STUDY TITLE

HR-001: Reverse Mutation Test

DATA REQUIREMENT

Required under U.S. EPA FIFRA Guidelines, Subdivision F

AUTHOR

[redacted]

STUDY COMPLETED ON

April 3, 1995

PERFORMING LABORATORY

The Institute of Environmental Toxicology
Suzuki-cho 2-772, Kodaira-shi, Tokyo 187, Japan

LABORATORY PROJECT ID

IET 94-0142

SPONSOR

Sankyo Co., Ltd.
7-12, Ginza 2-chome, Chuo-ku,
Tokyo 104, Japan

STATEMENT OF DATA CONFIDENTIALITY CLAIMS

HR-001: Reverse Mutation Test
(IET 94-0142)

This report contains the unpublished results of research sponsored by Sankyo Co., Ltd. These results may not be published, either wholly or in part, or reviewed or quoted in any other publication without the authorization of Sankyo Co., Ltd.

GLP STATEMENT

HR-001: Reverse Mutation Test
(IET 94-0142)

This study was conducted in conformity to Good Laboratory Practice standards (GLPs) of MAFF in Japan (59 NohSan No. 3850, 1984), EPA in U.S.A. (FIFRA: 40 CFR 160, 1989), and OECD (OECD Principles of GLP, 1981).

The Institute of Environmental Toxicology

Administrator:

[redacted] | Apr. 3, 1995
Director General | Date

Study Director:

[redacted] Ph.D. | Apr. 3, 1995
Senior Scientist | Date
Laboratory of Genetic Toxicology
Toxicology Division

Sponsor:

________________ | ________
Sankyo Co., Ltd. | Date

Submitter:

________________ | ________
 | Date

FLAGGING STATEMENT

HR-001: Reverse Mutation Test
(IET 94-0142)

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HR-001: Reverse Mutation Test
(IET 94-0142)

OBJECTIVE

The purpose of this study was to evaluate the mutagenic potential of HR-001 for bacteria.

SPONSOR

Name: Sankyo Co., LTD.

Address: 7-12, Ginza 2-chome, Chuo-ku, Tokyo 104, Japan

TESTING INSTITUTION

Name: The Institute of Environmental Toxicology

Address: Suzuki-cho 2-772, Kodaira, Tokyo 187, Japan

TESTING FACILITY

Name: Kodaira Laboratories
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HR-001: Reverse Mutation Test
(IET 94-0142)

STUDY PERIOD

Establishment of contract:	January 12, 1995
Approval of protocol:	February 7, 1995
Testing period:	
Initiation of experiment:	February 21, 1995
Termination of experiment:	March 9, 1995
Draft report preparation:	March 20, 1995
Comments from sponsor:	March 31, 1995
Final report preparation:	April 3, 1995

STORAGE OF RECORDS

All records obtained during the conduct of this study will be retained in the archive of this institution for ten years after the submission of the final report to the sponsor. Storage after this period will be negotiated between the Institute of Environmental Toxicology and the sponsor.

HR-001: Reverse Mutatin Test
(IET 94-0142)

STUDY DIRECTOR AND SUPERVISORY PERSONNEL

We, the undersigned, hereby declare that the study was performed under our supervision in conformity to the GLPs of MAFF in Japan (59 NohSan No. 3850, 1984), EPA in U.S.A. (FIFRA: 40 CFR 160, 1989), and OECD (OECD Principles of GLP, 1981) and the Guidelines of MAFF in Japan (59 NohSan No. 4200, 1985), EPA in U.S.A. (Pesticide Assessment Guidelines, Subdivision F, 1991) and OECD (OECD Guideline Nos. 471, 472, 1983).

Study Director:

[redacted] Ph.D. Apr. 3, 1995
Date

Senior Scientist
Laboratory of Genetic Toxicology
Toxicology Division

Mutagenicity:

[redacted] April 3, 1995
Date

Acting Chief
Laboratory of Genetic Toxicology
Toxicology Division

STUDY DIRECTOR AND SUPERVISORY PERSONNEL (continued)

Executive Supervisor:

[redacted] .J.C.V.P. Date

D.J.S.T.P.
Director of Toxicology

PERSONNEL IN CHARGE

Mutagenicity Examination:

[redacted] D.V.M., Ph.D.
Senior Scientist, Laboratory of Genetic Toxicology
Toxicology Division

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Technician, Laboratory of Genetic Toxicology
Toxicology Division

Report Preparation:

[redacted] D.V.M., Ph.D.
Senior Scientist, Laboratory of Genetic Toxicology
Toxicology Division

QUALITY ASSURANCE AUTHORIZATION

HR-001: Reverse Mutation Test

(IET 94-0142)

Report

	Inspection date	Report date to the study director	Report date to the administrator
Protocol	1/10/1995	1/10/1995	1/11/1995
	2/ 2/1995	2/ 2/1995	2/ 2/1995
Study procedure	3/ 7/1995	3/ 7/1995	3/ 7/1995
Raw data	3/17/1995	3/17/1995	3/20/1995
Report	3/17/1995	3/17/1995	3/20/1995
	4/ 3/1995	4/ 3/1995	4/ 3/1995

By the above inspections, it was assured that the reported methods and procedures were found to describe those used and the results to reflect the raw data generated during the conduct of this study accurately.

Quality Assurance Manager:

[redacted]

April 3, 1995
Date

D.J.C.V.P.

Chief, Quality Assurance Unit

CONTENTS

1. SUMMARY

Reverse mutation tests were performed on HR-001 in *Escherichia coli* WP2 *uvrA* and four tester strains of *Salmonella typhimurium* (TA100, TA1535, TA98, and TA1537). Experiments were carried out with and without metabolic activation system (S9 Mix) at dose levels up to the highest dose of 5000 μg/plate. The mean number of revertant colonies did not exceed the factor of 2 above that of the corresponding solvent control in any strain at any dose with or without S9 Mix.

