Summary of Drinking Water Carcinogenicity Study of 2-Hydroxyethyl Acrylate in BDF1 Mice

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Japan Bioassay Research Center

Japan Industrial Safety and Health Association

PREFACE

The tests were contracted and supported by the Ministry of Health, Labour and Welfare of Japan. The tests were conducted by Japan Bioassay Research Center (JBRC) and the report was prepared by JBRC and peer reviewed by outside expert pathologist. Complete report was submitted to Ministry of Health, Labour and Welfare of Japan on September 24, 2003.

This English Summary was translated by JBRC from Japanese complete report.

Summary of Drinking Water Carcinogenicity Study of 2-Hydroxyethyl Acrylate in BDF1 Mice

Purpose, materials and methods

2-Hydroxyethyl acrylate (HEA, CAS No. 818-61-1) is a transparent liquid with a boiling point of 82°C and is soluble in water.

The carcinogenicity and chronic toxicity of HEA were examined in groups of 50 Crj:BDF1 mice of both sexes administered HEA in drinking water for 2 years (104 weeks). The drinking water concentration of HEA was 0, 750, 1500 or 3000 ppm (w/w) for male mice and 0, 500, 1500 or 4500 ppm for female mice. The highest dose levels were chosen so as not to exceed the maximum tolerated dose (MTD), based on both growth rate and toxicity in the previous 13-week toxicity study. HEA was analyzed for purity and stability by both infrared spectrometry and gas chromatography before and after its use. The concentrations of HEA in drinking water were determined by gas chromatography at the time of preparation, and on the 4th, 8th and 11th days after preparation, while stored at room temperature. The animals were observed daily for clinical signs and mortality. Body weight, water consumption and food consumption were measured once a week for the first 14 weeks and every 4 weeks thereafter. Animals found dead, in a moribund state, or surviving to the end of the 2-year administration period underwent complete necropsy. Urinalysis was performed near the end of the 2-year administration period. For hematology and blood biochemistry, the surviving animals were bled under ether anesthesia, after they were fasted overnight, at the terminal necropsy. Organs and tissues were removed, weighed and examined for macroscopic lesions at necropsy. The organs and tissues were fixed and embedded in paraffin. Tissue sections of 5 μm thick were prepared and stained with hematoxylin and eosin and examined for histopathology. Incidences of neoplastic lesions were statistically analyzed by Fisher's exact test. A positive trend of dose-response relationship for the neoplastic incidence was analyzed by Peto's test. Incidences of non-neoplastic lesions and urinalysis were analyzed by Chi-square test. Changes in body weight, water consumption, food consumption, hematological and blood biochemical parameters, and organ weights were analyzed by Dunnett's test. The present studies were conducted in accordance with the Organisation for Economic Co-operation and Development (OECD) Good Laboratory Practice and with reference to the OECD Guideline for Testing of Chemicals 451 "Carcinogenicity Studies".

Results

Body weights, water consumption and food consumption were decreased dose-dependently. There was no difference in survival rate between any HEA-administered group of either sex and the respective control.

The incidence of neoplastic lesions was not increased in any HEA-administered group of either sex. On the contrary, the incidences of hepatocellular carcinomas in the HEA-administered males, and pituitary adenomas and adenocarcinomas in the HEA-administered females were decreased. As non-neoplastic lesions, the incidence of forestomach squamous cell hyperplasia was increased in the 3000 ppm-administered males and in all the HEA-administered female groups. The incidence of urothelial desquamation of pelvis was increased in the 3000 ppm-administered males and in the females administered 1500 ppm and above. Respiratory metaplasia in the nasal gland was observed in all the HEA-administered male groups and in the females administered 1500 ppm and above.

A lowest-observed-adverse-effect-level (LOAEL) for the endpoint of forestomach squamous cell hyperplasia was estimated at 500 ppm (equivalent to 0.048 to 0.093 g/kg/day). A lower confidence limit of the benchmark dose yielding a response with 10% extra risk ($BMDL_{10}$) for the same endpoint was determined at 489 ppm.

Conclusions

In mice, there was no evidence of carcinogenic activity of HEA in males or females. The incidences of hepatocellular tumors in the HEA -administered males and pituitary tumors in the HEA -administered females were decreased. As non-neoplastic lesions, forestomach squamous cell hyperplasia, desquamation of pelvis and respiratory metaplasia of the nasal gland were observed.

TABLES

TABLES (Continued)

TABLES (Continued)

TABLE 1 SURVIVAL ANIMAL NUMBERS AND BODY WEIGHT CHANGES OF MALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group	Control		750 ppm			1500 ppm			3000 ppm		
	<50>		<50>			<50>			<50>		
Week on Study	Survival No.	BW g	Survival No.	BW g	%	Survival No.	BW g	%	Survival No.	BW g	%
0	50	23.0 (50)	50	23.0 (50)	100	50	23.0 (50)	100	50	23.0 (50)	100
1	50	23.6 (50)	50	23.4 (50)	99	50	23.4 (50)	99	50	22.8 (50)	97 **
2	50	24.6 (50)	50	24.6 (50)	100	50	24.6 (50)	100	50	23.8 (50)	97 **
3	50	25.2 (50)	49	25.0 (49)	99	50	25.2 (50)	100	50	24.3 (50)	96 **
4	50	25.8 (50)	49	26.0 (49)	101	50	25.6 (50)	99	50	24.8 (50)	96 **
5	50	26.9 (50)	49	26.7 (49)	99	50	26.1 (50)	97 **	50	25.3 (50)	94 **
6	50	27.6 (50)	49	27.2 (49)	99	50	26.2 (50)	95 **	50	25.1 (50)	91 **
7	50	28.6 (50)	49	28.0 (49)	98	50	27.2 (50)	95 **	50	26.0 (50)	91 **
8	50	29.2 (50)	49	28.7 (49)	98	50	27.6 (50)	95 **	50	26.3 (50)	90 **
9	50	29.8 (50)	49	29.2 (49)	98	50	28.4 (50)	95 **	50	26.9 (50)	90 **
10	50	30.8 (50)	49	30.4 (49)	99	50	29.1 (50)	94 **	50	27.3 (50)	89 **
11	50	32.1 (50)	49	31.3 (49)	98	49	30.0 (49)	93 **	50	27.9 (50)	87 **
12	50	32.3 (50)	49	31.5 (49)	98	49	30.1 (49)	93 **	50	28.1 (50)	87 **
13	50	33.2 (50)	49	32.4 (49)	98	49	31.1 (49)	94 **	50	29.1 (50)	88 **
14	50	33.4 (50)	49	32.5 (49)	97	49	31.2 (49)	93 **	50	29.1 (50)	87 **
18	50	36.2 (50)	49	35.4 (49)	98	49	33.5 (49)	93 **	50	30.9 (50)	85 **
22	50	38.1 (50)	49	36.7 (49)	96	49	34.5 (49)	91 **	50	31.7 (50)	83 **
26	50	40.1 (50)	49	38.5 (49)	96	49	35.8 (49)	89 **	50	32.7 (50)	82 **
30	50	41.9 (50)	49	40.0 (49)	95	49	36.9 (49)	88 **	50	33.5 (50)	80 **
34	50	42.7 (50)	49	41.0 (49)	96	49	37.5 (49)	88 **	50	33.9 (50)	79 **
38	50	44.2 (50)	49	42.2 (49)	95 *	49	38.4 (49)	87 **	50	34.7 (50)	79 **
42	50	44.9 (50)	49	43.3 (49)	96	49	39.4 (49)	88 **	50	35.6 (50)	79 **
46	50	46.5 (50)	49	44.7 (49)	96	49	40.3 (49)	87 **	50	36.4 (50)	78 **
50	50	47.4 (50)	49	45.6 (49)	96	49	40.8 (49)	86 **	50	37.0 (50)	78 **
54	50	48.1 (50)	49	46.3 (49)	96	49	41.7 (49)	87 **	49	37.9 (49)	79 **
58	49	48.5 (49)	49	46.4 (49)	96	49	41.4 (49)	85 **	49	38.3 (49)	79 **
62	49	49.4 (49)	48	48.0 (48)	97	49	42.5 (49)	86 **	49	39.3 (49)	80 **
66	49	50.5 (49)	47	48.7 (47)	96	49	42.6 (49)	84 **	49	39.3 (49)	78 **
70	49	51.1 (49)	47	49.1 (47)	96	48	42.9 (48)	84 **	47	39.5 (47)	77 **
74	49	51.1 (49)	47	48.4 (47)	95 *	47	42.8 (47)	84 **	46	39.2 (46)	77 **
78	47	51.2 (47)	46	48.8 (46)	95	46	42.9 (46)	84 **	45	39.0 (45)	76 **
82	46	51.4 (46)	46	48.8 (46)	95 *	46	43.0 (46)	84 **	44	39.0 (44)	76 **
86	46	51.2 (46)	44	47.9 (44)	94 *	45	42.0 (45)	82 **	44	38.3 (44)	75 **
90	45	51.8 (45)	43	47.8 (43)	92 **	44	42.0 (44)	81 **	44	38.6 (44)	75 **
94	43	52.0 (43)	40	47.3 (40)	91 **	43	41.4 (43)	80 **	44	38.0 (44)	73 **
98	43	50.7 (43)	36	47.1 (36)	93	41	41.3 (41)	81 **	44	37.7 (44)	74 **
102	38	51.0 (38)	36	45.9 (36)	90	41	40.8 (41)	80 **	44	37.5 (44)	74 **
104	37	51.1 (37)	36	45.8 (36)	90 **	38	40.4 (38)	79 **	43	37.4 (43)	73 **

