

Degussa 

8EHQ-0497-13910

Degussa
Corporation

CERTIFIED MAIL R.R.R

April 7, 1997



8EHQ-97-13910

OPPT Document Control Officer
East Tower, Room G-99 [7407]
U.S. Environmental Protection Agency
401 M Street, S.W.
Washington, D.C. 20460
Attn: Mr. James F. Darr
TSCA § 8(e) Coordinator

Contains No CBI

RE: TSCA § 8(e) Submission
Triallycyanurate (CASRN 101-37-1)

Dear Mr. Darr:

Degussa Corporation (Degussa) has determined that the results of several studies performed with the substance known as Triallycyanurate or "TAC" (CASRN 101-37-1), recently received from Degussa's parent company in Germany, Degussa AG, may be reportable as substantial risk information under TSCA § 8(e). In reviewing the results of the test identified herein, Degussa relied on the TSCA § 8(e) reporting guidance contained in EPA's "TSCA § 8(e) Reporting Guide" that was issued in June of 1991 in conjunction with the implementation of EPA's TSCA § 8(e) Compliance Audit Program ("CAP"). Degussa was one of the companies that participated in the TSCA § 8(e) CAP (ref. 8ECAP-0081). The titles of the three studies that are the subject of this TSCA § 8(e) submission are shown below.

1. Triallycyanurate ("TAC"): 4-Week Oral Toxicity Study after Repeated Administration in Rats and a Subsequent 6-week Recovery Period - Report.
2. Bericht über die Untersuchung von Triallycyanurat (TAC) auf Abwasserverhalten (Reaktion, Belastung, toxikologisches Verhalten).
3. Biodegradability of Triallycyanurate According to OECD 301B (modified Sturm-Test).

TAC is a commercial chemical the Degussa imports into the U.S. from Degussa AG. Degussa interprets the 28-day subchronic study results to indicate that TAC is moderately toxic and may cause neurotoxic effects and changes in blood chemistry. These conclusions, coupled with the fact that TAC is a commercial product, are the basis for Degussa's decision to report the results of the test under TSCA § 8(e).



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The results of the acute aquatic toxicity test support a conclusion that TAC is moderately toxic to fish. The results of the modified Sturm test to determine the biodegradability of TAC indicate that the substance may not be readily biodegradable and therefore may bioaccumulate. These conclusions, coupled with the fact that TAC is a commercial product that may potentially be released to surface waters, constitute Degussa's basis for reporting the results of these studies under TSCA § 8(e).

As a final point, Degussa is now reviewing its current Material Safety Data Sheet (MSDS) for TAC to determine if it should be modified or revised in light of the results of the aforementioned studies.

Degussa has prepared the following summaries of the results of the tests that were received from Degussa AG.

1. **28 day oral gavage study:**

Test substance: Triallylcyanurate (TAC), solid, purity 99.7%

Study Design

28 day repeated dose oral gavage study with an additional 4 week recovery period according to OECD guideline 407 (1) and under GLP conditions.

Dosage/route/duration:

Groups of 5 Wistar rats of each sex were treated once daily, 7 days per week with 2.15 ml of solutions of the test substance in peanut oil at concentrations of 8.25, 26.1 and 82.5 or 100mg/ml, corresponding to doses of 17.8, 56.2 and 178 or 215 mg/kg bw. by gavage. The high dose was changed to 215 mg/kg from day 22 of the study until termination. Two control groups, one solvent control and one receiving water by gavage were included in the study. The recovery group (recovery period: 6 weeks) consisted of 5 high dose animals and 5 vehicle controls of each sex.

Results

No increase in mortality was reported in the treated animals. Only small decreases in body weight gain were observed in male animals of the high dose group.

In the high dose group the following clinical signs were observed:

Coordination disturbance, decreased muscle tone, loss of righting reflexes, salivation. Clonic convulsions were observed at single time points in individual animals. In the medium dose group the effects were restricted to coordination disturbances observed directly after dosing and being reversible within 60 min to 4 hours. No effects were observed in the medium dose group between day 10 and 21 of treatment. No effects were observed in the animals of the recovery group from the first day of recovery to

the end of the observation period. These clinical symptoms indicate that TAC seems to reversibly affect the central nervous system at dose levels at or above 56.2 mg/kg bw.

Examination of reflexes, eyes, hearing and teeth did not show any substance related effects.

Hematology parameters did not differ significantly from controls. Statistically significant changes in clinical chemical parameters were reported in the high and medium dose groups including a slight increase in alanine amino transferase activity in males and females of the high dose group in week 4. Increased inorganic phosphate, serum ion, and sodium levels as well as decreased albumin levels were reported in high dose group males. Albumin levels were also slightly decreased and inorganic phosphate slightly increased in high dose group females.

At the end of the 6 week recovery period only males showed deviation statistically significant from controls with slightly decreased values in triglycerides, alanine amino transferase, cholinesterase, glutamate dehydrogenase levels compared to controls.

No macroscopical treatment related pathological changes were observed. Relative weights of the following organs in high dose males were increased: brain, liver, kidneys, adrenals. In female rats of the high and mid-dose groups significant increases were detected in relative and absolute liver weight and in the high dose group in absolute and relative adrenal weight and relative weight of the right kidneys. At the end of the recovery period no significant differences in organ weights between treated and control animals were observed. Microscopical changes were only seen in livers of high dose animals with a marginally increased incidence of microgranulomas. However, the incidence and severity of this lesion was within the normal range of this species. The effect on the liver may be regarded as an adaptation phenomenon rather than a true adverse effect.

All changes were fully reversible within the 6 weeks recovery period.

A NOEL of 17.8 mg/kg was identified (based on clinical signs of toxicity)

2. **Acute toxicity to fish**

Test substance: Triallylcyanurate (TAC)

Study Design

A 48 h fish toxicity study was conducted with *Leuciscus Idus melanotus* (golden orfe) in different dilutions under static conditions according to the German DIN 38 412 part 20 method. No GLP was applied. Five fish per concentration were used. As a stock solution for the dilutions a saturated solution of 250 g TAC/51 water stirred at 40 degrees C for 2 hours and then filtrated was used. The test was carried out without adjustment of pH (initial pH ca. 7).

Results

At a concentration of about 3.9 mg/l no mortality was observed (LCO). At 7.8 mg/l 80% of the fish died within 48 h (LC30). The results are calculated from the nominal concentration of the test substance and no analytical determination of the concentration was performed.

Comment: It cannot be excluded that the toxicity observed is due to hydrolysis products.

3. **Biodegradation**

Test substance: Triallylcyanurate (TAC)

Study design

A modified Sturm Test was conducted according to OECD guideline No. 301 B for ready biodegradability, duration: 28 days and under GLP.