It is concluded that HR-001 is non-mutagenic to bacteria under the conditions used in this experiment.

2. OBJECTIVE

The purpose of this study is to evaluate the mutagenic potential of HR-001 for bacteria.

3. TEST SUBSTANCE

Name:	HR-001
Lot No.:	940908-1
Purity:	95.68%
Appearance at normal temperature:	white crystal
Melting point:	200℃
Solubility:	water, 12 g/l (25℃)
Storage condition:	dark cold room (5℃)

4. MATERIALS AND METHODS

1) Bacterial strains

Four histidine auxotroph strains of *Salmonella typhimurium* (TA100, TA1535, TA98 and TA1537) and *Escherichia coli* strain WP2 *uvrA* requiring tryptophan were used. Strains TA100 and TA98 were obtained on March 6, 1975 and others were on March 26, 1973 from Dr. [redacted], National Institute of Genetics, Mishima, Japan.

2) Examination of tester strains

The following genetic markers and other characters of the tester strains were checked regularly:

(1) Histidine and biotin requirements in *S. typhimurium* strains or tryptophan auxotroph in *E. coli* strain

(2) UV sensitivity (*uvrA*, *uvrB*)

(3) Sensitivity to crystal violet (*rfa*) in *S. typhimurium* strains

(4) Presence of the ampicillin-resistant plasmid (pKM101) in *S. typhimurium* strains TA100 and TA98

(5) Number of spontaneous revertants

(6) Response to positive control chemicals

3) Storage of tester strains and preculture

Each stock culture of tester strains has been stored at -80℃ (Ultra Low Temperature Cabinet, MDF-382AT, Sanyo Electric Co. Ltd., Japan) in the presence of dimethyl sulfoxide (DMSO, Dehydrous, Wako Pure Chemical Industries, Ltd., Japan) at the final concentration of 8% (v/v). Each of the five tester strains was inoculated with the nutrient broth medium (Oxoid nutrient broth No.2, Oxoid Ltd., U.K.) and cultured at 37℃ with shaking for 8hr. OD_{660} measured by Spectronic 21 (BAUSCH & LOMB, U.S.A.) was from 0.95 to 1.15 (1.4 - 2.3 x 10^9 cells/ml) at the end of culture.

4) Preparation of S9 Mix

A metabolic activation system (S9 mix) is a cofactor-supplemented post-mitochondrial fraction (S9 fraction) of liver homogenate of rats. S9 fraction with the following data was purchased from Kikkoman Corporation (Chiba, Japan) on October 26, 1994 and stored at -80℃.

(1) Used animal: Sprague-Dawley rat (Slc:SD)

(2) Sex: male

(3) Age: 7 weeks old

(4) Body weight: 192 - 229 g

(5) Inducer: phenobarbital (PB: Wako Pure Chemical Industries Ltd.,Japan)
5,6-benzoflavone (BF: Aldrich Chemical Co., Inc., U.S.A.)

(6) Treatment: intraperitoneal injection

(7) Dosage: Day 1: PB 30 mg/kg
Day 2: PB 60 mg/kg
Day 3: PB 60 mg/kg and BF 80 mg/kg
Day 4: PB 60 mg/kg

(8) Protein content: 24.40 mg/ml

(9) P-450 content: 1.04 nmol/mg protein

(10) Date of preparation: October 6, 1994

(11) Lot No.: RAA-316

(12) Sterility test: pass

(13) Enzyme activity measured by mutagenicity: good

The enzyme activity of this fraction was checked again by mutagenicity of 7,12-dimethylbenz(a)anthracene (Sigma Chemical Co., U.S.A., 95%) and 2-aminoanthracene (Wako Pure Chemical Industries Ltd., 96.5%) against *S. typhimurium* TA100 and TA98 in advance. The sterility of S9 fraction was also confirmed again in advance.

S9 Mix was prepared immediately before the experiment by mixing S9 fraction and Co-factor (freeze-drying co-factor mixture, Lot No. 718; Boehringer-Mannheim K. K., Japan). The components of S9 mix were 10% (v/v) S9 fraction, 8 mM $MgCl_2$, 33 mM KCl, 5 mM glucose-6-phosphate, 4 mM NADH, 4 mM NADPH, and 100 mM sodium phosphate buffer (pH 7.4).

5) Preparation of the test substance solution and dose levels

The solubility in water of HR-001 was known to be 12 mg/ml, while it was insoluble in DMSO at this concentration. Therefore, sterile water prepared by Milli-RO · 10 and Milli-Q Ultra-pure Water System (Nihon Millipore Ltd., Japan) was used as a solvent. HR-001 was suspended in sterile pure water at concentrations more than 12 mg/ml. The solution of the test substance was prepared immediately before the experiment.

In preliminary dose range finding tests (Table-1), HR-001 did not show any toxicity to any strain up to the highest dose of 5000 μg/plate with and without S9 Mix. Based on these results, 5000 μg/plate was used as the highest dose and experiments were carried out at 6 dose levels (156, 313, 625, 1250, 2500, and 5000 μg/plate). Toxicity was judged by a reduction in the number of revertant colonies or a clearing of the background lawn of histidine-biotin- or tryptophan-requiring cells.

