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August 10, 2009

MIR#320963

TSCA Confidential Business Information Center (7407M)  
EPA East-Room 6428 Attn: Section 8(e)  
U.S. Environmental Protection Agency  
1200 Pennsylvania Avenue, NW  
Washington, DC 20460-0001



8EHQ-0809-17609A

Dear Sir/Madam:

The purpose of this letter is to inform you under Section 8(e) of TSCA of the results obtained from the recent three aquatic toxicity studies on EPOMIN SP-200, the chemical being produced by my client, Nippon Shokubai Co., Ltd. The reports are entitled "Acute Toxicity Test of EPOMIN SP-200 with Medaka (*Oryzias latipes*)", "Acute Immobilisation Test of EPOMIN SP-200 with *Daphnia magna*", and "Growth Inhibition Test of EPOMIN SP-200 with *Pseudokirchneriella subcapitata*". EPOMIN SP-200, a commercial chemical product has been manufactured in Japan by Nippon Shokubai Co., Ltd., and imported to the US by my client, NA Industries Inc. , P.O. Box 5407 Chattanooga, TN 37406. EPOMIN SP-200's CAS name and number are Aziridine homopolymer and 9002-98-6, respectively.

Enclosed please find the copy of the reports and its TSCA Health & Safety Study Cover Sheet.

If you have any questions or comments on this submission, please feel free to call me at (703)920-5440 or email me at [hiyama@ix.netcom.com](mailto:hiyama@ix.netcom.com).

Sincerely yours,

Hiroshi Uyama

**Contains No CBI**



DCN: (88090000333)

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**TSCA HEALTH & SAFETY STUDY COVER SHEET** - revised 6/25/96

TSCA CBI STATUS:

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Clearly mark the confidential information with bracketing and check the box in the appropriate section ( *Contains CBI*)  
 Submit a sanitized cover sheet with CBI deleted. Mark the sanitized copy, "Public Display Copy" in the heading.

<b>1.0 SUBMISSION TYPE</b> <input type="checkbox"/> <i>Contains CBI</i> <input type="checkbox"/> 8(d) <input type="checkbox"/> 8(e) <input type="checkbox"/> FYI <input type="checkbox"/> 4 <input type="checkbox"/> OTHER: Specify _____ <input checked="" type="checkbox"/> Initial Submission <input type="checkbox"/> Follow-up Submission <input checked="" type="checkbox"/> Final Report Submission Previous EPA Submission Number or Title if update or follow-up: _____ Docket Number, if any: # _____ ITC Submission <input type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> continuation sheet attached				
<b>2.1 SUMMARY/ABSTRACT ATTACHED</b> (may be required for 8(e); optional for 8(d), 8(d) & FYI) <input type="checkbox"/> YES <input checked="" type="checkbox"/> NO		<b>2.2 SUBMITTER TRACKING NUMBER OR INTERNAL ID</b>	<b>2.3 FOR EPA USE ONLY</b>	<b>2.4 Study</b> _____ <b>of</b> _____
<b>3.0 CHEMICAL/TEST SUBSTANCE IDENTITY</b> <input type="checkbox"/> <i>Contains CBI</i> <p align="center"><i>Reported Chemical Name (specify nomenclature if other than CAS name):</i></p> CAS# <u>4002-98-6</u> <u>Aziridine homopolymer</u> Purity _____ % <input checked="" type="checkbox"/> Single Ingredient    Trade Name: <u>EPOMIN SP-200</u> Common Name: <u>Polyethyleneimine</u> <input type="checkbox"/> Commercial/Tech Grade <u>CAS Number</u> <u>NAME</u> <u>% WEIGHT</u> <input type="checkbox"/> Mixture				
Other chemical(s) present in tested mixture <input type="checkbox"/> continuation sheet attached				
<b>4.0 REPORT/STUDY TITLE</b> <input type="checkbox"/> <i>Contains CBI</i> <p align="center"><u>Growth Inhibition Test of EPOMIN SP-200 with Pseudomonas subcapitata</u></p> <input type="checkbox"/> continuation sheet attached				
<b>5.1 STUDY/TSCATS INDEXING TERMS</b> [CHECK ONE] HEALTH EFFECTS (HE): _____ ENVIRONMENTAL EFFECTS (EE): <input checked="" type="checkbox"/> ENVIRONMENTAL FATE (EF): _____				
<b>5.2 STUDY/TSCATS INDEXING TERMS</b> (see instructions for 4 digit codes) STUDY TYPE: <u>ATOX</u> SUBJECT ORGANISM (HE, EE only): <u>ALGA</u> ROUTE OF EXPOSURE (HE only): <u>0</u> VEHICLE OF EXPOSURE (HE only): _____ Other: _____    Other: _____    Other: _____				
<b>6.0 REPORT/STUDY INFORMATION</b> <input type="checkbox"/> <i>Contains CBI</i> <input type="checkbox"/> Study is GLP Laboratory: <u>Mitsubishi Chemical Medicine Corporation</u> Report/Study Date: <u>6/30/09</u> Source of Data/Sponsor (if different than submitter): _____ Number of pages: <u>10</u> <input type="checkbox"/> continuation sheet attached				
<b>7.0 SUBMITTER INFORMATION</b> <input type="checkbox"/> <i>Contains CBI</i> Submitter: <u>Takashi Tomita</u> Title: <u>Manager</u> Phone: <u>(423) 624-6496</u> Company Name: <u>NA Industries Inc</u> Company Address: <u>P.O. Box 5407 Chattanooga TN 37406</u> Submitter Address (if different): _____ Technical Contact: <u>Hiroshi Uyama, Japan Technical Information Center</u> Phone: <u>(603) 920-5440</u> <input type="checkbox"/> continuation sheet attached    e-mail address: _____				
<b>8.0 ADDITIONAL COMMENTS</b> <input type="checkbox"/> <i>Contains CBI</i> <p align="center"><u>[Signature]</u>  <u>[Signature]</u>    Date: <u>8/10/09</u></p> <input type="checkbox"/> continuation sheet attached				