< > : No.of effective animals, ( ) : No.of measured animals % : % of control group
Significant Difference, * : $p \leqq 0.05$, ** : $p \leqq 0.01$, Test of Dunnett

TABLE 2 SURVIVAL ANIMAL NUMBERS AND BODY WEIGHT CHANGES OF FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group	Control		500 ppm			1500 ppm			4500 ppm		
	<50>		<50>			<49>			<50>		
Week on Study	Survival No.	BW g	Survival No.	BW g	%	Survival No.	BW g	%	Survival No.	BW g	%
0	50	18.9 (50)	50	18.9 (50)	100	49	18.9 (49)	100	50	18.9 (50)	100
1	50	19.4 (50)	50	18.9 (50)	97	49	19.3 (49)	99	50	18.5 (50)	95 **
2	50	19.8 (50)	50	20.0 (50)	101	49	19.8 (49)	100	50	19.2 (50)	97 **
3	50	20.2 (50)	50	20.2 (50)	100	49	20.2 (49)	100	50	19.7 (50)	98 **
4	50	21.0 (50)	50	21.2 (50)	101	49	21.1 (49)	100	50	20.1 (50)	96 **
5	50	21.4 (50)	50	21.5 (50)	100	49	21.3 (49)	100	50	20.6 (50)	96 **
6	50	21.9 (50)	50	21.9 (50)	100	49	21.9 (49)	100	50	21.1 (50)	96 **
7	50	22.4 (50)	50	22.5 (50)	100	49	22.4 (49)	100	50	21.6 (50)	96 **
8	50	22.8 (50)	50	22.7 (50)	100	49	22.6 (49)	99	50	21.8 (50)	96 **
9	50	23.3 (50)	50	23.2 (50)	100	49	23.1 (49)	99	50	22.5 (50)	97 **
10	50	23.4 (50)	50	23.4 (50)	100	49	23.1 (49)	99	50	22.4 (50)	96 **
11	50	24.0 (50)	50	24.0 (50)	100	49	23.6 (49)	98	50	23.0 (50)	96 **
12	50	24.0 (50)	50	24.0 (50)	100	49	23.5 (49)	98	50	23.0 (50)	96 **
13	50	24.4 (50)	50	24.3 (50)	100	49	24.0 (49)	98	50	23.2 (50)	95 **
14	50	24.6 (50)	50	24.4 (50)	99	49	23.9 (49)	97 *	50	23.3 (50)	95 **
18	50	26.0 (50)	50	25.6 (50)	98	49	24.6 (49)	95 **	50	23.9 (50)	92 **
22	50	26.6 (50)	50	26.5 (50)	100	49	25.4 (49)	95 **	50	24.5 (50)	92 **
26	49	27.9 (49)	50	27.6 (50)	99	49	26.4 (49)	95 **	50	25.0 (50)	90 **
30	49	29.0 (49)	50	28.4 (50)	98	49	26.6 (49)	92 **	50	25.2 (50)	87 **
34	49	29.4 (49)	50	28.8 (50)	98	49	26.7 (49)	91 **	50	25.5 (50)	87 **
38	49	29.8 (49)	50	29.7 (50)	100	49	27.1 (49)	91 **	50	25.8 (50)	87 **
42	49	30.6 (49)	50	30.1 (50)	98	49	27.7 (49)	91 **	49	25.9 (49)	85 **
46	49	31.3 (49)	50	30.9 (50)	99	49	28.3 (49)	90 **	49	26.2 (49)	84 **
50	49	32.1 (49)	50	31.5 (50)	98	49	28.5 (49)	89 **	49	26.4 (49)	82 **
54	49	32.6 (49)	50	31.7 (50)	97	49	28.5 (49)	87 **	49	26.7 (49)	82 **
58	49	32.8 (49)	49	32.5 (49)	99	48	29.1 (48)	89 **	49	26.6 (49)	81 **
62	48	33.6 (48)	49	32.9 (49)	98	48	29.2 (48)	87 **	49	27.0 (49)	80 **
66	48	34.5 (48)	48	33.1 (48)	96	48	29.6 (48)	86 **	49	27.1 (49)	79 **
70	47	35.1 (47)	47	33.5 (47)	95	48	29.7 (48)	85 **	47	27.2 (47)	77 **
74	46	35.2 (46)	47	33.2 (47)	94	48	29.4 (48)	84 **	46	27.1 (46)	77 **
78	45	35.5 (45)	45	33.8 (45)	95	48	30.1 (48)	85 **	46	27.2 (46)	77 **
82	44	36.1 (44)	45	34.1 (45)	94	47	29.6 (47)	82 **	45	27.5 (45)	76 **
86	43	34.8 (43)	43	33.7 (43)	97	46	29.7 (46)	85 **	44	27.1 (44)	78 **
90	41	35.7 (41)	40	34.1 (40)	96	43	30.1 (43)	84 **	42	27.0 (42)	76 **
94	39	36.0 (39)	35	33.6 (35)	93	39	30.3 (39)	84 **	41	27.2 (41)	76 **
98	38	36.5 (38)	30	33.6 (30)	92	36	30.1 (36)	82 **	40	27.1 (40)	74 **
102	33	34.4 (33)	25	32.9 (25)	96	27	29.6 (27)	86 **	38	26.8 (38)	78 **
104	31	34.6 (31)	24	33.3 (24)	96	27	29.6 (27)	86 **	36	26.7 (36)	77 **