6) Negative control and positive controls

For a negative control (solvent control), sterile water was used. The following mutagens were used as positive controls:

Strain	without S9 Mix (μg/plate)		with S9 Mix (μg/plate)	
TA100	AF-2	(0.01)	2-AA	(1)
TA1535	NaN_3	(0.5)	2-AA	(2)
WP2 *uvrA*	AF-2	(0.01)	2-AA	(10)
TA98	AF-2	(0.1)	2-AA	(0.5)
TA1537	9-AA	(80)	2-AA	(2)

AF-2; 2-(2-furyl)-3-(5-nitro-2-furyl)acrylamide (Wako Pure Chemical Industries Ltd., 99.4%, Lot No. SAJ0748)

2-AA; 2-aminoanthracene (Wako Pure Chemical Industries Ltd., 96.5%, Lot No. DSJ3206)

NaN_3; sodium azide (Wako Pure Chemical Industries Ltd., 92.2%, Lot No. DSG1561)

9-AA; 9-aminoacridine hydrochloride (Aldrich Chemical Co. Inc., 98%, Lot No. 09518LX)

AF-2 and 2-AA were dissolved in DMSO (Tokyo Kasei Kogyo Co., Japan, guaranteed reagent, >99.0%). NaN_3 and 9-AA were dissolved in sterile water.

7) Preparation of amino acid-supplemented soft agar

For the *S. typhimurium* strains, a sterile solution of 0.5 mM D-biotin and 0.5 mM L-histidine was added to molten soft agar consisting of 0.6% agar (Wako Pure Chemical Industries Ltd., Lot No. PTE7487) and 0.5% NaCl at a rate of 1/10 (v/v), and for the *E. coli* strain a sterile solution of 0.5 mM L-tryptophan was added at the same rate.

8) Experimental procedures

(1) Preincubation method without metabolic activation

An aliquot of 0.5 ml of 100 mM sodium phosphate buffer (pH 7.4), 0.1 ml of a culture of the tester strain, and 0.1 ml of a solution of the test substance were added to a small sterile test tube, and incubated with shaking for 20 min at 37℃. After that 2 ml of the amino acid-supplemented molten soft agar kept at 45℃

was added to the test tube. The contents were mixed uniformly and overlaid on a minimal glucose agar plate consisted of salt mix (0.2% citric acid monohydrate, 1% K_2HPO_4, 0.192% $NH_4H_2PO_4$, 0.066% NaOH, and 0.02% $MgSO_47H_2O$), 1.5% agar (OXOID Agar No. 1, Oxoid Ltd.), and 2% glucose. Prepared minimal glucose agar plates (Climedia AM-N, 30 ml/plate, Lot No. AN570KJ) were purchased from Oriental Yeast Co., Ltd., Japan. All plates were incubated at 37℃ for 48 hr, after which the number of revertant colonies was counted by a colony analyzer (Model CA-7II, Oriental Instruments Ltd., Japan). Triplicate plates were made for each dose. In addition, the solvent control and positive controls were included in the experiment.

(2) Preincubation method with metabolic activation

An aliquot of 0.5 ml of S9 Mix, 0.1 ml of a culture of the tester strain, and 0.1 ml of a solution of the test substance were added to a small sterile test tube, and incubated with shaking for 20 min at 37℃. After that, 2 ml of the amino acid-supplemented molten soft agar kept at 45℃ was added to the small test tube. The contents were mixed uniformly and overlaid on a minimal glucose agar plate. All plates were incubated at 37℃ for 48 hr, after which the number of revertant colonies was counted by the colony analyzer. Triplicate plates were made for each dose. In addition, the solvent control and positive controls were included in the experiment.

9) Assay acceptance criteria

An assay is considered acceptable for evaluation of the test results only if all of the criteria listed below are satisfied.

(1) The culture of tester strains, the solution of the test substance, and S9 mix are free from contamination by other bacteria.

(2) Normal number of spontaneous revertant colonies is observed in solvent control.

(3) At least 3-fold increase above solvent control in the mean number of revertants is observed in positive control.

10) Evaluation criteria

The tests were carried out twice. Reproducibility of results was confirmed by two independent experiments. Results were judged positive without statistical analysis when the following criteria are all satisfied:

(1) A two-fold or greater increase above solvent control in the mean number of revertants is observed.

(2) This increase in the number of revertants is accompanied by a dose-response relationship.

(3) This increase in the number of revertants is reproducible.

5. RESULTS

Results are shown in Tables 2-(1), 2-(2), 3-(1), and 3-(2). The mean number of revertant colonies did not exceed the factor of 2 above that of the corresponding solvent control in any strain at any dose of HR-001 whether S9 Mix was added or not. Dose-response curves are shown in Figs. 1-(1) to 1-(3) and 2-(1) to (3).

Normal number of spontaneous revertant colonies was observed in solvent control for all the strains. In contrast, AF-2, NaN_3, and 9-AA used as positive controls showed mutagenicity in the absence of S9 Mix, and 2-AA was mutagenic for all the strains in the presence of S9 Mix. All the cultures of the tester strains, the solution of the test substance, and S9 Mix were checked to be free from contamination by other bacteria. Consequently, all assays were considered acceptable for evaluation.

6. CONCLUSION

As described in the above results, a two-fold or greater increase in the mean number of revertant colonies was not observed in any strain at any dose of HR-001 in the reverse mutation tests with or without metabolic activation.

It is concluded that HR-001 is non-mutagenic for bacteria under the conditions used in this experiment.

7. REFERENCES

1) Ames, B.N., J. McCann, and E. Yamasaki: Mutation Res., 31: 347-364, 1975.

2) Mutagenicity Tests in Occupational Safety and Health Acts: Test Guideline and GLP (Ed., Investigation Division of Chemical Substances, Ministry of Labor), Japan Industrial Safety and Health Assoc., Tokyo, 1991 (in Japanese).