Activated sludge was taken from an oxidation ditch in Delft, Netherlands for domestic sewage treatment. Concentration of the test substance in the medium was 12 and 24 mg/l respectively. Sodium acetate (100 mg/l) was used as reference compound to determine inoculum activity. A control series without test substance was used in order to allow correction for background carbon dioxide production by the inoculum. In order to detect possible toxic effects of the test substance, sodium acetate was also added to further duplicate bottles with 12 and 24 mg/l of the test substance. Carbon dioxide development was determined by trapping the developed gas with 0.4 M sodium hydroxide and titration with 0.1 M hydrogen chloride solution after 7, 14, 21 and 28 days.

Results

TAC was not readily biodegradable in this test as only small amounts of carbon dioxide were produced within 28 days. The control tests showed that the activity of the inoculum was sufficient and TAC had no significant effects on the activity of the inoculum.

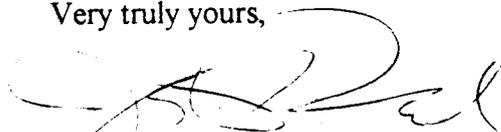
Evaluation of the ecotoxicological data

As TAC is not readily biodegradable and is moderately toxic to fish, it may pose a hazard to the environment when it enters the aquatic compartment. However, as the substance has a octanol-water partition coefficient of below 3 (2.8 determined according to OECD No. 107, Degussa AG, unpublished report no. 89-0148-DGP). It is not expected to bioaccumulate to a high extent and the hazard may be reduced by its low to moderate bioaccumulation potential.

Copies of the oral gavage and biodegradation studies are enclosed. The fish toxicity report is in German, and a copy is not enclosed. Degussa will provide a copy of an English translation of the fish toxicity report upon request.

If you have any questions regarding the contents of Degussa's TSCA § 8(e) submission on TAC, please do not hesitate to contact me at (201) 807-3161.

Very truly yours,

A handwritten signature in black ink, appearing to read "Jayne A. Pritchard". The signature is fluid and cursive, with a large initial "J" and "P".

Jayne A. Pritchard, Esq.
Regulatory Compliance Attorney

Enclosures

DEGUSSA AG - US-IT - NR.

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TRIALLYLCYANURATE (TAC)

4-Week Oral Toxicity Study after
Repeated Administration in Rats
and a Subsequent 6-Week
Recovery Period

REPORT

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August 24, 1992

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The report comprises 254 pages.

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1. GENERAL INFORMATION

1.1 Study Specification

Sponsor : Degussa AG/ZN Wolfgang
Industrielle Toxikologie
Postfach 13 45
D-6450 Hanau 1

Testing Facility : ASTA Medica AG
Institute of Toxicology
Kantstraße 2
D-4802 Halle-Künsebeck

Study Director : Dr. med. vet. H.-J. Zeche
ASTA Medica AG
Institute of Toxicology
Kantstraße 2
D-4802 Halle-Künsebeck

Further Testing Facility Involved (Analytical Investigations) : Degussa AG/ZN Wolfgang
Department IC AT0
Postfach 13 45
D-6450 Hanau 1

Test Substance : Triallylcyanurate (TAC)

Objective : Evaluation of the toxicity after 4 weeks of repeated daily oral administration in rats and a subsequent 6-week recovery period.

Test Guidelines : OECD Guideline No. 407 (1)
EEC Guideline (86/449/EEC) (2)

Study No. : 871154

The Schedule : Protocol : Jan. 10, 1990
Amendments : 4
First day of substance administration : Jan. 30, 1990
Start of autopsy :
Groups 1 - 3 : Feb. 27, 1990
Recovery animals : Apr. 10, 1990
Report : Aug. 24, 1992

Quality Assurance : The study was performed according to the principles of Good Laboratory Practice (GLP) (3).

Archivation : The approved protocol, all raw data obtained in the course of the study, all preserved tissues, paraffin blocks, slides, blood smears and a sample of the test substance as well as a copy of the final report are kept in the archives of the Institute of Toxicology for at least 30 years (starting with the report date). Afterwards the sponsor will decide on further use. The preserved tissues and the sample of the test substance may be discarded as soon as an inspection indicates that their quality does not allow any further evaluation. Data on analytical investigations are retained by sponsor.

Personnel of the Testing Facility :

Study Director,
Veterinary Care,
Clinical
Investigations : Dr. med. vet. H.-J. Zeebel

Autor of Report : Dr. rer. nat. K. Berthold

Clinical Pathology,
Necropsy,
Histopathology : Dr. J. H. Harleman DVS, PhD

Study Performance : M. Wellerdiek

Quality Assurance : K. E. Fichtner

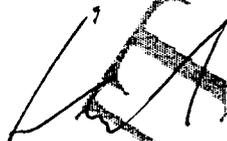
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1.2 Authentication

I, the undersigned, hereby declare that to the best of my knowledge the study, respectively all parts carried out in the testing facility, was performed under my supervision in accordance with the current OECD Principles of Good Laboratory Practice.

This report represents a true and accurate record of the results obtained and these results are the basis of the conclusion derived thereof.

Study Director


: Dr. med. vet. H.-J. Zechel

Report Review

Head of Institute
of Toxicology


: Dr. med. vet. W. Jahn

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1.3 Signature List

Author of Report : *V. B. Kucel*
Dr. rer. nat. K. B. old

Clinical Pathology,
Necropsy,
Histopathology : *[Signature]*
Dr. H. Harleman DVS, PhD

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1.4 ASTA Medica AG
Corporate Quality Assurance
Section GLP/GCP

Quality Assurance Statement

The non-clinical laboratory study

Triallylcyanurate (TAC)

4-week oral toxicity study after repeated administration to rats and a subsequent 6-week recovery period

performed at ASTA Medica AG, Bielefeld,

was inspected and audited for conformance to the principles of Good Laboratory Practice (GLP).

The dates of inspections and reports to study director and management are given below.

<u>Phase of Study</u>	<u>Date of QA Inspection</u>	<u>Date of QA Report</u>
Protocol Review	Jan. 18, 1990	
Experimental Period	Jan. 22, 1990	Jan. 22, 1990
	Feb. 1, 1990	
	Feb. 8, 1990	
	Feb. 20, 1990	Feb. 20, 1990
	Feb. 28, 1990	
	Mar. 29, 1990	Mar. 30, 1990
Final Report Audit	Apr. 9, 1990	
	Apr. 11, 1990	Apr. 12, 1990
	July 9, 1992	July 13, 1992
	Sep. 21, 1992	Sep. 21, 1992

Date: Sept. 22, 1992

Signed: _____

K. E. Fichtner

2. SUMMARY, EVALUATION, and CONCLUSION

General

To examine the oral toxicity of triallylcyanurate (TAC) after repeated administration, the test substance was given to 3 groups of Wistar rats for 28 days by gavage. The dose regimen was 17.8, 56.2, and 178 mg/kg b.w.. The high dose had to be changed to 215 mg/kg during the study. Additionally there were 2 groups of control animals, of which one was treated with tap water while the other received the vehicle (peanut oil). Each group consisted of 5 male and 5 female rats. In the vehicle control group as well as in the high dose group additionally 5 animals of each sex were included to serve as recovery animals. These rats were observed for further 6 weeks without administration of the test material after the end of the 28-day treatment period.