## FINAL REPORT

### Growth Inhibition Test of EPOMIN SP-200 with

#### *Pseudokirchneriella subcapitata*

(Report No. : A090106)

June 30, 2009

Yokohama Research Center

Toxicological Science Division

Medi-chem Business Segment

Mitsubishi Chemical Medicine Corporation.

1000 Kamoshida-cho, Aoba-ku, Yokohama,

Kanagawa, Japan

Study Director:

*Yoshiyuki NIIKURA*  
Yoshiyuki NIIKURA

#### Sponsor

NIPPON SHOKUBAI CO., LTD.

#### Purpose

In order to clarify the effects of the test substance, green algae (*Pseudokirchneriella subcapitata*) was exposed to the test substance for 72-hours.

The growth inhibition test was conducted under the two different concentration ranges.

#### Test Guidelines

OECD Guidelines for the Testing of Chemicals 201 (2006) "Freshwater Alga and Cyanobacteria, Growth Inhibition Test"

#### Testing Date

Exposure period :	Test 1	May 11-14, 2009
	Test 2	June 2-5, 2009

#### Test Methods

- |                             |   |
|-----------------------------|---|
| 1) Test substance:          | EPOMIN SP-200 (Lot No.: 9C06K1)   |
| 2) Exposure procedure:      | static, shake culture (100 rpm)   |
| 3) Test species:            | green algae ( <i>Pseudokirchneriella subcapitata</i> )                            |
| 4) Duration:                | 72 hours  |
| 5) Test concentrations:     | Test 1    1.0, 10, 100 mg/L and control<br>Test 2    0.010, 0.10 mg/L and control |
| 6) Dilution water:          | medium (recommended by the Test Guideline)  |
| 7) Volume of test solution: | 100 mL/vessel   |
| 8) Replicates:              | 6 vessels/control groups, 3 vessels/concentration groups                          |
| 9) Initial biomass:         | $5 \times 10^3$ cells/mL  |

- 10) Temperature: 22°C, variation range over the exposure period is within  $\pm 2^\circ\text{C}$   
 11) pH: not adjusted  
 12) Light: 65 to 75  $\mu\text{E}/\text{m}^2/\text{s}$ , continuously illuminated with white fluorescent lamp (near liquid surface)  
 13) Biomass measurement: electric particle counter

### **Test Procedure**

The stock solution of the test substance was diluted with dilution water to prepare the test solution for each concentration. The test solution for control group is only dilution water.

