (previous name: Scenedesmus subspicatus)

Any other information on materials and methods incl. tables

- An algae culture in the exponential growth phase was used as inoculum.

- Test temperature: 24 +/- 1 degree C

Results and discussion

Any other information on results incl. tables:

- Nominal test concentrations: 0, 49, 98, 196, 392, 784,1569, 3137 mg chlorate/L (is equal to 0, 62, 125, 250, 500, 1000, 2001,4001 mg sodium chlorate/l)

- Visual examination of the cultures at 48 and 72 hours indicated that algal cells were smaller and paler at 3137 mg/l compared to the control.

Based on visual observations:

NOEC = 1569 mg chlorate/L (2001 mg sodium chlorate/l)

LOEC = 3137 mg chlorate/L (4001 mg sodium chlorate/l)

Biomass: NOEC = 3137 mg chlorate/L (4001 mg sodium chlorate/l)

Growth rate: NOEC = 3137 mg chlorate/L (4001 mg sodium chlorate/l)

7.7. Study 7 (Study report 1994 b)

ENDPOINT_STUDY_RECORD: 7775-09-9,Toxicity to aquatic algae and cyanobacteria, Study report,1994b, SS

Administrative data

Adequacy of study: supporting study

Robust study summary: true

Reliability: 2 (reliable with restrictions)

Rationale for reliability incl. deficiencies

other: Test is not performed under GLP, but described in enough detail.

Data source

Reference: Sodium chlorate: toxicity to the marine alga Phaedactilum tricornutum. / study report

Materials and methods

Principles of method if other than guideline

Method: other

GLP compliance: no

Test material

Test material information: sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: no

Test organisms

Test organisms (species): Phaeodactylum tricornutum

Any other information on materials and methods incl. tables

- Test method was conducted according to own protocol based on the draft ISO standard 10253.

- Algae from a culture in the exponential growth phase were added to the test solutions.

- Nominal concentrations: 0, 50, 100, 200, 400, 800, 1600, 3200 mg chlorate/l (is equal to 0, 64, 128, 255, 510, 1020, 2041, 4082 mg sodium chlorate/l)

- Temperature: 20 +/- 1.0 degree C

Results and discussion

Any other information on results incl. tables

Biomass:

- NOEC = 50 mg chlorate/l (64 mg sodium chlorate/l)

- EbC50 = 298 mg chlorate/l (95% c.i. 177-468 mg chlorate/l) (380 mg sodium chlorate/l)

Growth rate:

- NOEC = 100 mg chlorate/l (128 mg sodium chlorate/l)

- ErC50 = 444 mg chlorate/l (95% c.i. 274-719 mg chlorate/l) (566 mg sodium chlorate/l)

7.8. Study 8 (study report 1994c)

ENDPOINT_STUDY_RECORD: 7775-09-9,Toxicity to aquatic algae and cyanobacteria, Rosemarin-Lehtinen,1994, S

Endpoint

toxicity to aquatic algae and cyanobacteria

Adequacy of study

other information

Robust study summary

false

Used for classification

false

Used for SDS

false

Reliability

3 (not reliable)

Rationale for reliability incl. deficiencies

other: see 'Remark' The test was not performed under GLP and not according to a standard protocol. Though certain aspects of the test are described in detail, there are parts which are not clear:

- Detailed information is missing on the test substance.

- It is not clear how many controls were tested and what the variability was in the results of the controls.

- The method of analysis is sensitive down to concentrations to about 0.5 mg ClO3-/l. Much lower concentrations were tested and it is not clear if and how these were analysed.

- Raw seawater was let into the pools. There are no details on the substances present in this water and it is not known if this water was treated before it entered the pools.

- For a long term study of 6 months it would be better to give a NOEC, in this case only EC50s are determined.

- Baltic sea has a low salinity (7 ppt) and is therefore brackish. The species tested are marine species and are

living in conditions of stress at low salinity. Therefore these circumstances are not optimal and can interfere with

the outcome of the test.

- Organisms which eat the algae are present in the pools and it is difficult to determine the influence of this behavior on the test results.

- Fucus vesiculosus on original stone substrate with associated organisms were put in the pools. It is not known

what these associated organisms were, whether a similar number was introduced into each pool and if they had an

impact on the test result.

Because of all these missing points the test is considered to be invalid.