ResultsClinical Investigations

Common signs of toxicity were coordination disturbances, decrease of muscle tone, loss of righting reflexes (dorsal position), and salivation. Additionally clonic convulsions were detected and individuals also displayed chromodacryorrhea and piloerection.

Signs were present only in groups 4 (56.2 mg/kg) and 5 (178/215 mg/kg) during the treatment period.

None of the rats died in the course of the study.

Only a minimal reduction in food consumption was recorded in group 4 males during the first 2 weeks of treatment. In group 5 the food consumption of males as well as females was reduced during the first week of treatment by about 20% as compared to the vehicle control group, but increased to almost normal values within one week. In males there was another slight decrease after the dose had been increased to 215 mg/kg at the beginning of week 4.

After the end of the administration period food consumption rapidly increased in treated and vehicle control rats to reach or even exceed the values of the water control groups within one week.

In contrast to these changes in food consumption the body weight gain was almost not affected by the test substance administration. Only males of the high dose group slightly fell behind the other groups during the treatment period. In females no differences could be detected.

At examinations of reflexes, eyes, hearing, and teeth there were no substance related effects recorded.

Clinical Pathology

The examination of hematology parameters in week 4 revealed only a marginal decrease in the number of platelets in males of the high dose group.

Clinical chemistry parameters did show statistically significant deviations from the control values predominantly in groups 4 and 5. In week 4 a slight increase in the activity of alanine aminotransferase was observed in males and females of group 5. The values for inorganic phosphate, serum iron, and sodium were also increased slightly in high dose males, whereas albumin levels were slightly decreased. In female group 5 animals also albumin was slightly decreased and inorganic phosphate slightly increased. At the end of the 6-week recovery period only males showed deviations of statistical significance from controls. The values for triglycerides, alanine aminotransferase, cholinesterase, and glutamate dehydrogenase were decreased slightly.

Analysis of the urine in week 4 did not reveal any alterations.

Pathology

Macroscopical examination revealed no treatment related changes.

At the end of the treatment period the absolute weights of spleens were minimally decreased in males of group 5 only. At the same time the relative weights of a number of organs of these animals were increased. The organs affected were brains, livers, kidneys, and adrenals.

In the female rats significant increases were detected in the relative as well as absolute weights of livers in groups 4 and 5 and in the adrenals of group 5 as well as in the relative weights of the right kidneys.

At the end of the recovery period significant differences in organ weights or organ/body weight ratios were no longer present.

Microscopical examination revealed no treatment related changes in any of the organs examined, except for the liver. High dose animals of both sexes had a marginally increased incidence of microgranulomas in the liver (microgranulomas are small focal aggregates of predominantly macrophages and mononuclear cells, occasionally containing individual necrotic hepatocytes. It is generally considered a reaction to cell turnover).

The incidence and severity of this lesion was however within normal range for this species. It may however explain the slightly increased values of alanine aminotransferase and glutamate dehydrogenase observed in the high dose group animals.

EVALUATION

From the results of this 4-week oral toxicity study on triallylcyanurate (TAC) in rats it is obvious that the test material predominantly induces central nervous effects and impedes the ability of the animals to move in a normal fashion. This is especially true at the high dose of 178 mg/kg b.w..

There also seems to be a rather fast mechanism of adaptation, which is the cause of the decreasing effects during the course of the study and made necessary the rise of the high dose to 215 mg/kg in week 4. This adaptation was detected in food consumption also, which was impaired only during the first week of treatment. The steep increase in food consumption after the end of the administration period may be explained by a cessation of the energy supply by the vehicle (peanut oil).

A marginal reduction in the mean number of platelets in males of group 5 (178/215 mg/kg) was noted. This value nevertheless is within the normal range of the species. As no dose relation existed and the change was only mild, the effect is not caused by the treatment.

Clinical chemistry revealed slight increases in alanine aminotransferase activity in both males and females of group 5. This finding is in good correlation with the result of the histopathological examination. The change was fully reversible within the 6-week recovery period. Similar small effects were detected for inorganic phosphate and albumin in both sexes, while iron and sodium were affected only in males.

The increases in relative weights of livers of males of group 5 as well as the absolute and relative increased liver weights in females of groups 4 and 5 can be ascribed to an induction of liver enzymes. This also would be consistent with the adaptation phenomenon observed during the treatment period. For the increases in organ/body weight ratios of kidneys and adrenals in males and females of group 5 a treatment relation cannot be excluded. Because no histomorphological correlation was found, this finding cannot be explained on the bases of the present study.

All changes were fully reversible within the six week recovery period.

Microscopical examination revealed no treatment related changes in any of the organs examined, except for the liver. The liver had a marginally increased incidence of microgranulomas in high dose group animals of both sexes.

CONCLUSION

According to the results of this study the central nervous system and the liver are considered to be target organs after oral administration of triallylcyanurate (TAC). The toxic action of the test substance is characterized by an impairment of the coordination of movements with loss of reflexes. Therefore a central site of action can be assumed. Whereas the effects on the liver probably indicate an adaption phenomenon which occurs at doses of 56.2 mg/kg and higher.

All changes were reversible within the recovery period of 6 weeks.

A dose of 17.8 mg/kg represents a no observed effect level.

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3. INTRODUCTION

The study was conducted to investigate the toxicity profile of triallylcyanurate (TAC) after repeated oral administration in rats during 4 weeks and a subsequent 6-week recovery period.

The results of clinical investigations, clinical pathology, and pathology were used for the detection of adverse effects.

4. DOSE FINDING

Triallylcyanurate (TAC) was administered orally during a dose range finding study (No. 874078) at dose levels of 21.5 to 215 mg/kg. The test substance caused an impairment of coordination of movements and righting reflexes. Doses of 7.8, 26.2, and 178 mg/kg were selected.

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5. METHODS

5.1 Test Substance

Test Substance/
Trade Name/Identity : Triallylcyanurate (TAC)

CAS No. : 101-37-1

Batch No. : Lot.-No. 681

Physical Appearance : Clear solid

Content/Purity : 99.7 %

Solubility : In water: 0.03 g/100 ml in ethanol: well soluble

Storage : The test substance was kept in a closed container in a refrigerator.

Stability : According to information from the sponsor the test substance was stable throughout the experimental period.

Additional Information : See enclosure 11.1 and under substance No. 88605 in file in the Institute of Toxicology.

5.2 Test System

5.2.1 Animal Species : Rat

Strain : Bor: WISW (SPFCpb)

Origin/Breeder : Winkelmann Versuchstierzucht GmbH & Co. KG.,
D-4799 BorchelnAge of the Animals
at Start ofAdministration : Males 8 weeks
Females 3 weeksBody Weight of the
Animals at Start
of Administration: Males 177 - 217 g
Females 123 - 165 gJustification for
the Selection of
the Test System

: The test system was selected on the basis of international recommendations. According to these, the rat is suitable for detecting potential toxic properties of test substances in rodents.