A certain quantity of the pre-cultured algae was added to each of the test solution in the flask. Consequently, the biomass in the solution, cell concentration as a substitute parameter, was  $5 \times 10^3$  cells/mL.

The biomass in each flask was determined every 24 hours. The biomass was determined with the electric particle counter. At 72 hours, the cell shapes of algae were also observed through the microscope.

The pH of the test solution was measured at the beginning of exposure and 72 hours after the beginning of exposure. During the exposure period, temperatures, light intensities, and revolutions in the culturing apparatus were measured once a day.

The mean value of the biomass for the concentration and control group were plotted against time to produce growth curves. It was confirmed that the control group was in logarithmic growth during the exposure period. The growth rate ( $\mu$ ) from the beginning of exposure to 72 hours after the beginning of exposure were calculated. The inhibition percentage ( $I\mu = (\mu_{\text{c}} - \mu) / \mu_{\text{c}} \times 100$ ) of the concentration group was calculated by subtraction of the average rate for the concentration group from the mean value for rate in the control group.

### **Results and Discussion**

Biomass of algae during the 72 hours exposure are shown in Table 1. Algal Growth Curve are shown in Figure 1. Growth Inhibitions are shown in Table 2.

Growth Inhibition Concentration and Cell shape of algae are shown as follows.

The pH values of test solutions and cultures is shown in Table 3. The conditions (temperatures, light intensities, and revolutions) in the culturing apparatus during 72-hours exposure period are shown in Table 4. The conditions were in normal ranges.

#### 1) Growth Inhibitions $I\mu(0-72\text{h})$

0.010 mg/L	$I\mu(0-72\text{h})$	:	8.3%
0.10 mg/L	$I\mu(0-72\text{h})$	:	78.1%
1.0 mg/L	$I\mu(0-72\text{h})$	:	>100%*
10 mg/L	$I\mu(0-72\text{h})$	:	>100%*
100 mg/L	$I\mu(0-72\text{h})$	:	>100%*

\* : The growth inhibition rate exceeded on 100 percent, because the biomass was below the quantitative limits of the electric particle counter.

## 2) Cell shape of algae

At the end of exposure, the microscopic observation in concentration groups(0.010, 0.10mg/L) showed neither unusual cell shape of algae (contraction, expansion, damaged cell etc.) nor agglutination was observed, and the algae looked normal compared with the control. In concentration groups(1.0, 10, 100mg/L), it couldn't observe because the biomass in each flask was small quantity.

Table 1 Biomass of *Pseudokirchneriella subcapitata* during the 72-Hour Exposure

## (1) Test 1

Test Group	Nominal Concentration (mg/L)	Vessel No.	Biomass (cells/mL)			
			0 Hour*	24 Hours	48 Hours	72 Hours
Control	--	1	5000	27600	217000	1580000
		2	5000	30100	229000	1660000
		3	5000	30600	223000	1540000
		4	5000	27900	215000	1660000
		5	5000	27900	203000	1490000
		6	5000	31000	241000	1840000
		Average	5000	29200	221000	1630000
SD	0	1550	13000	123000		
Conc.1	1.0	1	5000	<1000**	<1000**	<1000**
		2	5000	<1000**	<1000**	<1000**
		3	5000	<1000**	<1000**	<1000**
		Average	5000	<1000	<1000	<1000
		SD	0	0	0	0
Conc.2	10	1	5000	<1000**	<1000**	<1000**
		2	5000	<1000**	<1000**	<1000**
		3	5000	<1000**	<1000**	<1000**
		Average	5000	<1000	<1000	<1000
		SD	0	0	0	0
Conc.3	100	1	5000	<1000**	<1000**	<1000**
		2	5000	<1000**	<1000**	<1000**
		3	5000	<1000**	<1000**	<1000**
		Average	5000	<1000	<1000	<1000
		SD	0	0	0	0

SD : Standard deviation

\* : Nominal initial biomass

\*\* : Biomass was decided on below 1000 cells/mL, because the measurement was below the quantitative limits of the electric particle counter.