Table 1. Dose range finding tests

Test substance : HR-001

S9 Mix		Dose (μg/plate)	No. of revertant colonies/plate				
			Base-change type			Frameshift type	
			TA100	TA1535	WP2 uvrA	TA98	TA1537
−		Solvent control (H_2O)	124 111 (118)	14 18 (16)	25 21 (23)	17 25 (21)	7 5 (6)
		200	113	16	24	20	7
		500	116	15	23	10	3
		1000	116	16	20	14	4
		2000	82	10	15	5	5
		5000	79	3	19	8	3
+		Solvent control (H_2O)	83 86 (85)	11 9 (10)	21 25 (23)	29 28 (29)	6 10 (8)
		200	99	11	20	28	9
		500	82	7	12	30	6
		1000	97	8	28	25	6
		2000	96	9	18	38	7
		5000	33	4	17	20	5
Positive control	S9 Mix (−)	Compound	AF-2	NaN_3	AF-2	AF-2	9-AA
		μg/plate	0.01	0.5	0.01	0.1	80
		Revertants /plate	648 724 (686)	583 559 (571)	312 344 (328)	669 708 (689)	798 775 (787)
	S9 Mix (+)	Compound	2-AA	2-AA	2-AA	2-AA	2-AA
		μg/plate	1	2	10	0.5	2
		Revertants /plate	640 658 (649)	371 372 (372)	610 645 (628)	285 304 (295)	71 81 (76)

( ) : Average.

Table 2-(1) Reverse mutation tests without metabolic activation (Exp. I)

Test substance : HR-001

S9 Mix	Dose (μg/plate)	No. of revertant colonies/plate				
		Base-change type			Frameshift type	
		TA100	TA1535	WP2 uvrA	TA98	TA1537
−	Solvent control (H_2O)	110 121 119 (117± 6)	15 9 11 (12±3)	23 18 23 (21±3)	48 26 38 (37±11)	5 3 2 (3±2)
	156	124 108 124 (119± 9)	11 13 9 (11±2)	14 11 10 (12±2)	51 38 32 (40±10)	1 5 4 (3±2)
	313	130 106 116 (117±12)	12 10 12 (11±1)	13 13 22 (16±5)	37 48 40 (42± 6)	4 3 5 (4±1)
	625	135 134 148 (139± 8)	12 8 7 (9±3)	14 14 16 (15±1)	27 42 47 (39±10)	5 1 1 (2±2)
	1250	127 118 131 (125± 7)	10 10 8 (9±1)	18 22 25 (22±4)	37 45 48 (43± 6)	1 9 5 (5±4)
	2500	111 114 92 (106±12)	4 3 2 (3±1)	13 18 15 (15±3)	42 46 27 (38±10)	1 4 4 (3±2)
	5000	107 115 93 (105±11)	4 5 2 (4±2)	25 16 18 (20±5)	40 45 31 (39± 7)	1 2 2 (2±1)
Positive control	Compound	AF-2	NaN_3	AF-2	AF-2	9-AA
	μg/plat	0.01	0.5	0.01	0.1	80
	Revertants /plate	525 499 507 (510±13)	539 551 482 (524±37)	296 282 336 (305±28)	624 626 613 (621± 7)	691 839 828 (786±82)

( ) : Average ± S.D.

Table 2-(2) Reverse mutation tests with metabolic activation (Exp. I)

Test substance : HR-001

S9 Mix	Dose (μg/plate)	No. of revertant colonies/plate				
		Base-change type			Frameshift type	
		TA100	TA1535	WP2 uvrA	TA98	TA1537
+	Solvent control (H_2O)	79 73 83 (78± 5)	9 9 9 (9±0)	22 22 20 (21±1)	36 39 31 (35±4)	4 13 5 (7±5)
	156	80 88 82 (83± 4)	7 2 10 (6±4)	21 15 20 (19±3)	38 35 35 (36±2)	5 10 11 (9±3)
	313	82 73 76 (77± 5)	9 6 6 (7±2)	18 16 23 (19±4)	34 29 29 (31±3)	9 3 3 (5±3)
	625	111 92 93 (99±11)	2 10 7 (6±4)	20 18 18 (19±1)	27 36 27 (30±5)	9 10 4 (8±3)
	1250	106 98 74 (93±17)	4 8 6 (6±2)	18 20 28 (22±5)	36 41 33 (37±4)	7 5 5 (6±1)
	2500	73 61 85 (73±12)	13 2 5 (7±6)	14 16 18 (16±2)	39 38 41 (39±2)	6 5 10 (7±3)
	5000	55 48 66 (56± 9)	1 5 4 (3±2)	15 23 11 (16±6)	22 30 22 (25±5)	5 2 4 (4±2)
Positive control	Compound	2-AA	2-AA	2-AA	2-AA	2-AA
	μg/plat	1	2	10	0.5	2
	Revertants /plate	530 668 620 (606±70)	335 451 389 (392±58)	504 529 532 (522±15)	405 349 325 (360±41)	78 80 68 (75±6)

( ) : Average ± S.D.

Table 3-(1) Reverse mutation tests without metabolic activation (Exp. II)

Test substance : HR-001

S9 Mix	Dose (μg/plate)	No. of revertant colonies/plate				
		Base-change type			Frameshift type	
		TA100	TA1535	WP2 uvrA	TA98	TA1537
−	Solvent control (H_2O)	140 158 139 (146±11)	9 10 9 (9±1)	19 12 17 (16±4)	23 20 28 (24±4)	5 9 2 (5±4)
	156	137 144 131 (137± 7)	13 7 9 (10±3)	20 18 15 (18±3)	19 24 11 (18±7)	4 11 5 (7±4)
	313	154 128 137 (140±13)	7 9 5 (7±2)	16 20 20 (19±2)	27 18 14 (20±7)	3 4 5 (4±1)
	625	157 136 114 (136±22)	9 5 10 (8±3)	18 13 19 (17±3)	20 13 20 (18±4)	5 1 3 (3±2)
	1250	132 127 150 (136±12)	5 7 9 (7±2)	19 16 10 (15±5)	11 11 23 (15±7)	5 2 1 (3±2)
	2500	126 153 152 (144±15)	7 6 4 (6±2)	15 16 23 (18±4)	11 9 10 (10±1)	4 4 2 (3±1)
	5000	129 121 102 (117±14)	14 6 9 (10±4)	13 19 9 (14±5)	10 9 7 (9±2)	6 1 4 (4±3)
Positive control	Compound	AF-2	NaN_3	AF-2	AF-2	9-AA
	μg/plat	0.01	0.5	0.01	0.1	80
	Revertants /plate	610 607 569 (595±23)	427 568 587 (527±87)	284 235 238 (252±27)	701 760 765 (742±36)	1031 771 925 (909±131)

( ) : Average ± S.D.