< > : No.of effective animals, ( ) : No.of measured animals % : % of control group
Significant Difference, * : $p \leqq 0.05$, ** : $p \leqq 0.01$, Test of Dunnett

TABLE 3 WATER CONSUMPTION CHANGES OF MALE MICE
IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group	Control		750 ppm				1500 ppm				3000 ppm			
Week on Study	<50> Survival No.	WC g	<50> Survival No.	WC g	%		<50> Survival No.	WC g	%		<50> Survival No.	WC g	%	
1	50	4.5 (49)	50	3.6 (50)	80	**	50	3.1 (50)	69	**	50	2.5 (50)	56	**
2	50	4.4 (48)	50	3.5 (49)	80	**	50	2.8 (50)	64	**	50	2.2 (50)	50	**
3	50	4.4 (48)	49	3.6 (49)	82	**	50	2.8 (50)	64	**	50	2.3 (50)	52	**
4	50	4.4 (49)	49	3.5 (49)	80	**	50	2.8 (50)	64	**	50	2.3 (50)	52	**
5	50	4.5 (48)	49	3.5 (49)	78	**	50	2.9 (50)	64	**	50	2.4 (50)	53	**
6	50	4.3 (48)	49	3.6 (49)	84	**	50	3.0 (50)	70	**	50	2.3 (50)	53	**
7	50	4.4 (49)	49	3.7 (49)	84	**	50	2.9 (50)	66	**	50	2.5 (50)	57	**
8	50	4.3 (50)	49	3.4 (49)	79	**	50	2.7 (50)	63	**	50	2.3 (50)	53	**
9	50	4.2 (50)	49	3.3 (49)	79	**	50	2.6 (50)	62	**	50	2.3 (50)	55	**
10	50	4.1 (50)	49	3.2 (49)	78	**	50	2.5 (50)	61	**	50	2.3 (50)	56	**
11	50	4.0 (50)	49	3.2 (49)	80	**	49	2.6 (49)	65	**	50	2.2 (50)	55	**
12	50	3.9 (50)	49	3.1 (49)	79	**	49	2.6 (49)	67	**	50	2.2 (50)	56	**
13	50	3.7 (50)	49	3.1 (49)	84	**	49	2.6 (49)	70	**	50	2.2 (50)	59	**
14	50	3.8 (50)	49	3.1 (49)	82	**	49	2.5 (49)	66	**	50	2.2 (50)	58	**
18	50	3.5 (50)	49	3.0 (49)	86	**	49	2.4 (49)	69	**	50	2.1 (50)	60	**
22	50	3.3 (50)	49	2.8 (49)	85	**	49	2.4 (49)	73	**	50	2.1 (50)	64	**
26	50	3.6 (50)	49	3.0 (49)	83	**	49	2.5 (49)	69	**	50	2.1 (50)	58	**
30	50	3.6 (50)	49	3.1 (49)	86	**	49	2.6 (49)	72	**	50	2.1 (50)	58	**
34	50	3.6 (50)	49	3.1 (49)	86	**	49	2.5 (49)	69	**	50	2.2 (50)	61	**
38	50	3.7 (50)	49	3.1 (49)	84	**	49	2.6 (49)	70	**	50	2.2 (50)	59	**
42	50	3.8 (50)	49	3.3 (49)	87	**	49	2.8 (49)	74	**	50	2.3 (50)	61	**
46	50	3.9 (50)	49	3.4 (49)	87	**	49	2.8 (49)	72	**	50	2.4 (49)	62	**
50	50	3.8 (50)	49	3.3 (49)	87	**	49	2.7 (49)	71	**	50	2.3 (49)	61	**
54	50	3.9 (50)	49	3.5 (49)	90	**	49	2.8 (49)	72	**	49	2.4 (49)	62	**
58	49	4.1 (49)	49	3.4 (49)	83	**	49	2.8 (49)	68	**	49	2.4 (49)	59	**
62	49	4.1 (49)	48	3.6 (48)	88	**	49	2.9 (49)	71	**	49	2.5 (49)	61	**
66	49	4.3 (49)	47	3.6 (47)	84	**	49	3.0 (49)	70	**	49	2.6 (49)	60	**
70	49	4.4 (49)	47	3.6 (47)	82	**	48	3.0 (48)	68	**	47	2.6 (47)	59	**
74	49	4.2 (49)	47	3.6 (47)	86	**	47	3.0 (47)	71	**	46	2.6 (46)	62	**
78	47	4.3 (47)	46	3.6 (45)	84	**	46	3.1 (46)	72	**	45	2.7 (45)	63	**
82	46	4.3 (46)	46	3.5 (46)	81	**	46	3.0 (46)	70	**	44	2.5 (44)	58	**
86	46	4.5 (46)	44	3.7 (44)	82	**	45	3.0 (45)	67	**	44	2.6 (44)	58	**
90	45	4.7 (45)	43	4.0 (43)	85	**	44	3.2 (44)	68	**	44	2.7 (44)	57	**
94	43	4.6 (43)	40	3.9 (39)	85	**	43	3.2 (43)	70	**	44	2.8 (44)	61	**
98	43	4.8 (43)	36	4.0 (36)	83	*	41	3.3 (41)	69	**	44	2.8 (44)	58	**
102	38	4.9 (38)	36	4.0 (36)	82	**	41	3.4 (41)	69	**	44	2.8 (44)	57	**
104	37	4.7 (37)	36	4.0 (36)	85	*	38	3.3 (38)	70	**	43	2.7 (43)	57	**

< > : No.of effective animals, ( ) : No.of measured animals % : % of control group
Significant Difference, * : p≦0.05, ** : p≦0.01, Test of Dunnett