For more than 5 years the rat strain Bor: WISW (SPFCpb) is being used in the Institute of Toxicology, so that historic control data of a number of toxicological studies are available.

5.2.2 Husbandry

- Location** : ASTA Medica AG
Institute of Toxicology
Building B, Room No. 214-3
Artur-Ladebeck-Str. 128 - 152
D-4800 Bielefeld 14
- Caging** : Macrolon cages, type II
- Number of Animals per Cage** : 1
- Bedding** : Animal bedding chips, supplied by JeluWerk, J. Ehrlar, Industriehhle, D-7092 Rosenberg, Württ.
According to information from the manufacturer contaminant analyses of the bedding chips are performed in appropriate intervals. Certificates of analyses are on file in the testing facility.
- Diet** : Standard diet ad libitum, ssniff R, "Special Diet for Rats", supplied by ssniff Spezialdiäten GmbH, D-4770 Soest (composition see enclosure 11.2)
According to information from the manufacturer contaminant analyses of the diet are performed in appropriate intervals. Certificates of analyses are on file in the testing facility.
- Water** : Water was provided ad libitum in drinking water quality from the Stadtwerke Bielefeld. An automatic watering system with drinking nipples was used.
According to information from the Stadtwerke Bielefeld the water is investigated in appropriate intervals. Certificates of analyses are on file in the testing facility.
The known contaminants present in bedding chips, diet, and water are toxicologically insignificant in the quantities detected for the experiment performed.
- Room Temperature** : 21.0 - 22.5°C
- Relative Humidity** : 40 - 65% (For short periods down to 25%; this deviation was without any influence on the results of the study.)

Room Lighting : 6 a.m. - 6 p.m. CET artificial lighting
6 p.m. - 6 a.m. CET natural light-dark-rhythm

Room Hygiene,
Cage Cleaning : The room was cleaned regularly with commercial antiseptics and the cages with the cage washing machine type HAMO-R-T-5000, supplied by Hamo AG, CH-Biel-Bienne. Instruments and apparatus were sterilized in an autoclave or a hot air sterilizer (list of antiseptics and rinsing products see enclosure 11.3).

5.2.3 Allocation and Identification of the Animals

Total Number of Animals : 70: 25 males and 35 females

Number of Animals per Dose Group : 10 male and 10 female animals each in groups 1 and 5. The last 5 animals of each sex of these 2 groups were used as recovery animals.

5 male and 5 female animals each in groups 2, 3, and 4.

Allocation of the Animals to the Groups: Allocation of animals to the groups 1 - 5 by consecutive numbers (see scheme under 5.3.2).

Randomization of the Animals : The assignment of the animals to the groups 1 - 5 was done at random using a computerized random figure generator.

Identification of the Animals and Cages

: The animals were individually identified with ear clipping codes.
The cages were labelled with:
Study number, name of the test substance, group with colour code, animal number, and sex.

5.2.4 Acclimatization Period (Pretest)

: The animals were kept 2 weeks under test conditions before administration of the test substance.
Veterinary supervision of the animals was done before start of the study.

5.3 Procedure

5.3.1 Administration of the Test Substance

- Route of Administration : Oral, by gavage (Nelator catheter, red rubber, supplied by Wilhelm BÜSCH, D-7053 Rommelshausen/Stuttgart).
- Justification for the Selection of the Route of Administration : The oral toxicity after repeated administration of the test substance was tested, as a potential oral exposure of man cannot be excluded.
- Preparation of the Solutions for Administration : Daily preparation of the solutions for administration with different substance concentrations according to the intended doses.
- Solvent/Vehicle : Peanut oil (batch 7719), supplied by H. Lamotte, D-2800 Bremen
- Stability of the Test Substance in the Solutions for Administration : Stability of the test substance in the solutions for administration were examined before start of the study (see dose finding study No. 874078).
- Determination of the Concentrations of the Test Substance in the Solutions for Administration : In test weeks 1 and 4 the concentrations of the test substance in the solutions for administration were determined by Degussa AG/ZN Wolfgang, Department IC-ATO and found to be within acceptable limits.
- Frequency Administration : Once daily a.m. (7 days per week)
- Control Animals : The animals of the control group 1 received peanut oil without test substance. The animals of the control group 2 were treated with tap water. Two control groups were used to detect possible secondary effects caused by the vehicle, peanut oil.

5.3.2 Dosing/Dose Groups

Dose Intervals : Geometric sequence

Factor : 3.16 or 3.83

Dose Groups :

Group	1	2	3	4	5
	Control (Peanut Oil) incl. 5 Rec. Animals*	Control (Water)	1.8 mg/kg	56.2 mg/kg	178/215 mg/kg incl. 5 Rec. Animals*
Administration Volume (ml/kg)	2.15	2.15	2.15	2.15	2.15
Concentration (mg/ml)	0	0	8.2	6.1	82.5/100
Animal No. m	1 - 10	11 - 15	16 - 20	21 - 25	26 - 35
f	36 - 45	46 - 50	51 - 55	56 - 60	61 - 70
* m: 6 - 10, 31 - 35; f: 41 - 45, 66 - 70					

Justification for the

Selection of Doses : The doses were selected on the basis of the results of a previous dose finding study (Study No: 874078).

- 5.3.3 Duration of the Study: 4 weeks (test weeks 1 - 4) treatment period (substance administration up to the respective day of autopsy)
6 weeks (test weeks 5 - 10) treatment-free recovery period

5. Investigations and Parameters Examined

5.1 Clinical Investigations

Mortality : Mortality was checked twice daily (a.m. and p.m.), on Saturdays, Sundays, on national and business holidays only once daily (a.m.).

Behaviour and General Condition

(Clinical Symptoms) : The animals were observed daily for the occurrence of toxicity symptoms as well as their severity and duration.

Food Consumption : Recorded on a weekly basis and converted to g/animal/day

Body Weight : Recorded once per week

Reflexes : Pain, pinna, and corneal reflexes were tested prior to first substance administration and thereafter once a week until test week 4.

Examinations of Eyes, Hearing, and Teeth : Prior to first substance administration and in test week 4. The hearing was checked with a simple noise test. The teeth were inspected. Eye examination using a focussed visual light beam (CLIPTRAX Lichtstift No. 645, supplied by Varta, D-3000 Hannover).

5.4.2 Clinical Pathology

Blood Collection : In test week 4 and additionally in test week 10 from recovery animals. Blood was collected from the retroorbital venous plexus of one eye under CO₂ anaesthesia from all animals.

Anticoagulants : EDTA, supplied by Sarstedt, D-5223 Nümbrecht-Rommelsdorf, was used as an anticoagulant for hematological parameters. The clinical chemistry was done with serum.

Urine Collection : Urine was collected in metabolism cages in test week 4 a.m. from all animals. Pretreatment before sampling with tap water (10 ml/kg b.w.).

5.4.2.1 Hematology

In test week 4 and additionally in test week 10 from recovery animals:

Erythrocytes (RBC), hematocrit (Hct), hemoglobin (Hb), leucocytes (WBC), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), mean corpuscular volume (MCV), thrombocytes (Platelets).