Table 3-(2) Reverse mutation tests with metabolic activation (Exp. II)

Test substance : HR-001

S9 Mix	Dose (μg/plate)	No. of revertant colonies/plate				
		Base-change type			Frameshift type	
		TA100	TA1535	WP2 uvrA	TA98	TA1537
+	Solvent control (H_2O)	114 118 137 (123±12)	10 7 7 (8±2)	15 11 26 (17±8)	37 41 32 (37±5)	6 9 7 (7±2)
	156	106 102 129 (112±15)	11 5 6 (7±3)	15 16 15 (15±1)	43 29 25 (32±9)	14 6 9 (10±4)
	313	118 119 139 (125±12)	5 5 10 (7±3)	11 11 18 (13±4)	36 25 27 (29±6)	9 7 11 (9±2)
	625	116 108 115 (113± 4)	9 7 7 (8±1)	24 21 14 (20±5)	30 42 34 (35±6)	7 6 13 (9±4)
	1250	113 96 111 (107± 9)	4 7 9 (7±3)	15 13 15 (14±1)	29 31 24 (28±4)	9 9 10 (9±1)
	2500	81 86 100 (89±10)	9 7 4 (7±3)	16 27 14 (19±7)	19 15 27 (20±6)	10 10 5 (8±3)
	5000	67 70 64 (67± 3)	5 5 1 (4±2)	22 16 14 (17±4)	20 13 19 (17±4)	2 6 5 (4±2)
Positive control	Compound	2-AA	2-AA	2-AA	2-AA	2-AA
	μg/plat	1	2	10	0.5	2
	Revertants /plate	725 748 831 (768±56)	286 346 333 (322±32)	583 645 586 (605±35)	350 316 315 (327±20)	83 81 98 (87±9)

( ) : Average ± S.D.

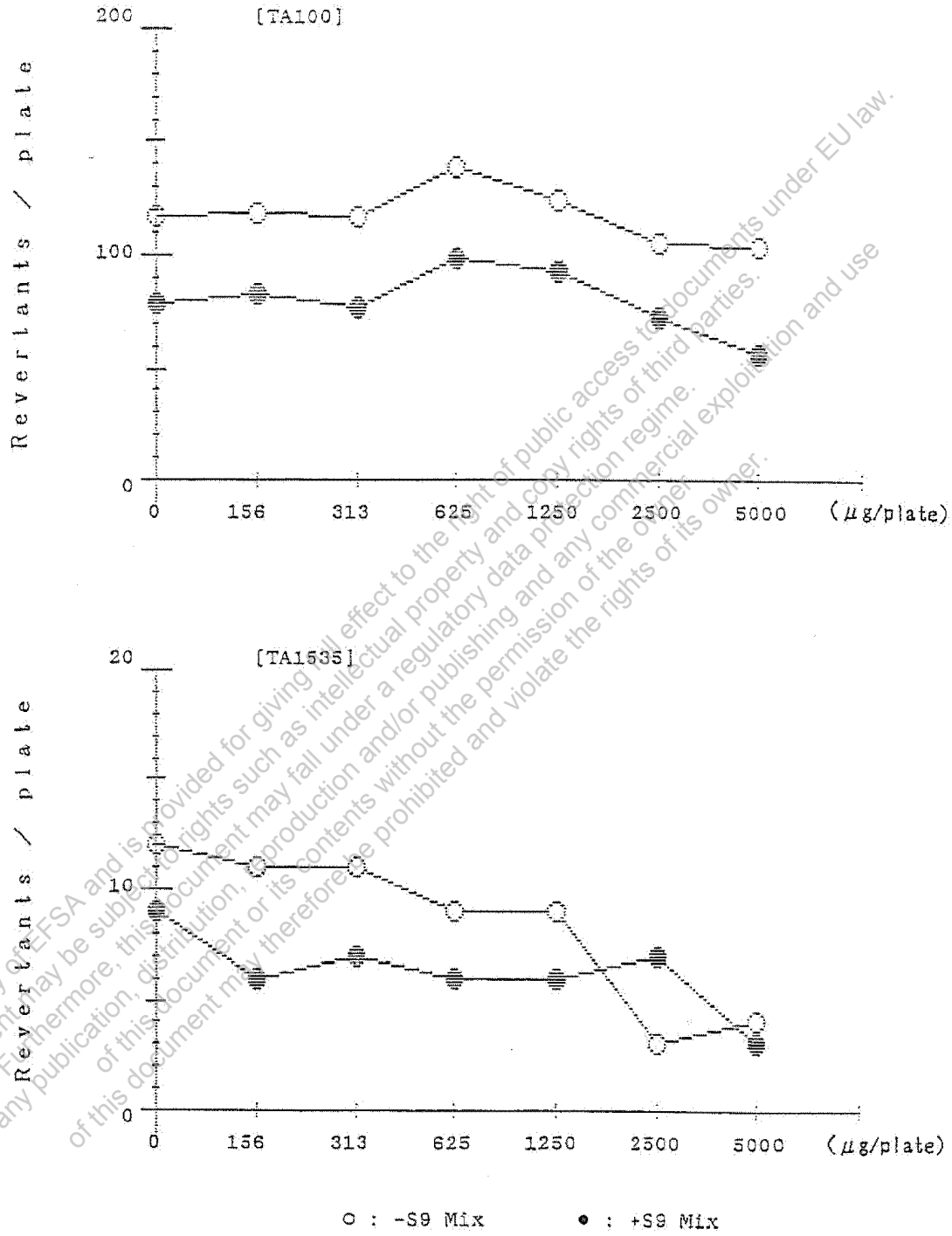

Fig. 1-(1). Dose-response curve (Exp.I, TA100, TA1535)