TABLE 4 WATER CONSUMPTION CHANGES OF FEMALE MICE
IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group	Control		500 ppm				1500 ppm				4500 ppm			
Week on Study	<50> Survival No.	WC g	<50> Survival No.	WC g	%		<49> Survival No.	WC g	%		<50> Survival No.	WC g	%	
1	50	4.3 (50)	50	3.4 (50)	79	**	49	3.0 (49)	70	**	50	2.0 (50)	47	**
2	50	4.7 (48)	50	3.7 (50)	79	**	49	2.9 (49)	62	**	50	1.9 (50)	40	**
3	50	4.3 (48)	50	3.5 (50)	81	**	49	2.9 (49)	67	**	50	2.0 (50)	47	**
4	50	4.0 (50)	50	3.2 (50)	80	**	49	2.7 (49)	68	**	50	1.8 (50)	45	**
5	50	4.3 (50)	50	3.3 (50)	77	**	49	2.7 (49)	63	**	50	1.9 (50)	44	**
6	50	4.2 (50)	50	3.3 (50)	79	**	49	2.7 (49)	64	**	50	2.0 (50)	48	**
7	50	4.3 (50)	50	3.4 (50)	79	**	49	2.9 (49)	67	**	50	2.1 (50)	49	**
8	50	4.3 (50)	50	3.4 (50)	79	**	49	2.7 (49)	63	**	50	2.0 (50)	47	**
9	50	4.2 (50)	50	3.3 (50)	79	**	49	2.7 (49)	64	**	50	2.1 (50)	50	**
10	50	4.3 (50)	50	3.3 (50)	77	**	49	2.7 (49)	63	**	50	2.1 (50)	49	**
11	50	4.4 (50)	50	3.4 (50)	77	**	49	2.8 (49)	64	**	50	2.1 (50)	48	**
12	50	4.3 (50)	50	3.4 (50)	79	**	49	2.7 (49)	63	**	50	2.1 (50)	49	**
13	50	4.2 (50)	50	3.3 (50)	79	**	49	2.8 (49)	67	**	50	2.1 (50)	50	**
14	50	4.1 (50)	50	3.3 (50)	80	**	49	2.7 (49)	66	**	50	2.1 (50)	51	**
18	50	3.9 (50)	50	3.2 (50)	82	**	49	2.5 (49)	64	**	50	2.0 (50)	51	**
22	50	3.9 (50)	50	3.1 (50)	79	**	49	2.4 (49)	62	**	50	1.8 (50)	46	**
26	49	3.8 (49)	50	3.0 (50)	79	**	49	2.6 (49)	68	**	50	1.5 (49)	39	**
30	49	3.9 (49)	50	3.1 (50)	79	**	49	2.6 (49)	67	**	50	1.9 (50)	49	**
34	49	4.0 (49)	50	3.2 (50)	80	**	49	2.6 (49)	65	**	50	2.1 (50)	53	**
38	49	3.9 (49)	50	3.2 (50)	82	**	49	2.4 (49)	62	**	50	1.9 (50)	49	**
42	49	4.0 (49)	50	3.2 (50)	80	**	49	2.5 (49)	63	**	49	2.0 (49)	50	**
46	49	3.9 (49)	50	3.1 (50)	79	**	49	2.5 (49)	64	**	49	2.0 (49)	51	**
50	49	4.1 (49)	50	3.2 (50)	78	**	49	2.5 (49)	61	**	49	2.0 (49)	49	**
54	49	4.0 (49)	50	3.2 (50)	80	**	49	2.5 (49)	63	**	49	2.1 (49)	53	**
58	49	4.1 (49)	49	3.2 (49)	78	**	48	2.6 (48)	63	**	49	2.2 (49)	54	**
62	48	4.1 (48)	49	3.1 (49)	76	**	48	2.5 (48)	61	**	49	2.1 (49)	51	**
66	48	4.4 (48)	48	3.4 (48)	77	**	48	2.7 (48)	61	**	49	2.2 (49)	50	**
70	47	4.1 (47)	47	3.2 (47)	78	**	48	2.7 (47)	66	**	47	2.1 (47)	51	**
74	46	4.2 (46)	47	3.2 (47)	76	**	48	2.6 (48)	62	**	46	2.1 (46)	50	**
78	45	4.1 (45)	45	3.3 (45)	80	**	48	2.6 (48)	63	**	46	2.2 (46)	54	**
82	44	4.1 (44)	45	3.2 (45)	78	**	47	2.9 (47)	71	**	45	2.1 (45)	51	**
86	43	4.1 (43)	43	3.6 (43)	88	*	46	2.9 (45)	71	**	44	2.2 (44)	54	**
90	41	4.1 (41)	40	3.6 (40)	88	**	43	3.0 (42)	73	**	42	2.4 (42)	59	**
94	39	4.4 (39)	35	3.5 (34)	80	**	39	3.1 (39)	70	**	41	2.6 (41)	59	**
98	38	4.5 (38)	30	3.8 (30)	84	*	36	2.9 (36)	64	**	40	2.6 (40)	58	**
102	33	4.8 (33)	25	3.5 (25)	73	**	27	3.1 (27)	65	**	38	2.6 (38)	54	**
104	31	4.4 (31)	24	3.7 (24)	84		27	3.0 (27)	68	**	36	2.6 (36)	59	**

< > : No.of effective animals, ( ) : No.of measured animals % : % of control group
Significant Difference, * : p≦0.05, ** : p≦0.01, Test of Dunnett

TABLE 5 FOOD CONSUMPTION CHANGES OF MALE MICE
IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group	Control		750 ppm			1500 ppm			3000 ppm		
	<50>		<50>			<50>			<50>		
Week on Study	Survival No.	FC g	Survival No.	FC g	%	Survival No.	FC g	%	Survival No.	FC g	%
1	50	3.8 (50)	50	3.7 (50)	97	50	3.7 (50)	97 *	50	3.5 (50)	92 **
2	50	3.7 (50)	50	3.6 (50)	97	50	3.6 (50)	97	50	3.5 (50)	95 **
3	50	3.8 (50)	49	3.8 (49)	100	50	3.6 (50)	95	50	3.4 (50)	89 **
4	50	3.8 (50)	49	3.8 (49)	100	50	3.7 (50)	97	50	3.6 (50)	95 **
5	50	4.0 (50)	49	4.0 (49)	100	50	3.6 (50)	90 **	50	3.5 (49)	88 **
6	50	3.9 (50)	49	3.7 (49)	95 *	50	3.5 (50)	90 **	50	3.4 (50)	87 **
7	50	3.9 (50)	49	3.8 (49)	97	50	3.7 (50)	95 *	50	3.5 (50)	90 **
8	50	4.1 (50)	49	4.0 (48)	98	50	3.8 (50)	93 **	50	3.6 (50)	88 **
9	50	4.0 (50)	49	3.9 (49)	98	50	3.8 (50)	95	50	3.6 (50)	90 **
10	50	4.1 (50)	49	4.2 (49)	102	50	3.9 (50)	95 **	50	3.7 (50)	90 **
11	50	4.1 (50)	49	4.0 (49)	98	49	3.9 (48)	95 **	50	3.7 (50)	90 **
12	50	4.1 (50)	49	4.0 (49)	98	49	3.9 (49)	95 **	50	3.6 (50)	88 **
13	50	4.0 (50)	49	4.1 (49)	103	49	4.0 (49)	100	50	3.8 (50)	95 **
14	50	4.2 (50)	49	4.1 (49)	98	49	3.9 (49)	93 **	50	3.7 (50)	88 **
18	50	4.1 (50)	49	4.1 (49)	100	49	3.9 (49)	95 **	50	3.7 (50)	90 **
22	50	4.2 (50)	49	4.1 (49)	98	49	3.9 (49)	93 **	50	3.9 (50)	93 **
26	50	4.3 (50)	49	4.2 (49)	98	49	4.1 (49)	95 *	50	3.9 (50)	91 **
30	50	4.4 (50)	49	4.4 (49)	100	49	4.1 (49)	93 **	50	3.9 (50)	89 **
34	50	4.4 (50)	49	4.4 (49)	100	49	4.2 (49)	95 **	50	4.0 (50)	91 **
38	50	4.5 (50)	49	4.4 (49)	98	49	4.3 (49)	96 **	50	4.0 (50)	89 **
42	50	4.5 (50)	49	4.5 (48)	100	49	4.4 (49)	98	50	4.2 (50)	93 **
46	50	4.6 (50)	49	4.6 (49)	100	49	4.4 (49)	96 **	50	4.2 (50)	91 **
50	50	4.5 (50)	49	4.5 (49)	100	49	4.3 (49)	96 **	50	4.2 (50)	93 **
54	50	4.6 (50)	49	4.5 (49)	98	49	4.2 (49)	91 **	49	4.1 (49)	89 **
58	49	4.6 (49)	49	4.5 (49)	98	49	4.1 (49)	89 **	49	4.1 (49)	89 **
62	49	4.7 (49)	48	4.6 (48)	98	49	4.4 (49)	94 **	49	4.3 (49)	91 **
66	49	4.9 (49)	47	4.7 (47)	96	49	4.4 (49)	90 **	49	4.2 (49)	86 **
70	49	4.8 (49)	47	4.7 (47)	98	48	4.4 (48)	92 **	47	4.2 (47)	88 **
74	49	4.8 (49)	47	4.7 (47)	98	47	4.4 (47)	92 **	46	4.3 (46)	90 **
78	47	4.8 (47)	46	4.7 (46)	98	46	4.4 (46)	92 **	45	4.2 (45)	88 **
82	46	4.9 (46)	46	4.8 (46)	98	46	4.5 (46)	92 **	44	4.3 (44)	88 **
86	46	4.8 (46)	44	4.6 (44)	96	45	4.3 (45)	90 **	44	4.2 (44)	88 **
90	45	5.0 (45)	43	5.0 (43)	100	44	4.6 (44)	92 **	44	4.4 (44)	88 **
94	43	5.1 (43)	40	5.0 (40)	98	43	4.6 (43)	90 **	44	4.4 (44)	86 **
98	43	4.9 (43)	36	4.9 (36)	100	41	4.6 (41)	94 **	44	4.4 (44)	90 **
102	38	5.1 (38)	36	4.8 (36)	94	41	4.7 (41)	92 **	44	4.5 (44)	88 **
104	37	4.9 (37)	36	4.7 (36)	96	38	4.5 (38)	92 **	43	4.2 (42)	86 **