Performance of examinations with COULTER COUNTER S Plus II, supplied by Coulter Electronics GmbH, D-4150 Krefeld, according to the methods of the manufacturer.

Differential leucocyte count
Staining of the blood smears according to PAPPENHEIM and microscopical evaluation (% and quantity per nl).

Reticulocytes

Staining of the blood smears with Brilliantkresyl-blue.

Evaluation was not performed because there were no pathological changes in the RBC.

5.4.2.2 Clinical Chemistry : In test week 4 and additionally in test week 10 from recovery animals:

Alanine aminotransferase (ALAT), albumin, alkaline phosphatase (AKP), aspartate aminotransferase (ASAT), blood urea, calcium (Ca), chloride (Cl), cholinesterase (CHE), creatine kinase (CK), creatinine, ferrum, γ -glutamyltransferase (Gamma-GT), glucose, glutamate dehydrogenase (GLDH), inorganic phosphate (P), potassium (K), sodium (Na), total bilirubin (Tot.Bili), total cholesterol (Chol), total protein (Tot. Prot), triglycerides (Triglyc).

Centrifugal Analyser CBAS BIO,
supplied by Hoffmann La Roche & Co. AG,
CH-4002 Basle

Flame Photometer FM/5051
(sodium, potassium, calcium)
supplied by Eppendorf, Gerätebau Netheler u.
Hinz GmbH, D-2000 Hamburg 63

The methods used in clinical chemistry are described in enclosure 11.4.

5.4.2.3 Urinalysis

: In test week 4:

Bilirubin, glucose, hemoglobin/erythrocytes, ketones, leucocytes, nitrite, osmolality, pH-value, protein and urobilinogen.

Microscopical examinations of the urine sediment were done in animals whose urine state showed pathological changes in hemoglobin/erythrocytes, leucocytes, or protein.

Test strips COMBUR⁹ Test(R) RL and photometer UFOTRON RL 9(R)
supplied by Boehringer Mannheim GmbH,
D-6800 Mannheim 31

Osmometer OM 801-D
(osmolality),
supplied by Vogel GmbH & Co. KG,
D-6300 Gießen

5.4.3 Pathology

Sacrifice : At the end of the treatment or recovery period the animals were anesthetized with CO₂ and sacrificed by exsanguination.

5.4.3.1 Gross Necropsy

Autopsy : All animals were subjected to full gross necropsy which included examination of the external surface of the body, all orifices, and the cranial, thoracic, and abdominal cavities and their contents.

The following organs or tissues of a representative sample were preserved:

All gross lesions, adrenal glands (r/l), bone marrow (as present in sternum), bone marrow smear (from femur), brain (cerebrum, cerebellum, brain stem), caecum, colon, duodenum, heart, ileum, jejunum, kidneys (r/l), liver, lungs (including main bronchi), ovaries (r/l), rectum, spleen, stomach, testes (r/l).

5.4.3.2 Organ Weights

Organs : Adrenals (r/l), brain, heart, kidneys (r/l), liver, ovaries (r/l), spleen, testes (r/l).

The organ weights (wet weights) were expressed as absolute values and relative to body weight* (organ/body weight ratio).

* Body weight after exsanguination

5.4.3.3 Histopathology

Fixation and Staining : All organs and tissues except for bone marrow smears were fixed in a 4 % neutral buffered formaldehyde solution (10 % formalin) according to LFLLE. The bone marrow smears were air dried.

All fixed organs and tissues of all animals were trimmed, embedded in paraffin wax, sectioned at approx. 4 μ m and stained with Hematoxylin and Eosin (H&E).

Bone marrow smears were stained according to PAPPENHEIM.

Microscopical
Examinations

: All tissues from control and high dose groups, except for bone marrow smears, were examined. Because the lesions were within the normal range no other tissues from the low and mid dose groups were examined, except for macroscopical changes.

5.5 Statistics

: Mean values of parameters and - where indicated - standard deviations were calculated separately for each group and sex.

For statistical evaluation of food consumption, body weights, and organ weights the DUNNETT-Test (4) was used. For values of hematological and clinical chemistry examinations the DUNNETT-Test was used in case of normal distribution, otherwise the STEEL-Test (5) was employed.

Significant differences between mean values of control group 1 and dose groups 3, 4, and 5 were marked with * or ** (significance level of $p < 0.05$ or $p < 0.01$ according to DUNNETT) or + (significance level $p < 0.05$ according to STEEL), respectively.

5.6 Data Processing
Systems

: Numerical values were recorded and processed using the RCC Software Package, supplied by Research and Consulting Company AG, CH-4452 Ittingen, Basle-Switzerland, on a VAX 8200 Computer, supplied by Digital Equipment Corp., D-8000 München 81. Statistical calculations were done by means of the RCC Program.

Macroscopic and microscopic findings were recorded and tabulated with a MAI Computer, supplied by MAI-Deutschland GmbH, D-6000 Frankfurt/M. 71, using the EPS-Pathology Data Computer System Software, supplied by Experimental Pathology Services, Basle-Switzerland.

7 Protocol Adherence

: The study was conducted in accordance with the original protocol and the 4 amendments with the following exceptions:

The name of ASTA Pharma AG was replaced by ASTA Medica AG and the name and address of Toxikologie Degussa-Asta changed because of the removal of the institute.

The duration of archivation for tissues, paraffin blocks, slides, blood smears and for the sample of the test substance has changed according to the modification of the German Chemicals Act of June 5, 1991 (3).

Following the dose elevation in the high dose group, the factor increased to 3.83.

At microscopical examination all tissues from control and high dose groups, except for bone marrow smears, were examined. Because the lesions were within the normal range, no other tissues from the low and mid dose groups were examined, except for macroscopical changes.

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6. RESULTS

6.1 Clinical Investigations

6.1.1 Clinical Symptoms

Behavioural changes were detected only in animals of groups 4 and 5 (56.2 and 178/215 mg/kg). While in group 4 coordination disturbances were present only temporarily and in about half of the animals, this symptom was recorded in all rats of the high dose group almost every day of the treatment period. Additionally all rats of this group exhibited loss of muscle tone and righting reflexes after up to about 50% of the administrations. Salivation also was observed in all animals of group 5 on 2 to 21 days. In 6 of these animals chronic diarrhoea occasionally was recorded. Only individual rats at rare occasions displayed slight clonic convulsions or piloerection.

None of the rats died in the course of the study.

6.1.2 Food Consumption

The uptake of food was affected only in animals of groups 4 (56.2 mg/kg) and 5 (178/215 mg/kg). In group 4 males only a minimal decrease was detected during the first 2 weeks of treatment. Females of this group were not affected.