Table 1 Continued

## (2) Test 2

Test Group	Nominal Concentration (mg/L)	Vessel No.	Biomass (cells/mL)			
			0 Hour*	24 Hours	48 Hours	72 Hours
Control	--	1	5000	30700	362000	2380000
		2	5000	29800	308000	2050000
		3	5000	27000	232000	1850000
		4	5000	34400	366000	2680000
		5	5000	29900	300000	2330000
		6	5000	29900	247000	1750000
		Average	5000	30300	303000	2170000
		SD	0	2380	56000	353000
Conc.1	0.010	1	5000	12100	168000	1310000
		2	5000	22100	178000	940000
		3	5000	19900	236000	1780000
		Average	5000	18000	194000	1340000
		SD	0	5250	36700	421000
Conc.2	0.10	1	5000	<1000**	<1000**	5290
		2	5000	<1000**	6680	54400
		3	5000	<1000**	1370	23000
		Average	5000	<1000**	<3020**	27600
		SD	0	0	3180	24900

SD : Standard deviation

\* : Nominal initial biomass

\*\* : Biomass was decided on below 1000 cells/mL, because the measurement was below the quantitative limits of the electric particle counter.

Table 2 Growth Inhibitions (%) of *Pseudokirchneriella subcapitata*

## (1) Test 1

Test Group	Nominal Concentration (mg/L)	Vessel No.	Growth Rate	
			Rate $\mu$ (0-72h)	Inhibition(%) <sup>*1</sup> $I_{\mu}$ (0-72h)
Control	--	1	0.0799	
		2	0.0806	
		3	0.0796	
		4	0.0806	
		5	0.0791	
		6	0.0821	
		Average SD	0.0803 0.0010	
Conc.1	1.0	1	<-0.0224 <sup>*2</sup>	
		2	<-0.0224 <sup>*2</sup>	
		3	<-0.0224 <sup>*2</sup>	
		Average SD	<-0.0224 <sup>*2</sup> 0.0000	
Conc.2	10	1	<-0.0224 <sup>*2</sup>	
		2	<-0.0224 <sup>*2</sup>	
		3	<-0.0224 <sup>*2</sup>	
		Average SD	<-0.0224 <sup>*2</sup> 0.0000	
Conc.3	100	1	<-0.0224 <sup>*2</sup>	
		2	<-0.0224 <sup>*2</sup>	
		3	<-0.0224 <sup>*2</sup>	
		Average SD	<-0.0224 <sup>*2</sup> 0.0000	

\*1 : Values are the growth inhibition (%) relative to the control.

\*2 : The growth rates were calculated using the quantitative limits

SD : Standard deviation

Table 2 Continued

(2) Test 2

Test Group	Nominal Concentration (mg/L)	Vessel No.	Growth Rate	
			Rate $\mu$ (0-72h)	Inhibition(%) <sup>*1</sup> $I_{\mu}$ (0-72h)
Control	--	1	0.0856	
		2	0.0836	
		3	0.0821	
		4	0.0873	
		5	0.0853	
		6	0.0814	
		Average SD	0.0842 0.0023	-
Conc.1	0.010	1	0.0773	
		2	0.0727	
		3	0.0816	
		Average SD	0.0772 0.0045	8.3
Conc.2	0.10	1	0.0008	
		2	0.0332	
		3	0.0212	
		Average SD	0.0184 0.0164	78.1

\*1 : Values are the growth inhibition (%) relative to the control.

SD : Standard deviation

Table 3 pH Values of Test Cultures

## (1) Test 1

Test Group	Nominal Concentration (mg/L)	pH		
		0 Hour	72 Hours (Vessel No.)	
Control	--	7.9	7.8	(1)
Conc.1	1.0	7.8	7.8	(1)
Conc.2	10	8.2	8.0	(1)
Conc.3	100	9.2	8.0	(1)

## (2) Test 2

Test Group	Nominal Concentration (mg/L)	pH		
		0 Hour	72 Hours (Vessel No.)	
Control	--	7.8	7.8	(1)
Conc.1	0.010	7.8	7.9	(1)
Conc.2	0.10	7.8	7.8 <sup>*1</sup>	(1)
		-	7.8 <sup>*1</sup>	(2)
		-	7.8 <sup>*1</sup>	(3)

\*1 : All vessels were measured, because the biomass of each vessel were different more than twice.