< > : No.of effective animals, ( ) : No.of measured animals % : % of control group
Significant Difference, * : $p \leqq 0.05$, ** : $p \leqq 0.01$, Test of Dunnett

TABLE 6 FOOD CONSUMPTION CHANGES OF FEMALE MICE
IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group	Control		500 ppm			1500 ppm			4500 ppm		
	<50>		<50>			<49>			<50>		
Week on Study	Survival No.	FC g	Survival No.	FC g	%	Survival No.	FC g	%	Survival No.	FC g	%
1	50	3.3 (50)	50	3.3 (50)	100	49	3.3 (49)	100	50	3.0 (50)	91 **
2	50	3.3 (50)	50	3.4 (50)	103	49	3.2 (49)	97	50	3.1 (50)	94 **
3	50	3.4 (50)	50	3.3 (50)	97	49	3.3 (49)	97	50	3.2 (50)	94 **
4	50	3.6 (50)	50	3.5 (50)	97	49	3.4 (49)	94 *	50	3.2 (50)	89 **
5	50	3.5 (50)	50	3.5 (50)	100	49	3.4 (49)	97	50	3.2 (50)	91 **
6	50	3.4 (50)	50	3.4 (50)	100	49	3.4 (49)	100	50	3.3 (49)	97 *
7	50	3.6 (50)	50	3.6 (50)	100	49	3.5 (49)	97 *	50	3.4 (50)	94 **
8	50	3.7 (50)	50	3.7 (50)	100	49	3.6 (49)	97 *	50	3.5 (50)	95 **
9	50	3.8 (50)	50	3.6 (50)	95 *	49	3.6 (49)	95 *	50	3.5 (50)	92 **
10	50	3.8 (50)	50	3.7 (50)	97	49	3.7 (49)	97	50	3.5 (50)	92 **
11	50	3.8 (50)	50	3.7 (50)	97	49	3.7 (49)	97 *	50	3.6 (50)	95 **
12	50	3.8 (50)	50	3.7 (50)	97	49	3.6 (49)	95 *	50	3.5 (50)	92 **
13	50	3.9 (50)	50	3.8 (50)	97	49	3.7 (49)	95 *	50	3.6 (50)	92 **
14	50	3.8 (50)	50	3.8 (50)	100	49	3.7 (49)	97	50	3.5 (50)	92 **
18	50	3.9 (50)	50	3.7 (50)	95 *	49	3.6 (49)	92 **	50	3.4 (50)	87 **
22	50	3.9 (50)	50	3.8 (50)	97	49	3.8 (49)	97	50	3.4 (50)	87 **
26	49	3.9 (49)	50	3.9 (50)	100	49	3.9 (49)	100	50	3.6 (50)	92 **
30	49	4.0 (49)	50	3.9 (50)	98	49	3.8 (49)	95 **	50	3.5 (50)	88 **
34	49	4.0 (49)	50	4.1 (50)	103	49	3.9 (49)	98	50	3.7 (50)	92 **
38	49	4.1 (49)	50	4.1 (50)	100	49	3.9 (49)	95	50	3.8 (50)	93 **
42	49	3.9 (49)	50	3.8 (50)	97	49	3.7 (49)	95 *	49	3.6 (49)	92 **
46	49	4.0 (49)	50	4.0 (50)	100	49	3.9 (49)	98	49	3.6 (48)	90 **
50	49	4.2 (49)	50	4.0 (50)	95	49	3.9 (49)	93 *	49	3.7 (49)	88 **
54	49	4.1 (49)	50	4.0 (50)	98	49	3.7 (49)	90 **	49	3.6 (49)	88 **
58	49	4.0 (49)	49	4.0 (49)	100	48	3.8 (48)	95 *	49	3.6 (49)	90 **
62	48	4.4 (48)	49	4.0 (49)	91 **	48	4.0 (48)	91 **	49	3.8 (49)	86 **
66	48	4.4 (48)	48	4.2 (48)	95	48	4.0 (48)	91 **	49	3.7 (49)	84 **
70	47	4.2 (47)	47	3.9 (47)	93 *	48	4.0 (48)	95	47	3.8 (47)	90 **
74	46	4.4 (46)	47	4.1 (47)	93 **	48	3.8 (48)	86 **	46	3.6 (46)	82 **
78	45	4.3 (45)	45	4.2 (45)	98	48	4.0 (48)	93 **	46	3.7 (46)	86 **
82	44	4.4 (44)	45	4.0 (45)	91 *	47	3.8 (47)	86 **	45	3.7 (45)	84 **
86	43	4.2 (43)	43	4.3 (43)	102	46	4.0 (46)	95	44	3.7 (44)	88 **
90	41	4.5 (41)	40	4.4 (40)	98	43	4.2 (43)	93	42	3.9 (42)	87 **
94	39	4.8 (39)	35	4.5 (35)	94	39	4.3 (39)	90 **	41	4.2 (41)	88 **
98	38	4.5 (38)	30	4.3 (30)	96	36	4.0 (36)	89 **	40	4.0 (40)	89 **
102	33	4.4 (33)	25	4.4 (25)	100	27	4.2 (27)	95	38	4.0 (38)	91 *
104	31	4.5 (31)	24	4.5 (24)	100	27	4.0 (27)	89 *	36	3.8 (36)	84 **

< > : No.of effective animals, ( ) : No.of measured animals % : % of control group
Significant Difference, * : $p \leqq 0.05$, ** : $p \leqq 0.01$, Test of Dunnett