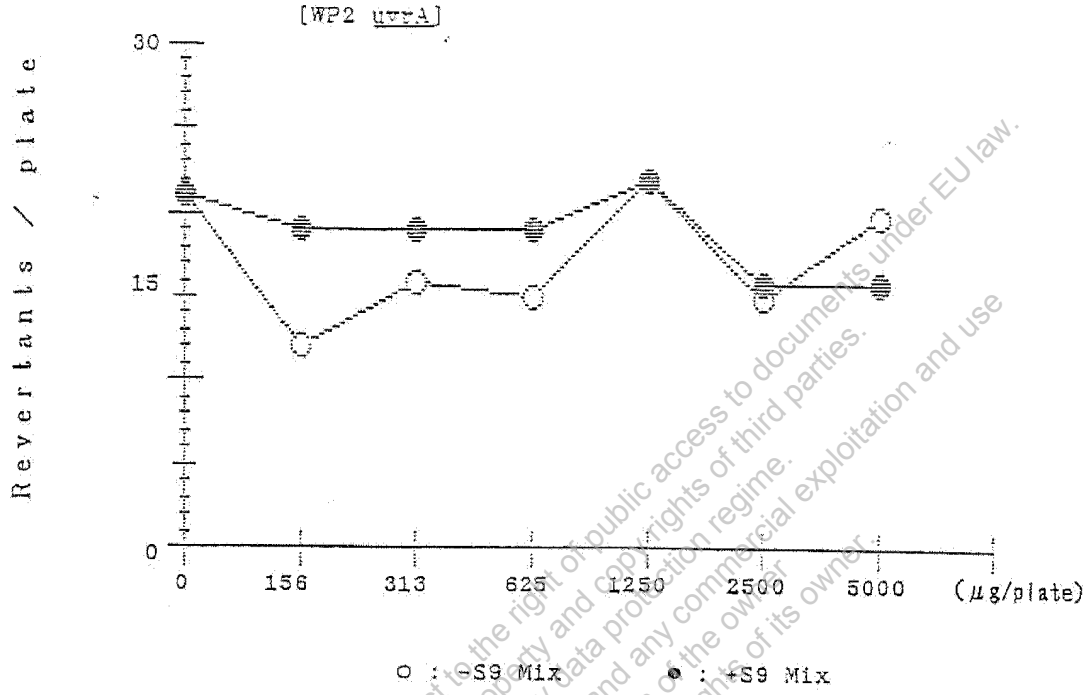

Fig. 1-(2). Dose-response curve (Exp.I, WP2 uvrA)