Data source

Reference: Effects of pulp mill chlorate on Baltic Sea algae. / Rosemarin. A., Lehtinen. K.J., Notini. M., Mattson. J. / publication

Materials and methods

Test guideline

Qualifier

according to

Principles of method if other than guideline

Method: other: own method

GLP compliance

no

Test material

Test material information

sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring

no

Test organisms

Test organisms (species)

other: other algae: Fucus vesiculosus

Any other information on materials and methods incl. tables

Circular pools of 8 m3 were created with a sand base 5 cm thick. Different species were added to the pools. 10 l of transplanted Fucus vesiculosus with associated organisms was introduced. Other algae were introduced as well. 100 stickleback (Gasterosteus aculeatus) larvae and five juvenile flounders (Platichthys flesus) were added to each pool. Zostera marine was placed into the pools anchored by small stones.

With a continuous flow of seawater (48h-renewal period) the pools were set up in April and run for 2 months prior to exposure to effluents. Salinity was 7 ppt throughout the exposure period and temperature varied from 3 to 20 degree C.

The tests duration was 6 months.

Bleachery effluent containing different amounts of chlorate was removed from a kraft pulp mill on the Swedish east coast. Chlorate content of the effluents was determined using high pressure ion chromatography. This method is only sensitive down to about 0.5 mg ClO3-/l.

Three pools received the same amount of chlorate, one with chlorate alone, one with chlorate plus effluent from a pulp mill and one with effluent alone. One control was tested as well.

Apical growth of F. vesiculosus was measured in August and December. Net growth was measured by volume displacement at the beginning and the end of the test (beginning of the test is probably at the start of the 2 months pre-exposure period).

F. serratum and Chorda filum were monitored by determining wet weight.

Net ecosystems production in each pool based on continuous automated measurements of pH, O2 and temperature in the incoming and outgoing water from each pool while taking into account O2 and CO2 gas diffusion constants.

Treatments:

Dilution	Concentration (ug ClO3-/l)

Control	-	0
Effl B	2000	21
Chlorate	-	58
Chlorate+Effl A	4000	58
Effl A	2000	58
Effl B	400	105
Effl A	400	288

Results and discussion

Effect concentrations

Key result

false

Dose descriptor

EC50

Effect conc.

ca. 80 µg/L

Any other information on results incl. tables

The interaction of several species exposed to chlorate in marine pools was evaluated.

- For the apical growth of Fucus vesiculosus the EC50 was about 80 ug ClO3-/l. No NOEC was determined.

- For Fucus serratus the EC50 was 130 ug ClO3-/l. Growth was clearly inhibited in pools receiving the two highest

chlorate concentrations.

- Chorda filum disappeared from all pools containing chlorate, after three months. Its availability to grazers should be taken into account in assessing the sensitivity, since a net loss of material occurred even in the controls.

- Pilayella littoralis and Ectocarpus siliculosus only survived in one pool with the lowest chlorate concentration.

- The growth of colonial form of a blue-green algae Rivularia, was stimulated at the 3 highest chlorate concentrations in the other concentrations it grew as in the control.

- The filamentous Lyngbya was co-dominant in all pools and Anabaena occurred in all pools at the same level. So these algae are not deleteriously affected by chlorate.

- No inhibitory effect was found on the dominant species of the filamentous green algae and neither on two red algae species.

- Diatoms as a group were not affected either by chlorate.

- Zostera marine did not show an effect at any of the concentrations.

Net ecosystem productivity was negatively influenced. The EC50 was again around 80 ug ClO3-/l.

7.9. Study 9 (study report 1986)

ENDPOINT_STUDY_RECORD: 7775-09-9,Toxicity to aquatic algae and cyanobacteria, Rosemarin-Mattson,1986, S

Endpoint

toxicity to aquatic algae and cyanobacteria

Adequacy of study

other information

Robust study summary

false

Used for classification

false

Used for SDS

false

Reliability

3 (not reliable)

Rationale for reliability incl. deficiencies

other: The test was not performed under GLP and not according to a standard protocol.

It is not known if there were other substance present in the effluent which could have had an effect on F. vesiculosus.

Data source

Reference

Effects of pulp mill chlorate (ClO3-) on fucus vesiculosus - A summary of projects. / Rosemarin, A., Mattson, J., Lehtinen, K.J., Notini, M., Nylén, E. / publication

Materials and methods

Test material

Test material information

sodium chlorate / 7775-09-9 / 231-887-4

Test organisms

Test organisms (species)

other: other algae: Fucus vesiculosus

Results and discussion

Any other information on results incl. tables

Chlorate has deleterious effects on F. vesiculosus. It causes reduction in growth and viability at concentrations as low as 20 ug/l in the Baltic sea. Chlorate uptake appears to be stimulated by nitrate. This may result in a seasonal variation in chlorate uptake. The Fucus belt over an area of at least 12 km2 was severely affected by the effluent from a kraft pulp mill (which contained chlorate levels around 50 mg/l).