TABLE 7 INCIDENCE AND TIME OF MASS OCCURRENCE IN CLINICAL OBSERVATION OF MALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Time of mass occurrence (week)	0～13	14～26	27～39	40～52	53～65	66～78	79～91	92～104	0～104
External mass									
Control	0/50	0/50	0/50	0/50	1/50	1/49	2/47	3/45	3/50 (2/13)
750 ppm	0/50	0/49	0/49	0/49	1/49	1/47	1/46	1/40	3/50 (2/14)
1500 ppm	0/50	0/49	1/49	1/49	1/49	1/49	1/46	3/44	3/50 (2/12)
3000 ppm	0/50	0/50	0/50	0/50	1/50	1/49	0/45	0/44	1/50 (1/ 7)
Internal mass									
Control	0/50	1/50	1/50	1/50	1/50	4/49	5/47	8/45	13/50 (5/13)
750 ppm	0/50	1/49	1/49	2/49	2/49	4/47	7/46	4/40	10/50 (8/14)
1500 ppm	1/50	2/49	1/49	1/49	2/49	3/49	3/46	8/44	12/50 (4/12)
3000 ppm	0/50	0/50	0/50	0/50	0/50	0/49	0/45	2/44	2/50 (0/ 7)

No. of animals with mass / No. of survival animals at first week on each period.
(No. of dead and moribund animals with mass / No. of dead and moribund animals)

TABLE 8 INCIDENCE AND TIME OF MASS OCCURRENCE IN CLINICAL OBSERVATION OF FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Time of mass occurrence (week)	0～13	14～26	27～39	40～52	53～65	66～78	79～91	92～104	0～104
External mass									
Control	0/50	0/50	0/49	0/49	0/49	0/48	2/45	4/41	4/50 (3/19)
500 ppm	0/50	0/50	0/50	0/50	1/50	2/48	1/45	3/37	5/50 (4/26)
1500 ppm	0/49	0/49	0/49	0/49	0/49	1/48	2/48	3/41	3/49 (1/23)
4500 ppm	0/50	0/50	0/50	0/49	0/49	2/49	1/45	3/41	4/50 (3/14)
Internal mass									
Control	0/50	0/50	0/49	0/49	2/49	4/48	2/45	8/41	12/50 (6/19)
500 ppm	0/50	0/50	0/50	0/50	1/50	4/48	8/45	9/37	16/50 (12/26)
1500 ppm	0/49	0/49	1/49	1/49	1/49	2/48	9/48	13/41	17/49 (13/23)
4500 ppm	0/50	0/50	0/50	0/49	0/49	4/49	4/45	5/41	10/50 (6/14)

No. of animals with mass / No. of survival animals at first week on each period.
(No. of dead and moribund animals with mass / No. of dead and moribund animals)

TABLE 9 HEMATOLOGY OF MALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE (SELECTED)

Group	Control	750 ppm	1500 ppm	3000 ppm	
No. of animals examined	34	31	35	40	
Red blood cell ($10^6/\mu$ L)	9.55 ± 0.81	9.67 ± 1.37	9.83 ± 0.86	9.52 ± 0.79	
Hemoglobin (g/dL)	13.6 ± 1.2	13.6 ± 1.8	13.7 ± 1.0	13.5 ± 1.1	
Hematocrit (%)	43.8 ± 3.5	44.2 ± 5.4	44.4 ± 3.3	44.0 ± 3.4	
MCV (fL)	45.9 ± 1.5	46.0 ± 3.0	45.2 ± 1.6	46.3 ± 1.1	
MCH (pg)	14.2 ± 0.5	14.1 ± 0.6	14.0 ± 0.6	14.2 ± 0.4	
MCHC (g/dL)	30.9 ± 0.7	30.8 ± 1.0	30.9 ± 0.6	30.7 ± 0.6	
Platelet ($10^3/\mu$ L)	1817 ± 347	1856 ± 501	1830 ± 302	1941 ± 218	
WBC ($10^3/\mu$ L)	2.94 ± 1.51	2.88 ± 1.60	2.25 ± 1.00	2.01 ± 2.56	**

Data represent means ± S.D.

Significant difference, ** : $p \leqq 0.01$, Test of Dunnett

TABLE 10 HEMATOLOGY OF FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE (SELECTED)

Group	Control	500 ppm	1500 ppm	4500 ppm	
No. of animals examined	28	24	24	33	
Red blood cell ($10^6/\mu$ L)	9.42 ± 1.83	9.04 ± 1.66	9.40 ± 1.45	9.23 ± 1.51	
Hemoglobin (g/dL)	13.7 ± 2.4	12.9 ± 2.3	13.3 ± 2.1	12.9 ± 2.1	
Hematocrit (%)	43.7 ± 7.4	42.0 ± 6.8	43.1 ± 5.4	42.2 ± 6.3	
MCV (fL)	46.9 ± 4.2	46.8 ± 3.5	46.3 ± 3.5	46.0 ± 2.3	
MCH (pg)	14.6 ± 0.9	14.4 ± 0.7	14.1 ± 0.5	14.0 ± 0.5	**
MCHC (g/dL)	31.2 ± 1.1	30.8 ± 1.1	30.7 ± 1.9	30.5 ± 1.1	**
Platelet ($10^3/\mu$ L)	1075 ± 304	1111 ± 293	1180 ± 313	1336 ± 241	**
WBC ($10^3/\mu$ L)	3.99 ± 9.86	2.00 ± 1.22	1.45 ± 0.72	3.25 ± 10.52	

Data represent means ± S.D.

Significant difference, ** : $p \leqq 0.01$, Test of Dunnett

TABLE 11 BIOCHEMISTRY OF MALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group	Control	750 ppm	1500 ppm	3000 ppm
No. of animals examined	35	33	36	41
Total protein (g/dL)	5.1 ± 0.5	5.3 ± 0.7	5.1 ± 0.5	4.9 ± 0.4 *
Albumin (g/dL)	2.8 ± 0.3	2.9 ± 0.5	2.8 ± 0.3	2.8 ± 0.2
A/G ratio	1.2 ± 0.1	1.2 ± 0.2	1.2 ± 0.2	1.3 ± 0.1 **
T-Bilirubin (mg/dL)	0.13 ± 0.03	0.13 ± 0.03	0.13 ± 0.03	0.13 ± 0.02
Glucose (mg/dL)	204 ± 36	186 ± 40	188 ± 43	204 ± 22
T-Cholesterol (mg/dL)	110 ± 28	122 ± 49	109 ± 44	102 ± 20
Triglyceride (mg/dL)	42 ± 18	48 ± 81	32 ± 13 *	31 ± 11 *
Phospholipid (mg/dL)	201 ± 42	218 ± 73	196 ± 48	198 ± 30
GOT (IU/L)	112 ± 253	246 ± 768 *	102 ± 158	52 ± 11
GPT (IU/L)	54 ± 118	166 ± 536	65 ± 128	22 ± 10 **
LDH (IU/L)	553 ± 1049	770 ± 1585	450 ± 555	265 ± 87
ALP (IU/L)	129 ± 44	138 ± 74	139 ± 85	130 ± 19
γ -GTP (IU/L)	2 ± 1	2 ± 1	2 ± 2	2 ± 3
CPK (IU/L)	52 ± 22	63 ± 33	59 ± 36	61 ± 29
Urea nitrogen (mg/L)	21.8 ± 2.5	23.6 ± 5.8	24.2 ± 11.1	21.9 ± 3.8
Sodium (mEq/L)	153 ± 1	154 ± 1	154 ± 1	154 ± 2
Potassium (mEq/L)	4.4 ± 0.4	4.3 ± 0.5	4.1 ± 0.7 **	4.1 ± 0.5 **
Chloride (mEq/L)	122 ± 3	122 ± 3	122 ± 3	121 ± 3
Calcium (mg/dL)	8.8 ± 0.4	8.9 ± 0.6	8.7 ± 0.4	8.6 ± 0.3
Inorganic phosphorus (mg/dL)	6.6 ± 0.8	6.5 ± 0.8	6.7 ± 1.0	6.3 ± 0.8