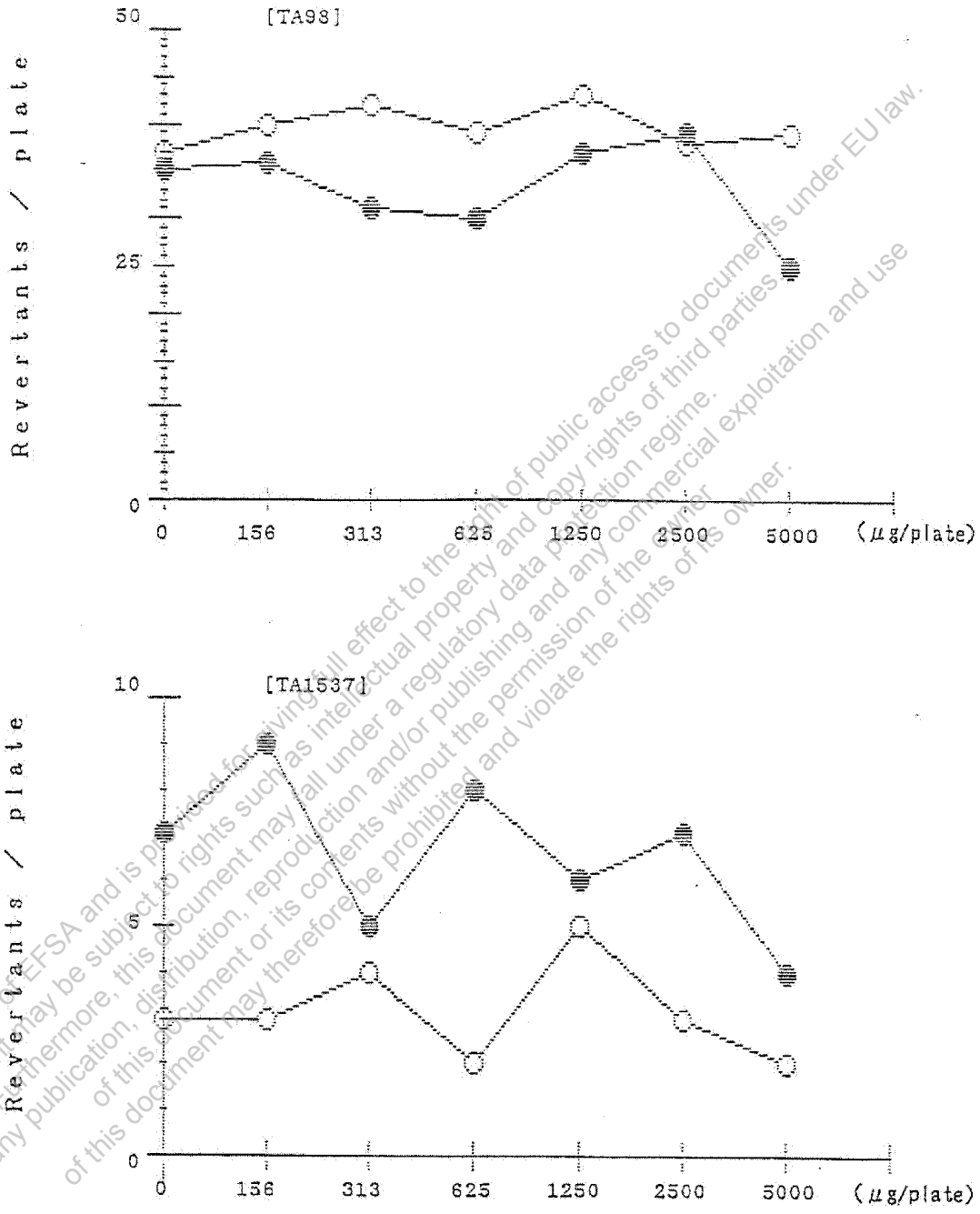

Fig. 1-(3). Dose-response curve (Exp.I, TA98, TA1537)

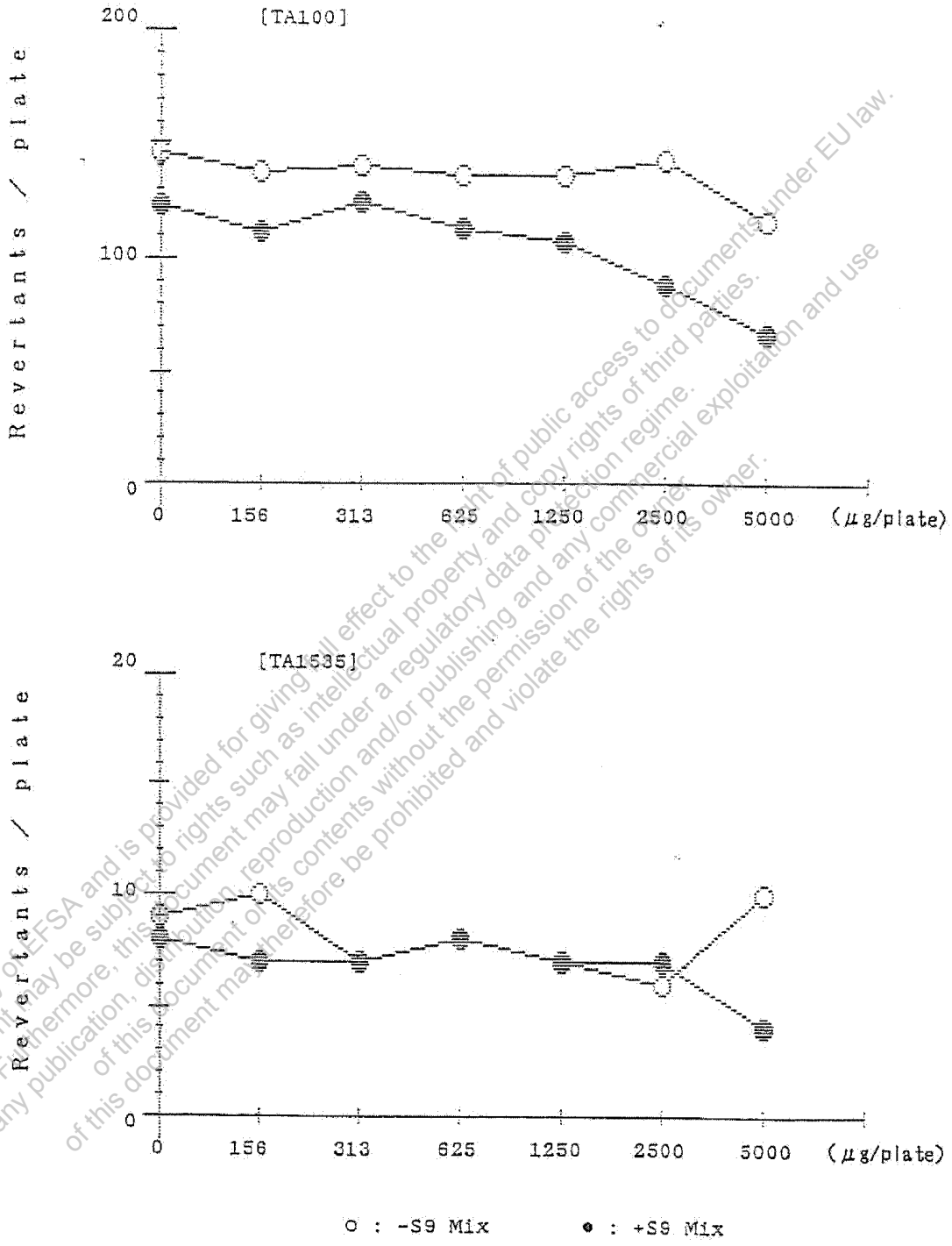

Fig. 2-(1). Dose-response curve (Exp.II, TA100, TA1535)