8. TOXICITY TO AQUATIC PLANTS OTHER THAN ALGAE

8.1. Study 1 (study report 2003)

ENDPOINT_STUDY_RECORD: 7775-09-9, Toxicity to aquatic plants other than algae, Study report, 2003, RS

Reliability

1 (reliable without restriction)

Rationale for reliability incl. deficiencies

other: Study generated according to generally valid and internationally accepted testing guideline and performed under GLP.

Data source

Reference: Sodium chlorate aquatic plant toxicity test, Lemna minor, static, 7 d. / study report

Materials and methods

Test guideline

Qualifier: according to OECD Guideline 221 (revised draft document 2002)

Deviations: yes, Swedish Standard (SIS) medium. Modified: NH4Cl was given as inorganic nitrogen source instead of NaNO3 to get sufficient recovery rates of concentration control analysis. growth sp. inhibition test"

GLP compliance: yes

Test material

Test material information: sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring: yes

Details on sampling:

At the start and the end of the test samples were taken for analyses.

Details on analytical methods:

HPLC, analytical determination was carried out on an anion exchange column using an external standard. Analysis was performed in isocratic mode with conductivity detection.

Test solutions

Vehicle: no

Details on test solutions:

- Dispersion treatment: Agitation

- Procedures: 320 mg/L prepared with dilution water

Test organisms

Test organisms (species): Lemna minor

Details on test organisms:

- Source/supplier: OekoTox Moser & Pickl Gbr, Stuttgart, Germany

- Laboratory culture: yes

- Method of cultivation: in 900 ml crystallization dishes, filled with 500 ml Swedish Standard (SIS) Medium.

Study design

Test type: static

Water media type: freshwater

Limit test: no

Total exposure duration: 7 d

Test conditions

Test temperature: 24 ± 1 °C

pH: 6.60-6.64 at the start of the test and 5.23-5.95 at the end

Nominal and measured concentrations:

- Nominal test concentrations: 0.0, 3.2, 10, 32, 100, 320 mg/L

- The measured concentrations of the test substance were between 90 and 107% of the nominal concentrations, therefore the nominal values were used for the calculations

Details on test conditions

Swedish Standard (SIS) medium, modified GROWTH/TEST MEDIUM CHEMISTRY: Swedish Standard (SIS) medium

- pH: 6.5 ± 0.2

- Exposure vessel type: Crystallization dishes with a volume of 500 ml, covered with glass tops and filled with 200 ml test solution.

- Number of replicates, plants per replicate: 3, 3 plants with 4 fronds each

- Intensity of irradiation: 6500-10000 lx (85-125 µmol/m2*s)

- Photoperiod: continuous fluorescent light

- Number of plants (start and end of the test), number of fronds (on day 0, 2, 5 and 7) and dry weight (at the end of the test).

On each observation day the pigmentation, destruction of roots etc. were determined.

Reference substance (positive control): yes Zinc chloride, 99.2% pure

Results and discussion

Effect concentrations:

Duration	Dose descriptor	Effect conc.	Nominal /	Conc. based on	Basis for effect	Remarks on

			measured			result
7 d	EC50	134 mg/L	nominal	test mat.	growth rate	other: 33.7- > 320 mg/L

Details on results

Details on results

Biomass growth (number of fronds):

EC50 = 73.7 mg/L (95% c.i. 34.5-158 mg/L)

LOEC = 32 mg/L NOEC = 10 mg/L

Growth rate (number of fronds):

EC50 = 134 mg/L (95% c.i. 33.7- > 320 mg/L)

LOEC = 32 mg/L NOEC = 10 mg/L

Biomass dry weight:

EC50 = 128 mg/L (95% c.i. 28.0- > 320 mg/L)

LOEC = 32 mg/L

NOEC = 10 mg/L

- Evaluation after day 7 (mean and standard deviation in brackets):

Conc (mg/L) log biomass Inhibition of integral log biomass (%)

control 7.49 (0.49)

3.2 8.37 (0.29) -11.74 (3.88)

10 8.30 (0.29) -10.75 (3.82)

32 6.12 (0.44) 18.31 (5.81)

100 3.25 (0.47) 56.58 (6.27)

320 0.24 (0.11) 96.83 (1.50)

Conc (mg/L) Specific Inhibition of growth rate specific growth rate (%)

control 0.350 (0.020)

3.2 0.366 (0.020) -4.45 (5.08)

10 0.355 (0.000) -1.44 (1.31)

32 0.289 (0.010) 17.29 (3.15)