In group 5 males as well as females consumption of food was reduced by about 20% as compared to the vehicle control group (group 1). This effect, however, was present only for the measurement during the first week. Afterwards the animals had adapted and differences were no longer detected until the dose was increased at the beginning of the fourth week. At that time male and female rats showed another slight decrease in food consumption. After the end of the treatment period both males and females of the high dose recovery group as well as the recovery control group increased their food uptake considerably to values previously found in the water treated controls or even higher. This effect is considered as a consequence of the cessation of energy supply by the vehicle (peanut oil).

Figures 1 and 2 and tables 1 and 2 give the mean values \pm S.D. of the food consumption.

The individual animal data are given under 10.1, page 119.

6.1.3 Body Weights

The development of the body weights was affected only in males of the high dose group. In these animals the body weight gain fell slightly behind the control animals during the treatment period.

Figures 3 and 4 and tables 3 and 4 give the mean values \pm S.D. of the body weights.

The individual animal data are given under 10.2, page 126.

6.1.4 Examinations of Reflexes, Eyes, Hearing, and Teeth

At examinations of reflexes, eyes, hearing, and teeth there were no substance related effects recorded.

6.2 Clinical Pathology

6.2.1 Hematology

Hematology was done in weeks 4 and 10. At week 4 the only possibly test substance related alteration in a hematology parameter was the marginal reduction of the number of platelets in males of the high dose group.

Tables 5 and 6 give the mean values \pm S.D. of each group and sex for the hematological parameters examined.

The individual animal data are given under 10.3, page 133.

6.2.2 Clinical Chemistry

Clinical chemistry parameters were examined in weeks 4 and 10. Statistically significant deviations from the control values were found predominantly in groups 4 and 5. At the end of the treatment period (week 4) a slight increase in the activity of alanine aminotransferase was detected in males as well as in females of group 4. The same was true for the concentration of inorganic phosphate while the amount of albumin was slightly reduced. The concentration of serum iron and sodium were slightly increased in males. All other statistically significant alterations, namely increased total bilirubin in water control females (group 2), increased glucose concentration in males of group 4, increase of total protein and cholesterol in group 4 females, and increased activity of alkaline phosphatase in group 5 females are considered as incidental findings, because the changes are not dose dependent and in the physiological range for this rat strain.

The second examination at the end of the recovery period (week 10) significant alterations were present only in males. The activities of alanine aminotransferase, glutamate dehydrogenase, and cholinesterase as well as the concentration of triglycerides were slightly decreased. Because the changes were only mild, no connection is to be assumed with this toxicity study.

Tables 7 and 8 give the mean values \pm S.D. of each group and sex for the clinical chemistry parameters examined.

The individual animal data are given under 10.4, page 150.

6.2.3 Urinalysis

Examination of the urine in week 4 did not reveal any significant differences between control and treated animals.

Tables 9 and 10 give the mean values \pm S.D. of each group and sex.

The individual animal data are given under 10.5, page 163.

6.3 Pathology

6.3.1 Gross Necropsy

On macroscopic inspection no test substance related alterations were detected.

The individual animal data are given under 10.8, page 194.

6.3.2 Organ Weights

After the 4-week treatment period high dose male rats showed minimally reduced absolute weights of the spleens only, while in animals of the water control group (group 2) the weights of brains, kidneys, and spleens were significantly increased. In female animals the weights of livers were slightly increased in groups 4 and 5 and the adrenal weights were increased in group 5 females.

The organ/body weight ratios showed significant differences mainly in group 5 males and females. All values were increased by more than 10% for brains, livers, kidneys, and adrenals in males and for livers, kidneys, and adrenals in females. The liver/body weight ratio was increased slightly in females of group 4 also.

With exception for the relatively increased liver weights (see histopathological findings) all other increased relative organ weights in high dose males can be attributed to the significantly lower body weight of this group.

At the end of the 6-week recovery period none of the findings of week 4 could be detected.

Tables 11-14 give the mean values \pm S.D. of each group and sex for the organ weights and the organ/body weight ratios respectively.

The individual animal data are given under 10.6 and 10.7, pages 180 and 187.

6.3.3 Histopathology

Microscopical examination revealed no treatment related changes in any of the organs examined, except for the liver. High dose animals of both sexes had a marginally increased incidence of microgranulomas in the liver (microgranulomas are small focal aggregates of predominantly macrophages and mononuclear cells occasionally containing individual necrotic hepatocytes. It is generally considered a reaction to such a cell turnover).

The incidence and severity of this lesion was however within normal range for this species. It may however explain the slightly increased values of alanine aminotransferase and glutamate dehydrogenase observed in the high dose group animals.

Summary tables of the microscopic findings are presented under 15, page 108.

The individual animal data are given under 10.8, page 194.

7. REFERENCES

1. OECD (1981). "Repeated Dose Oral Toxicity-Rodent 28-Day or 14-Day Study",
In OECD Guidelines for Testing of Chemicals
Section 4: Health effects, Method No. 407,
Organisation for Economic Cooperation and Development (OECD),
Paris, ISBN-92-64-12221-4.
2. EEC (1984). Commission Directive of 25 April 1984
adapting to technical progress for the sixth time
Council Directive 67/548/EEC on the approximation
of laws, regulations and administrative provisions
relating to the classification, packing and labelling
of dangerous substances (84/449/EEC),
Annex V, B.7. Subacute Toxicity - Oral.
3. Law on protection from dangerous substances (German Chemicals
Act), August 1, 1990, Amendment 1: Principles of Good Laboratory
Practice (GLP) amended at: "Dritte Verordnung zur Änderung der
Gefahrstoffverordnung" dated June 5, 1991, published in
Bundesgesetzblatt Part I, Volume 1990, No. 13, March 27, 1990
and Volume 1991, No. 35, June 11, 1991.
4. DUNNETT, C.W.,
(1) A multiple comparison procedure for comparing several
treatments with a control.
Journal of the American Statistical Association 50, 1096-1121
(1955).
(2) New Tables for multiple comparison with a control,
Biometrics 20, 482-492 (1964).
5. STEEL, R.G.D.,
A multiple comparison rank sum test: treatments versus control.
Biometrics 15, 560-582 (1959).



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TNO-report

Report no.: R 89/287

BIODEGRADABILITY OF TRIALLYLCYANURATE
ACCORDING TO OECD 301B
(MODIFIED STURM-TEST)

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Dr J.W. Vonk

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GLP COMPLIANCE STATEMENT

'I, the undersigned hereby declare that the work to which this report refers was performed under my supervision according to the procedures herein described. To the best of my knowledge this report provides an accurate record of the results obtained. The study was carried out in compliance with the OECD code of Good Laboratory Practice except in the case of characterization and verification of the test substance identity and properties, maintenance of these records is the responsibility of the sponsor.'



Dr J.W. Vonk
Study director

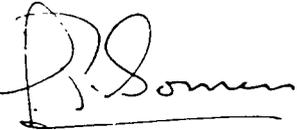
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TNO Quality Assurance Unit (Toxicology)
P. O. Box 360
3700 AJ ZEIST

Report no.: R89/287
Study no. : MTB-89-0034-01

QUALITY ASSURANCE AUDIT STATEMENT

STUDY TITLE: Biodegradability of triallylcyanurate according to OECD 301B
(modified Sturm-test)

REPORT DATE: 1989.09.01

The report of this study was audited by the TNO Quality Assurance Unit
(Toxicology), P. O. Box 360, 3700 AJ Zeist.