Table 4 Temperatures, Light Intensities and Revolutions in the Incubation Chamber

## (1) Test 1

Exposure Period (Hours)	Temperature (°C)	Light Intensity ( $\mu E/m^2/s$ )	Revolutions (rpm)
0	20.5	67-70	100
24	20.5	65-68	100
48	20.6	65-69	100
72	20.6	68-70	100

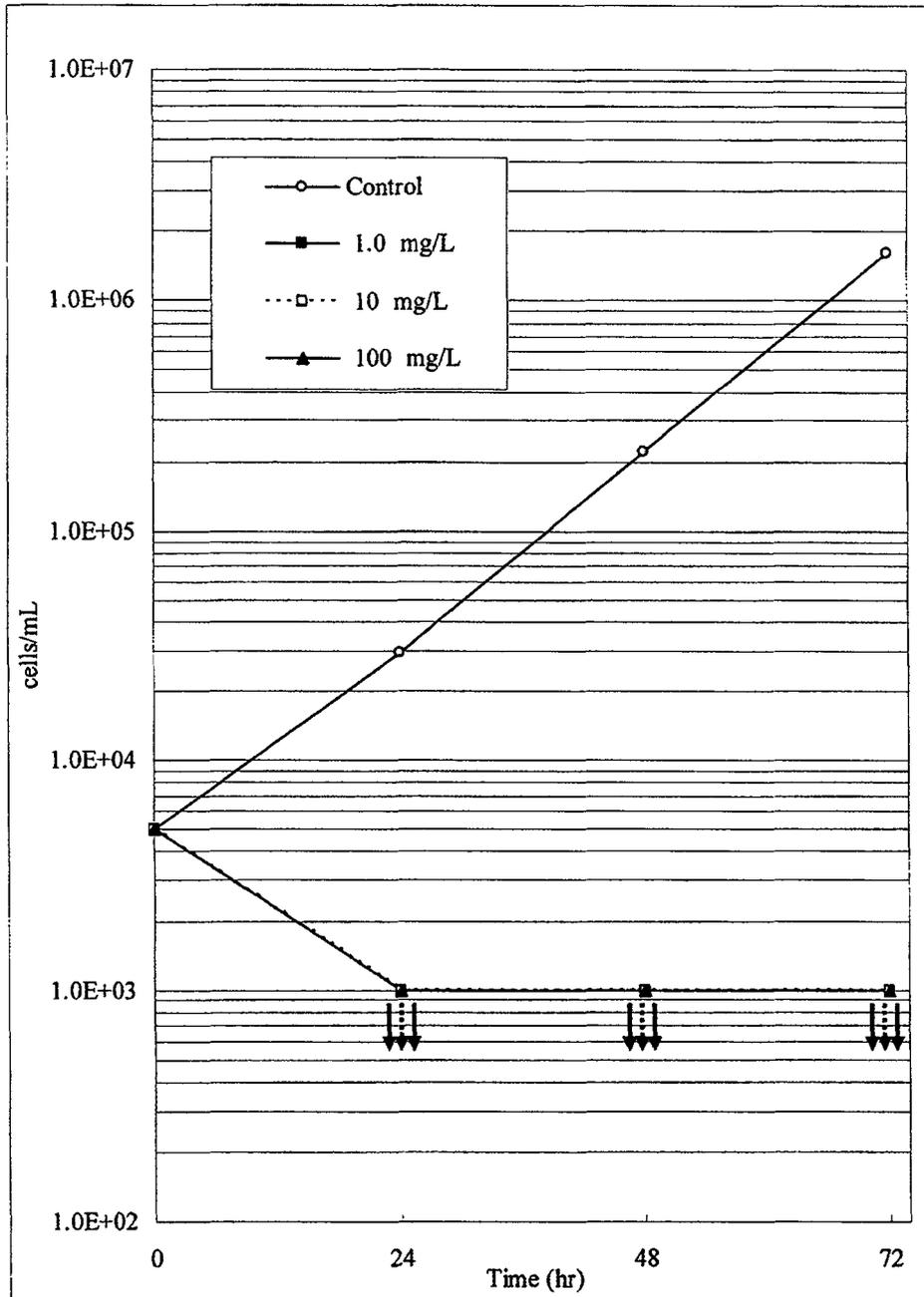
## (2) Test 2

Exposure Period (Hours)	Temperature (°C)	Light Intensity ( $\mu E/m^2/s$ )	Revolutions (rpm)
0	21.6	70-72	100
24	21.6	68-71	100
48	21.1	68-69	100
72	21.5	68-71	100