100 0.204 (0.010) 41.74 (2.04)

320 0.099 (0.20) 71.70 (6.18)

Conc (mg/L) Doubling Biomass dw Inhibition of time of (mg) biomass dw (%) fronds (d)

control 1.98 (0.12) 26.0 (2.1)

3.2 1.90 (0.09) 29.5 (0.6) -4.87 (0.79)

10 1.95 (0.03) 23.4 (0.9) 4.21 (1.52)

32 2.39 (0.09) 15.4 (2.0) 20.37 (4.95)

100 3.40 (0.12) 8.4 (0.6) 43.92 (2.81)

320 7.00 (1.86) 4.2 (0.6) 70.78 (5.41)

- Other observations: In the three highest concentrations the fronds were lighter green and partly covered with white spots at day 2. At day 5 the fronts in 320 and 100 mg/L were partly without pigmentation and in 32 mg/L fronds were partly covered with white spots.

At day 7 fronds in the three highest concentrations were partly without pigmentation.

Results with reference substance (positive control)

- Concentrations: 0.0, 0.32, 1, 3.2, 10, 32 mg/L

- Results:

- EC50 (biomass growth) = 9.5 mg/L (95% c.i. 1.8-50.2 mg/L)

- EC50 (growth rate) = 5.2 mg/L (95% c.i. 1.1-24.6 mg/L)

- EC50 (biomass dry weight) = 10.9 mg/L (95% c.i. 3.2-37.1 mg/L)

Reported statistics and error estimates

ANOVA and Dunnett's test were used for determination of significant differences compared control replicates. If normality of equal variance failed Bonferroni t-test was carried out. Probit analysis were used for calculation of EC50.

Applicant's summary and conclusion

Validity criteria fulfilled: yes

Conclusions

Test conducted under GLP with analysis, analysis certificate and to relevant guideline. This study can be considered reliable without restrictions.

The NOEC based on biomass dry weight is 10 mg/l. The lowest EC50 is based on biomass growth an d is 73.7 mg/L (95% c.i. 34.5-158 mg/L).

Executive summary

The effects of the test item Sodium Chlorate on growth of the monocotyledon species Lemna minor were determined according to the principles of OECD-Guideline 221 (Revised Draft Document July 2002), from October 29 to November 05, 2003 at DR.U.NoACK-LABORATORIEN, Sarstedt, Germany. The test item (batch number 1E01 03WF) contained 99.66 % Sodium Chlorate. The aim of the study was to determine the effects of the test item on the growth of Lemna minor over 7 days under static exposure conditions. On the basis of a preliminary test, 5 concentration levels were chosen in a geometrical series with a dilution factor of √1 0: nominal: 3.2 - 10 - 32 - 100 – 320 mg/L.

Three replicates were investigated for the test concentrations and the control. Frond numbers were assessed on days 0, 2, 5 and 7. Inhibition of log biomass growth. specific growth rate and biomass dry weight were determined. The concentrations of sodium chlorate were analysed on day 0 (freshly prepared solutions) and on day 7 (old solutions) via HPLC. The recovery rates of the active ingredient chlorate were> 80 % at start and end of the study (for details of the analytical method and results please refer to chapter 10).

Environmental parameters like water temperature and pH-value were determined to be within the acceptable limits. The effects of sodium chlorate, based on nominal concentrations, are summarized in table 1.

Table 1: Effects on Lemna minor after 2, 5 and 7 d (based on nominal concentrations of the test item)

Inhibition of log Biomass growth [mg/ L]	2d	5d	7d	
Elog bC50value	91.3	60.5	73.7	
95 % confidence Interval for ElogbC50values	68.6 -122	38.6 -95.0	34.5 -158	
LOEC	100	100	32	

NOEC	32	32	10	
Inhibition of specific Growth rate [mg/L]				
ErC50 value	91.3	57.8	134	
95 % confidence Interval for ErC50values	68.6-122	35.8-93.3	33.7 ->320	
LOEC	100	100	32	
NOEC	32	32	10	
Inhibition of log Biomass dry weight [mg/ L]				
ElogdwC50value	n.a	n.a	128	
95% confidence interval forElogdwC50value	n.a	n.a	28.0->320	
LOEC	n.a	n.a	32	
NOEC	n.a	n.a	10	

n.a. =not applicable

9. TOXICITY TO OTHER AQUATIC INVERTEBRATES

9.1. Study 1 (Study report 1991 g)

ENDPOINT_STUDY_RECORD: 7775-09-9, Toxicity to other aquatic organisms, Study report, 1991g, RS

Reliability
2 (reliable with restrictions)