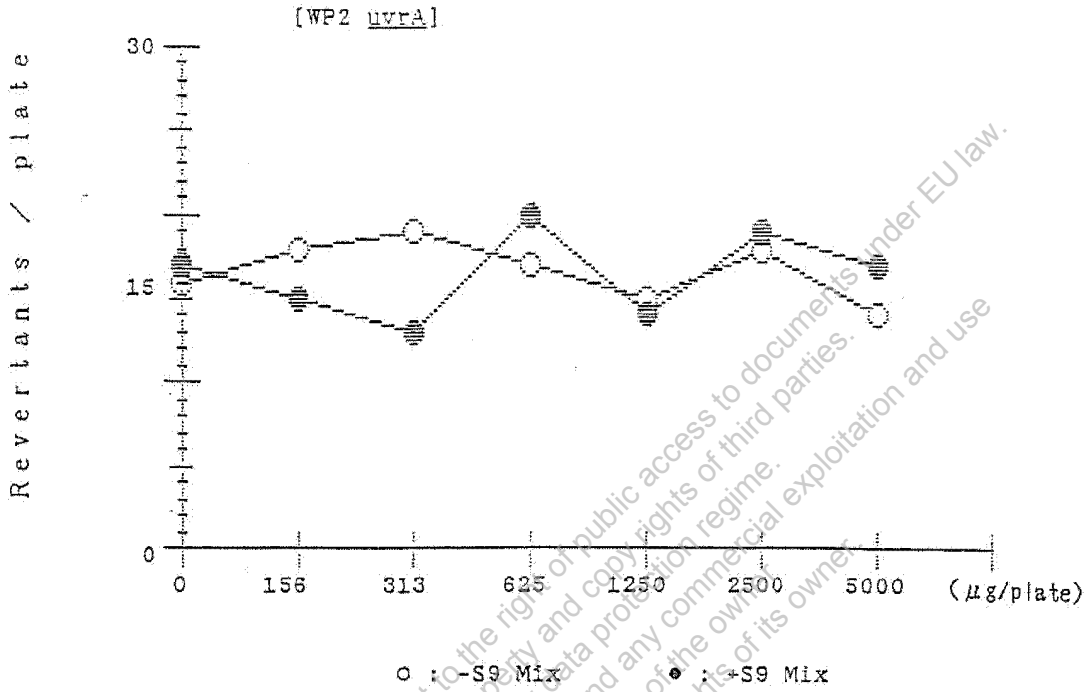

Fig. 2-(2). Dose-response curve (Exp.II, WP2 uvrA)

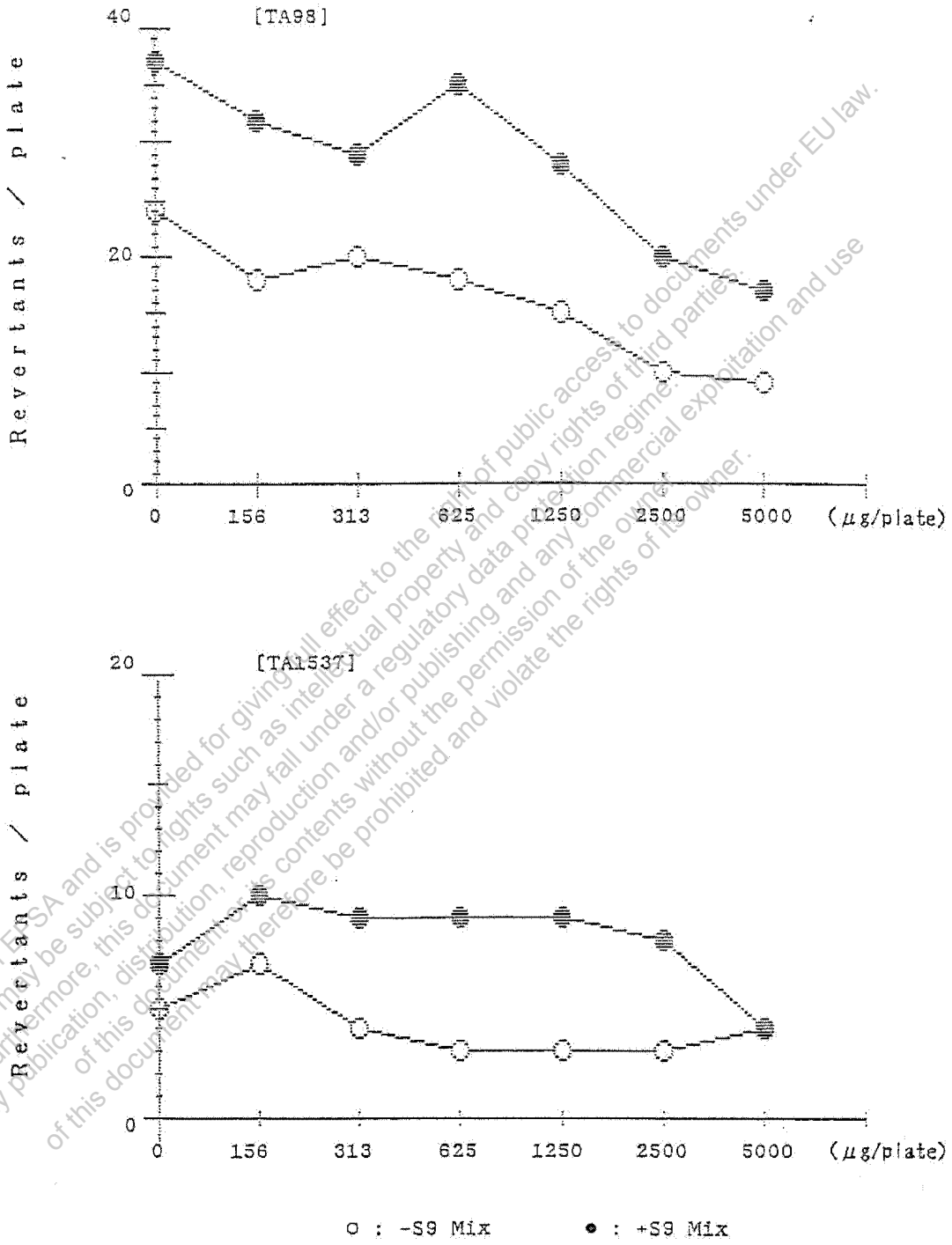

Fig. 2-(3). Dose-response curve (Exp.II, TA98, TA1537)