Data represent means ± S.D.
Significant difference, * : $p \leqq 0.05$, ** : $p \leqq 0.01$, Test of Dunnett

TABLE 12 BIOCHEMISTRY OF FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group	Control	500 ppm	1500 ppm	4500 ppm
No. of animals examined	30	24	26	34
Total protein (g/dL)	5.2 ± 1.1	5.2 ± 0.7	4.9 ± 0.3	4.7 ± 0.4 *
Albumin (g/dL)	2.8 ± 0.4	2.9 ± 0.5	2.8 ± 0.2	2.8 ± 0.3
A/G ratio	1.2 ± 0.3	1.3 ± 0.2	1.4 ± 0.2 *	1.5 ± 0.2 **
T-Bilirubin (mg/dL)	0.14 ± 0.03	0.15 ± 0.07	0.13 ± 0.03	0.12 ± 0.02
Glucose (mg/dL)	130 ± 41	149 ± 37	147 ± 22	157 ± 31 **
T-Cholesterol (mg/dL)	78 ± 36	98 ± 57	75 ± 15	92 ± 16 **
Triglyceride (mg/dL)	30 ± 17	29 ± 15	25 ± 14	19 ± 11 **
Phospholipid (mg/dL)	141 ± 55	182 ± 104	145 ± 26	172 ± 24 **
GOT (IU/L)	96 ± 52	84 ± 33	76 ± 23	85 ± 114 **
GPT (IU/L)	43 ± 34	40 ± 29	28 ± 10	28 ± 18 **
LDH (IU/L)	548 ± 559	403 ± 524	462 ± 752	495 ± 1365 **
ALP (IU/L)	191 ± 82	203 ± 119	310 ± 258 **	255 ± 94 **
γ -GTP (IU/L)	2 ± 1	2 ± 2	2 ± 2	2 ± 2
CPK (IU/L)	99 ± 77	75 ± 71	77 ± 35	89 ± 77
Urea nitrogen (mg/L)	23.3 ± 14.1	19.9 ± 8.4	18.8 ± 4.5	21.9 ± 11.5
Sodium (mEq/L)	154 ± 4	152 ± 2	154 ± 2	153 ± 2
Potassium (mEq/L)	4.3 ± 0.8	4.0 ± 0.5	4.1 ± 0.4	4.2 ± 0.4
Chloride (mEq/L)	123 ± 4	122 ± 3	123 ± 2	121 ± 3 *
Calcium (mg/dL)	8.8 ± 0.5	9.2 ± 0.9	8.9 ± 0.4	8.8 ± 0.3
Inorganic phosphorus (mg/dL)	6.8 ± 1.4	6.4 ± 0.7	6.8 ± 0.9	5.7 ± 1.1 **

Data represent means ± S.D.
Significant difference, * : $p \leqq 0.05$, ** : $p \leqq 0.01$, Test of Dunnett

TABLE 13 URINALYSIS OF MALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE (SELECTED)

Group		Control	750 ppm	1500 ppm	3000 ppm
No. of animals examined		37	36	39	43
pH	6.0	3	6 **	4 **	10 **
	6.5	6	19	32	33
	7.0	16	9	2	0
	7.5	12	2	0	0
	8.0	0	0	1	0
	8.5	0	0	0	0
	(Grade)				
Protein	−	0	0 *	0 **	0 **
	±	0	3	0	0
	+	31	21	17	14
	2+	3	11	19	28
	3+	2	0	2	1
	4+	1	1	1	0
Ketone body	−	29	13 **	11 **	10 **
	±	7	14	13	20
	+	1	8	12	11
	2+	0	1	2	2
	3+	0	0	1	0
Occult blood	−	28	29	37	39
	±	3	1	0	1
	+	1	1	0	0
	2+	1	0	0	0
	3+	4	5	2	3

Significant difference, * : p≦0.05, ** : p≦0.01 Chi square test

TABLE 14 URINALYSIS OF FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE (SELECTED)

Group		Control	500 ppm	1500 ppm	4500 ppm
No. of animals examined		31	24	27	36
pH	6.0	1	0	1 **	5 **
	6.5	5	3	9	23
	7.0	1	6	7	4
	7.5	3	4	6	3
	8.0	19	11	4	1
	8.5	2	0	0	0
	(Grade)				
Protein	−	0	0	0	0 **
	±	0	0	0	0
	+	17	9	13	7
	2+	13	15	12	25
	3+	1	0	2	4
	4+	0	0	0	0
Ketone body	−	9	1	0 **	4 **
	±	18	20	11	14
	+	4	3	10	11
	2+	0	0	6	7
	3+	0	0	0	0
Occult blood	−	24	20	22	19
	±	4	3	0	5
	+	0	0	1	1
	2+	0	0	3	3
	3+	3	1	1	8

Significant difference, * : p≦0.05, ** : p≦0.01 Chi square test

TABLE 15 ORGAN WEIGHTS OF MALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group		Control	750 ppm	1500 ppm	3000 ppm
No. of animals examined		<37>	<36>	<38>	<43>
Adrenal	(g)	0.010 ± 0.002	0.009 ± 0.002	0.010 ± 0.002	0.009 ± 0.002
	(%)	0.021 ± 0.006	0.022 ± 0.006	0.027 ± 0.008 **	0.027 ± 0.007 **
Testis	(g)	0.207 ± 0.036	0.227 ± 0.040 *	0.201 ± 0.027	0.214 ± 0.030
	(%)	0.445 ± 0.099	0.547 ± 0.124 **	0.549 ± 0.107 **	0.624 ± 0.101 **
Heart	(g)	0.227 ± 0.030	0.220 ± 0.027	0.203 ± 0.025 **	0.201 ± 0.018 **
	(%)	0.482 ± 0.057	0.527 ± 0.076 *	0.552 ± 0.080 **	0.585 ± 0.062 **
Lung	(g)	0.225 ± 0.026	0.246 ± 0.150	0.216 ± 0.040	0.202 ± 0.017 **
	(%)	0.481 ± 0.078	0.635 ± 0.676	0.591 ± 0.126 **	0.592 ± 0.084 **
Kidney	(g)	0.616 ± 0.048	1.261 ± 3.470	0.690 ± 0.340	0.636 ± 0.053
	(%)	1.313 ± 0.140	2.772 ± 6.753 **	1.883 ± 0.923 **	1.853 ± 0.186 **
Spleen	(g)	0.134 ± 0.219	0.115 ± 0.116	0.108 ± 0.139	0.078 ± 0.074 **
	(%)	0.317 ± 0.623	0.228 ± 0.313	0.298 ± 0.402	0.227 ± 0.220
Liver	(g)	1.799 ± 0.863	1.805 ± 0.509	1.551 ± 0.340 *	1.447 ± 0.205 **
	(%)	3.866 ± 2.147	4.346 ± 1.458 **	4.240 ± 1.212 **	4.222 ± 0.718 **
Brain	(g)	0.448 ± 0.018	0.448 ± 0.014	0.446 ± 0.014	0.448 ± 0.015
	(%)	0.960 ± 0.126	1.082 ± 0.187 **	1.222 ± 0.169 **	1.314 ± 0.179 **