Rationale for reliability incl. deficiencies
other: see 'Remark' GLP study according to protocol. No chemical analyses were performed on the test concentration only the stock solution were analyzed. From results in chronic tests it can be assumed that the test concentrations were stable and that the organisms were exposed.
Data source

Reference
Acute Flow-through Mollusc Shell Deposition Test with Sodium Chlorate / study report

Data access
data submitter is data owner

Data protection claimed
yes

Materials and methods
555

Test guideline

Qualifier
according to

Guideline
other: U.S. EPA-FIFRA, Guideline 72-3

GLP compliance
yes

Test material

Test material information
sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring
yes

Details on sampling
The concentration of sodium chlorate test media could not be determined because of naturally occurring interferences in the dilution water. The available analytical methods were effective in deionized water used to prepare toxicant stock solution but not in the dilution water. Approximately 5 ml of the 500,000 mg/L primary stock solution (prepared in deionized water) was withdrawn from the toxicant reservoir, placed into 40 ml glass VOA vial, and transferred to analytical laboratory for determination of toxicant concentration.

Details on analytical methods
The analytical method was validated in deionized water at 1 to 25 mg/L. Several attempts were made to validate the method in dilution water without success. Interfering compounds, chlorate and nitrate present in the natural groundwater used as dilution water, interfere with the analysis by eluting near the sodium chlorate peak.
The use of columns other than the conductivity detector used for these analyses would not eliminate the problem because the large interferences of nitrate and chlorate would elute at the same time on any column.
Analytical samples including standards and blanks) were filtered through a 0.5 micron filter into an HPLC autosampler vial and analyzed using a 510 pump [2] high performance liquid chromatograph (Waters Model 680 controller, 431 conductivity detector, 712 Wisp, with and HP 3350 data system or equivalent).
Test solutions

Vehicle
no

Details on test solutions
Dilution water:
Water use for acclimation of test organisms and for all toxicity testing was unfiltered seawater collected from the Atlantic Ocean at EnviroSystems in Hampton, New Hampshire. Water, which had a salinity of 21 to 26 ppt (parts per thousand) and a pH of 7.7 to 8.0, was stored in 500-gallon polyethylene tanks where it was aerated.
Stock solution: 556
An initial 500,000 mg/L stock solution was prepared by combining 1,000.0 g of test substance a
nd deionized water in a 2 liter class A volumetric flask and adjusting the volume to approximately
1,950 ml with deionized water. The stock solution was mixed until the test substance dissolved on a magnetic stir plate and the total volume was brought up to 2.0 L with deionized water. This procedure was repeated three more times to produce a total of 6 L more of the 500,000 mg/L stock solution.
Appropriate amounts of the stock solution were added directly to dilution water by a proportional diluter (5.0 ml of test substance was combined with 2,500 ml of water during each diluter cycle) and this diluter toxicant cell solution was mixed by a high shear pump equipped with a Teflon head.

Test organisms

Test organisms (species)
other: Crassostrea virginica

Details on test organisms
Juvenile oysters employed as test organisms were from a single source and were identified using an appropriate taxonomic key. They were procured from the Aquatic Research Organisms division of Resource Analysts Inc. in Hampton, New Hampshire and acclimated to test conditions in unfiltered seawater for more than 10 days. Prior to testing oysters were maintained in 100% dilution water under flow through conditions. During acclimation oysters were not treated for disease and they were free of apparent sickness, injury, and abnormality at the beginning of the test. Oysters were supplied with live marine phytoplankton to supplement the available food in the unfiltered natural seawater that was used as dilution water and for acclimation. During the last 10 days of acclimation the temperature ranged from 18.6 - 21.2 °C, salinity was maintained at 25 to 26 parts per thousand, and dissolved oxygen was maintained above 7.4 mg/L.
Oysters were 25 to 50 mm in height (measured along the long axis). At the initiation of the test, each oyster was ground to remove approximately 3 to 5 mm of shell and form a smooth edge.

Study design

Test type
flow-through

Water media type
saltwater

Limit test
no

Total exposure duration
96 h

Test conditions

Test temperature
20.0 to 23.4°C

pH
7.7 to 8.0

Dissolved oxygen
7.2 to 7.5 mg/L

Salinity
21 to 24 parts per thousand

Nominal and measured concentrations
Nominal concentrations: 0 (control), 70, 120, 250, 500, and 1,000 mg/L sodium chlorate.
The 500,000 moiL (nominal) stock solution had a measured concentration of 480,000 mg/L sodium

chlorate.