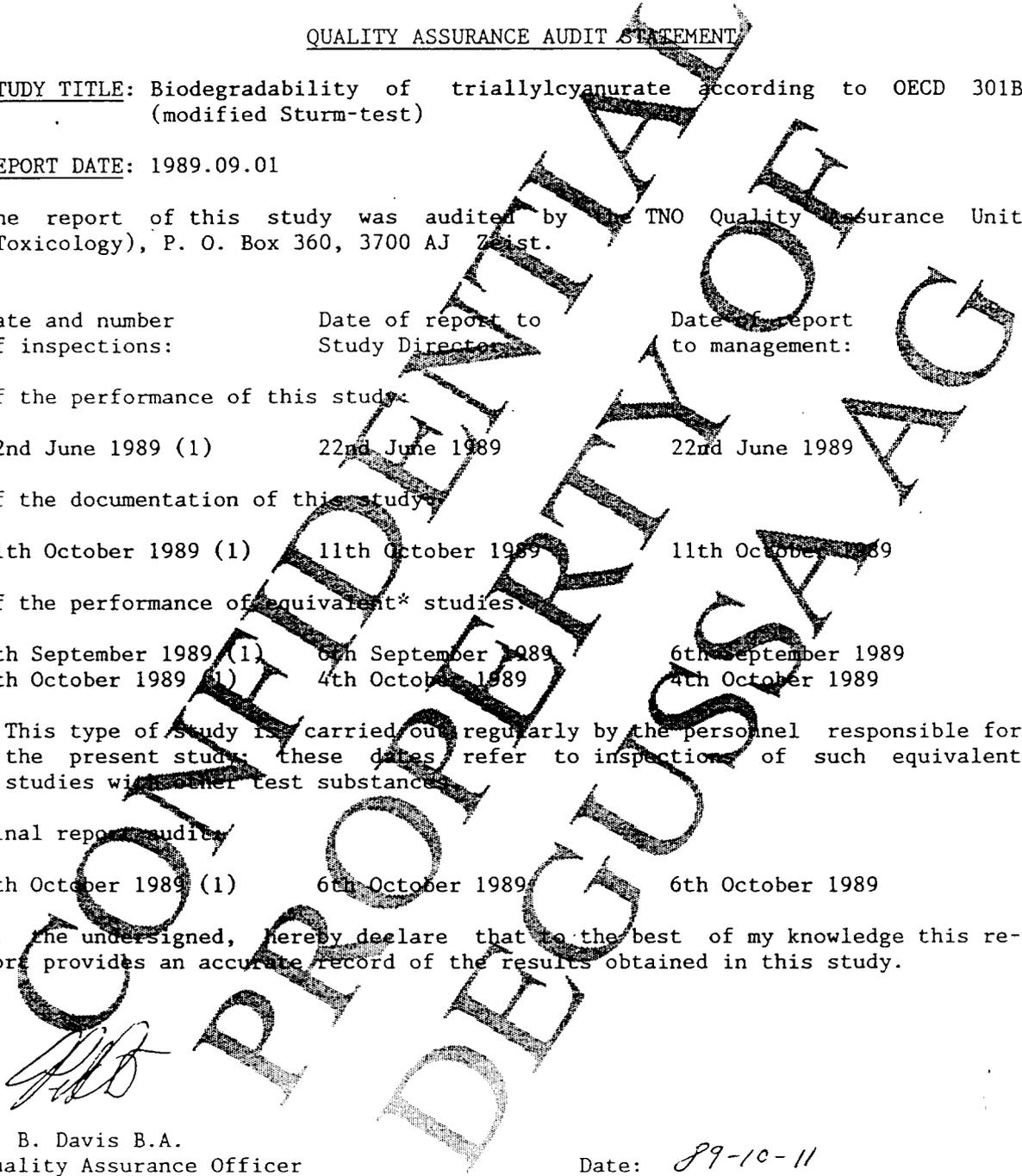
Date and number of inspections:	Date of report to Study Director:	Date of report to management:
of the performance of this study:		
22nd June 1989 (1)	22nd June 1989	22nd June 1989
of the documentation of this study:		
11th October 1989 (1)	11th October 1989	11th October 1989
of the performance of equivalent* studies:		
6th September 1989 (1)	6th September 1989	6th September 1989
4th October 1989 (1)	4th October 1989	4th October 1989
Final report audited:		
6th October 1989 (1)	6th October 1989	6th October 1989

* This type of study is carried out regularly by the personnel responsible for
the present study; these dates refer to inspections of such equivalent
studies with other test substance

I, the undersigned, hereby declare that to the best of my knowledge this re-
port provides an accurate record of the results obtained in this study.


P. B. Davis B.A.
Quality Assurance Officer

Date: 89-10-11



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The last page of this report is 18.

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SUMMARY AND CONCLUSION

The biodegradability of triallylcyanurate was determined by the method laid down in the OECD Guideline 301 B; Ready Biodegradability: Modified Sturm Test (1).

A control test showed that the activity of the inoculum was sufficient and that no significant effects of the test substance on this activity was found.

The CO₂-production resulting from biodegradation of triallylcyanurate was small and this substance is therefore regarded as not being readily biodegradable.

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1. INTRODUCTION

The biodegradability of triallylcyanurate was determined at sponsor's request.

The determination was conducted in accordance with the OECD Guideline 301 B: Ready Biodegradability: Modified Sturm Test (1).

This report presents a description of the methods used and the results obtained.

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2. MATERIALS AND METHODS

2.1 TEST SUBSTANCE

The substance investigated was triallylcyanurat, batch no. 9503, a clear crystalline solid which was supplied by sponsor in a plastic bottle labelled: 100 g Triallylcyanurat (TAC); CAS-RN-101-37-1 with 100 g sample was received on 1989-06-13. The solubility in water was 0.03 g/100 g according to sponsor.

The sample was stored at room temperature in the dark. The TOC of the sample was found to be 526 mg.g⁻¹.

The composition and properties of triallylcyanurat were specified by sponsor and are recorded in Annex C.

2.2 ACTIVATED SLUDGE

A sample of activated sludge was taken from an oxidation ditch situated on the premises of TNO, Bilt, The Netherlands. The oxidation ditch is used to treat domestic sewage.

The original sludge (2.7 g of solid substance.l⁻¹) was allowed to settle for 5 minutes and 10 ml of the supernatant was used to inoculate one litre of medium (2).

The inoculated medium was aerated with CO₂-free air for 24 hours before use.

2.3 TEST MEDIUM

A medium with a buffer capacity higher than the standard was used (2); its composition is given in Annex A.

2.4 TEST DATES

The protocol was signed by sponsor on 1989-06-05.

The biodegradability test was carried out in the period 1989-07-03 until 1989-08-01.