Figure 1 Algal Growth Curve of *Pseudokirchneriella subcapitata*

(Mean biomass vs time during the 72-hour exposure)

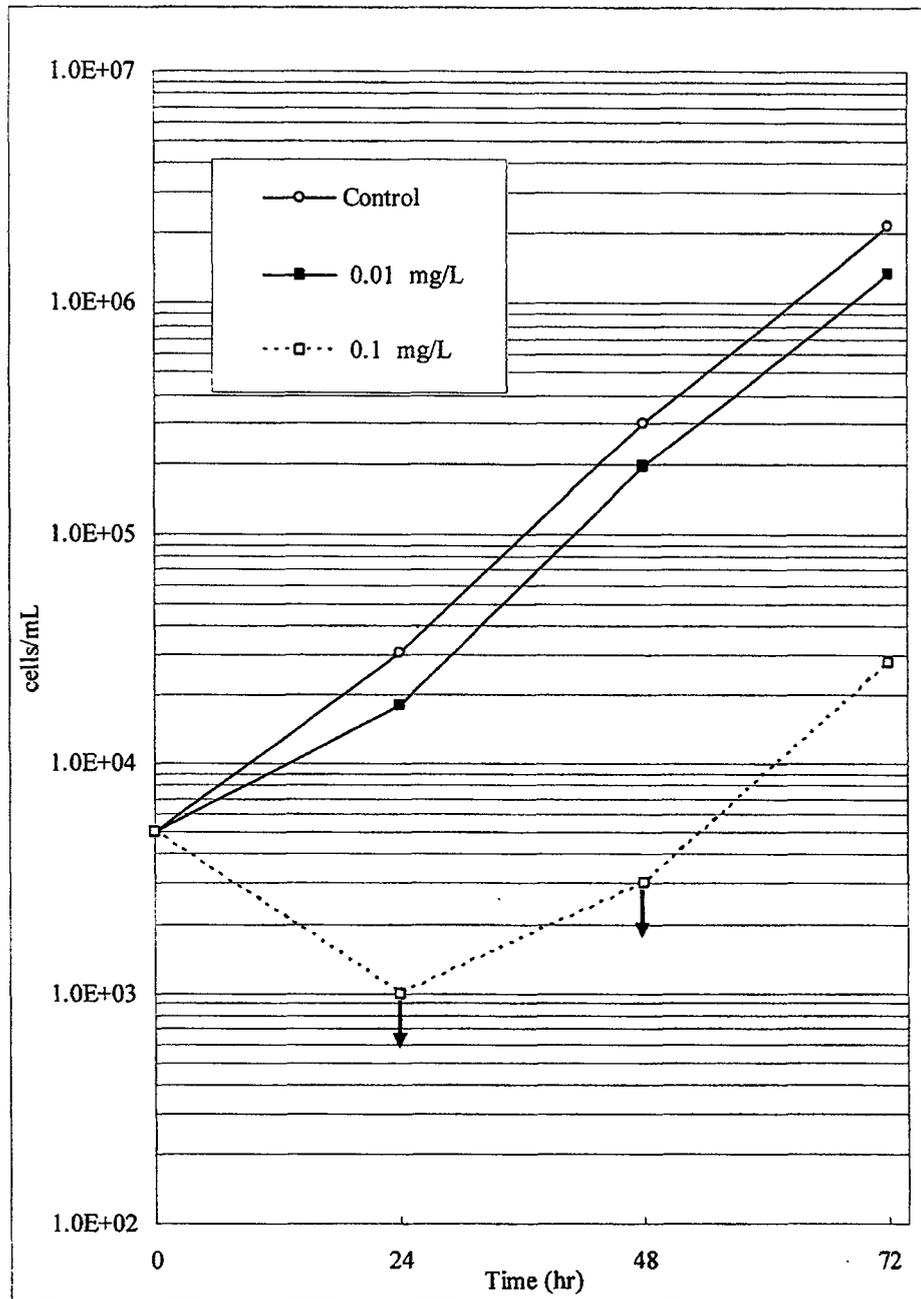
(1) Test 1



Values in legend are given in the nominal concentration.

Figure 1 Continued

(2) Test 2



Values in legend are given in the nominal concentration.