Details on test conditions
- Photoperiod: A 16 hour light and 8 hour dark photoperiod was automatically maintained
- Light intensity: with cool-white fluorescent lights that provided a light intensity of 18 pEs- 1/m2
- Aeration: Aeration was not required to maintain dissolved oxygen concentrations above acceptable levels.
- Feeding: Oysters were supplied with live marine phytoplankton to supplement the available food in the unfiltered natural seawater that was used as dilution water.

Reference substance (positive control)
no

Any other information on materials and methods incl. tables
Twenty oysters were indiscriminately and equally distributed among a single replicate of each treatment.
The test was performed in 20 liter glass aquaria that contained 15 liters of test solution (water depth was approximately 18 cm). Test vessels were randomly arranged in a water bath during the 96 hour test (a random numbers table was used to select the location of each vessel). The test substance was supplied to the test vessels under flow through conditions by an intermittent flow proportional diluter. The diluter, which was constructed at EnviroSystems, allowed test media to contact only glass or Teflon-coated surfaces. The diluter was calibrated before and after the test. During the test the diluter was activated 1,007 times, resulting in an average of 16.8 media exchanges per 24 hours in each test vessel and an average of 0.5 liters per oyster per hour.
The number of surviving organisms and the occurrence of sublethal effects were determined visually and recorded after 0, 24, 48, 72, and 96 hours. At the end of the study oysters were removed from test vessels and the longest finger of new growth was measured to the nearest 0.1 mm with a Manostat caliper.

Results and discussion

Effect concentrations

Duration	Dose descriptor	Effect conc.	Nominal / measured	Conc. based on	Basis for effect
96 h	EC50	> 1000 mg/L	nominal	test mat.	other: shell growth
96 h	LC50	> 1000 mg/L	nominal	test mat.	mortality

Details on results
One hundred percent survival occurred in the control and no sublethal effects were noted during the exposure period.
One oyster died in 250 mg/l, but this was not concentration related.
Reported statistics and error estimates
Results of the toxicity test could not be interpreted by standard statistical techniques (Stephan, 1983) because greater than 50% control shell deposition occurred at all tested concentrations.

Any other information on results incl. tables
Shell growth data from toxicity test with sodium chlorate

Conc. (mg/l)	0.0	70	120	250	500	1000
Mean (mm)	2.0	2.4	2.0	1.8	1.7	1.4
St. dev.	0.8	1.1	0.7	1.1	0.9	0.8
% of control	100.0	120.0	100.0	90.0	85.0	70.0

Overall remarks, attachments

Applicant's summary and conclusion

Validity criteria fulfilled
not specified

Conclusions
The 96h EC50 (based on shell growth) and LC50 were greater than the highest test concentration of 1000 mg/l.

Executive summary
The effect of sodium chlorate on shell deposition by the eastern oyster, Crassostrea virginica, is
described in this final report. The test was conducted for Albright and Wilson Americas for 96 hours during February 22 to 26, 1991, at the EnviroSystems Division of Resource Analysts, Inc. in Hampton, New Hampshire. It was conducted by Peter Kowalski, Ellen Stanford, Jeanne Magazu, Robert Boeri, and Timothy Ward according to the protocol developed for EnviroSystems Study Number 90116-DE.
The analytical portion of this study was conducted under the supervision of Gloria Switalski. Sodium chlorate (reported purity >= 99% active ingredient) was supplied by the sponsor. The test was performed under flow-through conditions with five concentrations of test substance and a dilution water control at a temperature of 15 to 30°C. The dilution water was unfiltered natural seawater collected at Hampton, New Hampshire. Nominal concentrations of sodium chlorate were: 0 mg/L (control), 70, 120, 250, 500, and 1,000 mg/L. Nominal concentrations were used for all calculations. Oysters used in the test were procured from a commercial supplier (the Aquatic Research Organisms division of Resource Analysts, Inc., Hampton, New Hampshire) and acclimated under test conditions for more than 10 days. All animals were in good condition at the beginning of the study. Exposure of oysters to the test substance resulted in a 96 hour EC50 greater than 1,000 mg/L sodium chlorate.

9.2. Study 2 (Study report 2010b)

ENDPOINT_STUDY_RECORD: 7775-09-9, Toxicity to other aquatic organisms, Study report, 2010b, RS

Reliability
1 (reliable without restriction)

Rationale for reliability incl. deficiencies
other: well conducted study according to standard protocol with chemical analyses and GLP.