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2.5 TEST METHOD

The biodegradation test was conducted essentially as detailed in OECD 301 B (1). Two-litre glass bottles (Schott Duran) closed with a plastic screw cap were used for the test. A 30 ml glass vial containing the CO₂-absorbing fluid was suspended from the screw cap in each bottle.

The bottles were filled with one litre of inoculated medium, and 5 ml of 0.40 M NaOH was placed in the vial.

The test substance was slightly warmed in order to melt it. Two concentrations of test substance (12 and 24 mg.l⁻¹ respectively) were prepared in triplicate by weighing the correct amount of melted test substance on a cover-glass and adding this cover-glass with test substance to each bottle containing inoculated medium. A control series without test substance and with cover-glass was included to allow correction for the background CO₂-production by the inoculum.

In order to determine the inoculum activity, 100 mg.l⁻¹ of sodium acetate as a carbon source was added to two additional bottles containing inoculated medium only.

In order to detect possible toxic effects of the test substance, sodium acetate (100 mg.l⁻¹) was also added to further duplicate bottles containing 12 or 24 mg.l⁻¹ of allyl cyanurate.

The bottles were incubated on a Braun Pilot Shaker orbital shaking machine in a room kept at 20°C for 28 days. The CO₂-absorption vials were replaced by fresh ones after 7, 14, 21 and 28 days. The amount of CO₂ absorbed was determined according to OECD 301 B (1) by titration with 0.1 M HCl. The pH-value of the medium was also determined.

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3. RESULTS AND DISCUSSION

3.1 INOCULUM ACTIVITY AND TOXICITY

The biodegradation of acetate in the control series in the absence of test substance was used as a measure for the microbial activity of the inoculum. A reduction of the biodegradation of acetate in the presence of the test substance would indicate a toxic effect on the inoculum activity.

The results of these tests are given in Table 1 and, more fully, in Annex B. They show that, as expected, acetate was significantly degraded (73%) within two weeks. No effect of the added triallylcyanurate on the inoculum activity could be found.

Table 1 Results of the control tests, for inoculum activity and the effect of triallylcyanurate on the biodegradation of sodium acetate by the inoculum. The results are expressed as mg CO₂ per litre (cumulative) after 7, 14, 21 and 28 days, and as percentage biodegradation of acetate. The theoretical CO₂-production of acetate is 1.07 mg CO₂.mg⁻¹, and 100 mg of acetate was added per litre. The results are the mean values of duplicate bottles.

days	triallylcyanurate (mg.l ⁻¹)					
	0		12		24	
	mg CO ₂ .l ⁻¹	% biodegr.	mg CO ₂ .l ⁻¹	% biodegr.	mg CO ₂ .l ⁻¹	% biodegr.
7	45	42	46	43	45	42
14	78	72	80	74	81	75
21	90	84	91	85	91	85
28	95	89	96	90	96	90

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3.2 BIODEGRADABILITY

The solubility of the test substance was 0.03 g/100 ml in water.

The pH of the medium remained constant at 7.8 ± 0.1 during the test.

The results of the determination of the biodegradability of triallylcyanurate are given in Table 2, and, more fully, in Annex B.

Table 2 Results of the biodegradation test with triallylcyanurate. The results are expressed as mg CO₂ per litre (cumulative) after 7, 14, 21 and 28 days, and as percentage biodegradation of triallylcyanurate and are mean values of triplicate bottles. The theoretical CO₂-production of triallylcyanurate is 2.12 mg CO₂.mg⁻¹.

days	12		24	
	mg CO ₂ .l ⁻¹	% biodegr.	mg CO ₂ .l ⁻¹	% biodegr.
7	0.1	0	1.1	2
14	0.4	2	1.4	3
21	1.5	6	2.5	6
28	2.2	9	3.1	7

Only slight CO₂ production was found within 28 days.

It was concluded that triallylcyanurate is not readily biodegradable in this test.

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4. REFERENCES

1. OECD 301 B: Ready Biodegradability: Modified Sturm Test.
OECD Guideline for Testing of Chemicals,
Paris, 1981.
2. NPR 6513: Water - Screening test for ready biodegradability
of organic compounds.
Nederlands Normalisatie-instituut, Delft, 1982.

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5. RETENTION OF RECORDS AND SAMPLES

All records concerning the observations, counts, analyses and all other information relevant to the quality and integrity of the study have been filed in the archives of the Department of Biology, TNO Division of Technology for Society in Delft, under the study reference MTR-89-0034-01.

These records will be retained for a period of at least ten years after the cover date of this report.

A sample of the test substance has been deposited in the sample archives of the Department of Biology, under the sample reference MTR-89-0034-A; this sample will also be stored for ten years and then disposed of.

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6. DEVIATIONS FROM THE PROTOCOL

The test was carried out by Ms Y.A. Matla.

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ANNEX ATest medium

.. Add 10 ml of solution a and 1 ml of solutions b to f to water and make up to 1000 ml

a. Solution a contains per 1000 ml water:

KH_2PO_4 8.5 g

K_2HPO_4 21.75 g

$\text{Na}_2\text{HPC}_4 \cdot 2\text{H}_2\text{O}$ 33.4 g

NH_4Cl 1.5 g

b. Solution b contains 22.5 g of $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ per 1,000 ml water

c. Solution c contains 27.5 g of $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ per 1,000 ml water

d. Solution d contains 0.25 g of $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ per 1,000 ml water
Prepare this solution freshly before use.

e. Trace element solution.

This solution contains per 1000 ml water:

$\text{MnSO}_4 \cdot 4\text{H}_2\text{O}$ 39.9 mg (30.23 mg $\text{MnSO}_4 \cdot \text{H}_2\text{O}$)

H_3BO_3 57.2 mg

$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ 42.8 mg

$(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}$ 34.7 mg (36.85 mg $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$)

EDFA 100 mg

If not used immediately, the trace element stock solution must be sterilized by filtration.

f. 15 mg yeast extract dissolved in 100 ml water.

Prepare this solution freshly immediately before use.

MTJT17/A

ANNEX BResults of the biodegradability test

Concentration trialllycyanurat mg.l ⁻¹	Na-acetate mg.l ⁻¹	mg of CO ₂ .l ⁻¹ collected in the periods ²⁾				
		0-7 days	7-14 days	14-21 days	21-28 days	
12	A ¹⁾	-	0.8	0.3	1.0	0.6
	B	-	-0.5	0.4	0	1.0
	C	-	0	0.3	1.3	0.2
24	A	-	0.4	0.2	1.4	0.8
	B	-	1.8	0.5	0.6	0.6
	C	-	1.0	0.4	1.2	0.6
0	A	100	45	33	12	5
	B	100	45	32	12	5
12	A	100	45	33	12	5
	B	100	46	34	12	5
24	A	100	44	35	11	5
	B	100	46	36	11	5

¹⁾ A, B (and C) are the replicate test flasks.

²⁾ The data have been corrected for the CO₂ production of the inoculum itself; this sometimes results in negative values.