Data source
Reference
Sodium Chlorate Effect on reproduction to the marine rotatoria Brachionus plicatilis / study report

Data access
data submitter is data owner

Data protection claimed
yes, but willing to share

Materials and methods
561

Test guideline
Qualifier
according to

Guideline
other: ISO/DC 20666

Deviations
yes test duration is 96h and performed in continuous light

Principles of method if other than guideline
The toxicity test was conducted with 8 wells with 1 animal in each for each test concentration, 16 control wells were used for control. The test animals were less than 24 hours old. Newly hatched rotifers were selected based on shape and size. A 24 Multiwell plate was used for incubation. A concentrated culture of Nannochloropsis oceanica and Tetraselmis suecica were added to each well at the start to give an algal cell concentration of >3x10E6 cells/ml.
GLP compliance
yes

Test material
Test material information
sodium chlorate / 7775-09-9 / 231-887-4

Sampling and analysis

Analytical monitoring
yes

Details on sampling
Sodium Chlorate levels were analysed in the test solution before addition of test organisms and feed algae suspension and at the end of the test.
Samples were filtered through 0.2 µm disposable cellulose acetat filters and stored at 4 °C in 20 ml scintillations vials. Storage time was less than 15 days for all samples.

Details on analytical methods
Preparation of a 1000 mg/l stock of the test substance by weighing 1 g of test compound and dissolving it in 1 liter of aged seawater. The stock solution was stored at 4 °C and was less than 1 week old when used for establishing a calibration curve (the stock solutions were actually only one day old.
Personal communication with the SD). From this stock a series of standard solutions were made to cover the test concentrations of 1 to 1000 mg/l of Sodium Chlorate. Because of the high amount of chlorine in seawater both standards and test solutions were diluted 100x before analysis. A simultaneous calibration with SO4 gave that the response factor for Chlorate relative to SO4 was 0.5365. The linearity of the calibration curve had correlation coefficient of 99.948 % with an intercept of -0.001 and slope value of 1.349 on the day samples were analysed.
Instrument DX 320 Dionex Ionchrometograph including: 2 modules of IC25, an Dionex Ionpac CS16 separation column for cations, an Dionex Ionpac AS15 separation column for anions, an Dionex Ionpac CG16 precolumn for cations, Dionex Ionpac AG15 precolumn for anions, Dionex Ionpac ATC-HC trap column for anions and 2 x EG40 Eluent generators.
Ion Cromatograph
- Column: Dionex IonPack AS15 3mm (id) x 150 mm
- Eluent Cartrigde: KOH Eluent cartridge from Dionex art no 058900
- Gard column: Dionex IonPac AG15 3mm(id) x 150 mm
- Gas: 99.999 % pure N2
- Injection sample volume: 200 µl
- Eluent flow: 0.7 ml/min
- Electrical Current: 100 mA
- Detector temperature: 27 °C
- Eluent: KOH

Test solutions

Vehicle
no

Details on test solutions
The test concentrations were prepared by diluting a stock solution of 1000 mg/l of Sodium Chlorate with appropriate amount of aged sea water.

Test organisms
Test organisms (species)
other: brachionus plicatilis

Details on test organisms
Source: SINTEF, Norway
Stock culture: Cultured in natural seawater fed with Nannochloropsis oceanica weekly batches, temperature 20 °C and 25 PSU
Lifestage/Age: Newly hatched rotifers, less than 24 h old
Adaptions to ISO/DC 20666, incorporated to use Brachionus plicatilis as test organism Brachionus
plicatilis is a marine rotifer of the same genus as freshwater rotifer Brachionus calcyflorus used in the ISO/DC 20666 guideline. However, Brachionus plicatilis has lower growth and reproduction rate and do not thrive at 26 °C. Therefore, the testperiod was extended to 96 hours and incubation temperature lowered (20 °C) compared to the guideline in order to achieve good reproduction. Best results were achieved when algae are in suspension; incubation in light gave more motile algal cells.

Study design
Test type: static
Water media type: saltwater
Limit test: no
Total exposure duration: 96 h

Test conditions
Test temperature: 20.1 - 20.5 °C
pH:7.89 - 8.16
Dissolved oxygen:> 7.7 mg/L

Nominal and measured concentrations

Sample	nominal	Measured Sodium Chlorate	% of nominal			
10 mg/ l	initial	8.2	82			
10 mg/ l	End	6.6	66			
22 mg/l	initial	l 19.3	88			
22 mg/l	End	16.8	76			
46 mg/l	initial	42	92			
46 mg/l	End	35	77			
100 mg/l	initial	99	99			
100 mg/l	End	92	92			
220 mg/l	initial	218	99			
220 mg/l	End	185	84			
460 mg/l	initial	471	102			
460 mg/l	End	392	85			
1000 mg/l	initial	1031	103			
1000 mg/l	End	865	86			

Details on test conditions

TEST SYSTEM
- Test vessel: 24 Multiwell plate
- No. of organisms per vessel: 1
- No. of vessels per concentration (replicates): 8
- No. of vessels per control (replicates): 16

TEST MEDIUM / WATER PARAMETERS
- Source/preparation of dilution water: Aged sea water was used as control medium and for dilution of Sodium Chlorate. The sea water was sieved (1 µm) in order to exclude particles and other organisms.
- Culture medium different from test medium: No

OTHER TEST CONDITIONS
- Adjustment of pH: No
- Photoperiod: continuous light
- Light intensity: not known

EFFECT PARAMETERS MEASURED (with observation intervals if applicable) : number of offspring using a low power stereoscopic microscope, mortality

Reference substance (positive control)
yes 3,5-dichlorophenol

Results and discussion

Effect concentrations

Duration	**Dose descriptor**	**Effect conc.**	**Nominal / measured**	**Conc. based on**	**Basis for effect**	**Remarks on result**
96 h	NOEC	46 mg/L	nominal	test mat.	other: reproduction	
96 h	EC10	21.9 mg/L	nominal	test mat.	other:	other: 95% CL

					reproduction	6.8 - 55.6 mg/L
96 h	EC50	596	nominal	test mat	other: reproduction	other: 95% CL 417 - 1215 mg/L

Remarks on result
Details on results
Chemical analysis
Initial test concentrations are measured to be between 82 and 103 % of nominal. After test end the test solutions were collected and filtered before chemical analysis. Test concentrations were then measured to be between 66 and 92 % of nominal. Most test concentrations showed a decrease of approximately 15 % from start to end.
Almost all mean concentrations were > 80% of the nominals. Only the lowest test concentration was below 80% (74%), but this is not a critical concentration, therefore nominals were used for effect calculations.
Results with reference substance (positive control)
EC50 = 5.6 mg/L for growth inhibition.
Prior tests have shown EC50 in the range of 4.2-5.6 mg/l. The EC50 is quite similar to that found for the algae Skeletonama costatum (2.0 mg/l) and Acartia tonsa (1.0 mg/l).
Reported statistics and error estimates
NOEC was determined using the Dunnett's test with JMP statistical package of SAS institute
(1989-1997). EC10 and EC50 values were determined using logistic regression analysis.

Any other information on results incl. tables
Number of offspring after 96 hours exposure to Sodium Chlorate.

Replicate no	Control	1000 mg/ L	460 mg/ L	220 mg/L	100 mg/L	46 mg/L	22 mg/L	10 mg/L
1	7	4	3	5	4	7	6	7
2	7	2	3	5	3	7	7	7
3	6	2	3	3	4	6	8	8
4	6	3	4	5	7	5	7	7
5	7	5	6	3	6	5	6	7
6	6	2	5	4	3	5	6	8
7	6	2	4	3	5	6	7	8
8	6	2	7	5	4	4	4	6
9	7							
10	6							
11	8							
12	8							
13	7							
14	7							
15	9							
16	8							
Total	111	22	35	33	36	45	51	58
Average	6.9	2.8	4.4	4.1	4.5	5.6	6.4	7.3
% inhib	0.0	60.4	36.9	40.5	35.1	18.9	8.1	-4.5

Applicant's summary and conclusion
Validity criteria fulfilled
yes
Conclusions
The test was well conducted according to standard protocol with chemical analysis and GLP.
Sodium chlorate is not very toxic to the marine rotifer Brachionus plicatilis.
Executive summary
The effect of Sodium Chlorate on the reproduction to the marine rotatoria Brachionus plicatilis has been investigated. The test was performed according to ISO/DC 20666- Water quality – Determination of the chronic toxicity to Brachionus calcyflorus in 48h. The test concentrations were 10, 22, 46, 100, 220, 460 and 1000 mg/l of Sodium Chlorate. Aged seawater was used as control in the test. The test was performed with 8 replicate vessels with 1 animal in each, for each test concentration and 16 control replicate vessels. The vessels were incubated for 96 hours at 20 ±1 °C. Mortality and number of off-springs were recorded after 96 hours.
There was a dose dependent reduction in reproduction observed for B. plicatilis, when exposed to
Sodium Chlorate. Statistical assessment indicates a NOEC of 46 mg/l Sodium Chlorate. EC50 was
estimated to be 596 mg/l Sodium Chlorate. Mortality of parent rotatoria was not observed at any
concentration. The test indicates that high concentration of Sodium Chlorate exerts a reproductive inhibition on B. plicatilis.