Data represent means ± S.D.
Significant difference, * : $p \leqq 0.05$, ** : $p \leqq 0.01$, Test of Dunnett

TABLE 16 ORGAN WEIGHTS OF FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group		Control	500 ppm	1500 ppm	4500 ppm
No. of animals examined		<31>	<24>	<27>	<36>
Adrenal	(g)	0.013 ± 0.002	0.015 ± 0.007	0.012 ± 0.003	0.012 ± 0.002
	(%)	0.040 ± 0.007	0.050 ± 0.026	0.045 ± 0.012	0.048 ± 0.009 **
Ovary	(g)	0.052 ± 0.052	0.142 ± 0.492	0.335 ± 1.196	0.067 ± 0.134
	(%)	0.162 ± 0.152	0.441 ± 1.476	1.088 ± 3.752	0.268 ± 0.543
Heart	(g)	0.176 ± 0.024	0.179 ± 0.025	0.159 ± 0.014 *	0.155 ± 0.021 **
	(%)	0.560 ± 0.089	0.605 ± 0.122	0.595 ± 0.067	0.632 ± 0.110 **
Lung	(g)	0.226 ± 0.049	0.209 ± 0.029	0.202 ± 0.021	0.192 ± 0.035 **
	(%)	0.720 ± 0.154	0.708 ± 0.135	0.756 ± 0.106	0.784 ± 0.195
Kidney	(g)	0.431 ± 0.065	0.463 ± 0.043 *	0.473 ± 0.117	0.531 ± 0.243 **
	(%)	1.370 ± 0.200	1.560 ± 0.218 **	1.771 ± 0.481 **	2.151 ± 0.953 **
Spleen	(g)	0.196 ± 0.156	0.220 ± 0.248	0.158 ± 0.160	0.179 ± 0.236
	(%)	0.624 ± 0.481	0.711 ± 0.739	0.573 ± 0.514	0.727 ± 0.957
Liver	(g)	1.754 ± 1.036	1.579 ± 0.514	1.470 ± 0.859 *	1.275 ± 0.235 **
	(%)	5.505 ± 2.942	5.270 ± 1.511	5.389 ± 2.637	5.166 ± 0.885 **
Brain	(g)	0.471 ± 0.020	0.470 ± 0.023	0.471 ± 0.017	0.453 ± 0.015 **
	(%)	1.508 ± 0.197	1.597 ± 0.286	1.767 ± 0.210 **	1.845 ± 0.155 **

Data represent means ± S.D.
Significant difference, * : $p \leqq 0.05$, ** : $p \leqq 0.01$, Test of Dunnett

TABLE 17 NEOPLASTIC LESIONS OF MALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE (SELECTED)

Group No. of animals examined	Control <50>	750 ppm <50>	1500 ppm <50>	3000 ppm <50>	
Lung					
Bronchiolar-alveolar adenoma	5 (10%) [a)]	6 (12%)	4 (8%)	3 (6%)	
Bronchiolar-alveolar carcinoma	4 (8%)	3 (6%)	5 (10%)	1 (2%)	
Lymph node					
Malignant lymphoma	7 (14%)	11 (22%)	9 (18%)	6 (12%)	
Spleen					
Malignant lymphoma	2 (4%)	3 (6%)	2 (4%)	1 (2%)	
Hemangioma	3 (6%)	1 (2%)	1 (2%)	0 (0%)	
Hemangiosarcoma	2 (4%)	1 (2%)	1 (2%)	0 (0%)	
Hemangioma / hemangiosarcoma	5 (10%)	2 (4%)	2 (4%)	0 (0%) *	⇩
Liver					
Hepatocellular adenoma	7 (14%)	8 (16%)	5 (10%)	3 (6%)	
Hepatocellular carcinoma	6 (12%)	7 (14%)	4 (8%)	0 (0%) *	⇩
Hepatocellular adenoma / carcinoma	11 (22%)	15 (30%)	9 (18%)	3 (6%) *	⇩⇩
Hemangioma	1 (2%)	0 (0%)	0 (0%)	0 (0%)	
Hemangiosarcoma	7 (14%)	3 (6%)	3 (6%)	0 (0%) **	⇩⇩
Hemangioma / hemangiosarcoma	8 (16%)	3 (6%)	3 (6%)	0 (0%) **	⇩⇩
Histiocytic sarcoma	3 (6%)	1 (2%)	0 (0%)	2 (4%)	
Stomach					
Squamous cell carcinoma	0 (0%)	0 (0%)	1 (2%)	0 (0%)	
Multi-site					
Histiocytic sarcoma	5 (10%)	2 (4%)	0 (0%) *	6 (12%)	
Malignant lymphoma	9 (18%)	14 (28%)	11 (22%)	7 (14%)	

a) : No. of animals with bearing tumor (incidence ; %)

* and ** : Statistically differenct from control group at $p \leqq 0.05$ and $p \leqq 0.01$ by Fisher exact test, respectively

⇩ and ⇩⇩ : The trend of treated groups statistically different from control group at $p \leqq 0.05$ and $p \leqq 0.01$ by Cochran-Armitage test, respectively.

TABLE 18 NEOPLASTIC LESIONS OF FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE (SELECTED)

Group No. of animals examined	Control <50>	500 ppm <50>	1500 ppm <49>	4500 ppm <50>	
Lung					
Bronchiolar-alveolar adenoma	0 (0%) [a)]	1 (2%)	1 (2%)	0 (0%)	
Bronchiolar-alveolar carcinoma	0 (0%)	0 (0%)	0 (0%)	2 (4%)	
Lymph node					
Malignant lymphoma	18 (36%)	17 (34%)	9 (18%) *	9 (18%) *	⇩
Spleen					
Malignant lymphoma	4 (8%)	2 (4%)	1 (2%)	3 (6%)	
Liver					
Hepatocellular adenoma	0 (0%)	6 (12%) *	1 (2%)	2 (4%)	
Hepatocellular carcinoma	1 (2%)	2 (4%)	0 (0%)	0 (0%)	
Hepatocellular adenoma / carcinoma	1 (2%)	8 (16%) *	1 (2%)	2 (4%)	
Kidney					
Renal cell carcinoma	0 (0%)	2 (4%)	0 (0%)	0 (0%)	
Stomach					
Squamous cell papilloma	0 (0%)	1 (2%)	0 (0%)	0 (0%)	
Pituitary					
Adenoma	5 (10%)	6 (12%)	1 (2%)	1 (2%)	⇩
Adenocarcinoma	0 (0%)	1 (2%)	1 (2%)	0 (0%)	
Adenoma / adenocarcinoma	5 (10%)	7 (14%)	2 (4%)	1 (2%)	⇩
Uterus					
Endometrial stromal polyp	1 (2%)	0 (0%)	3 (6%)	2 (4%)	
Histiocytic sarcoma	5 (10%)	6 (12%)	12 (24%) *	9 (18%)	
Multi-site					
Histiocytic sarcoma	6 (12%)	8 (16%)	13 (26%)	10 (20%)	
Malignant lymphoma	22 (44%)	19 (38%)	10 (20%) *	12 (24%) *	⇩

[a)] : No. of animals with tumor (incidence ; %)

* : Statistically differenct from control group at $p \leqq 0.05$ by Fisher exact test

⇩ : The trend of treated groups statistically different from control group at $p \leqq 0.05$ by Cochran-Armitage test

TABLE 19 NON-NEOPLASTIC LESIONS IN MALE AND FEMALE MICE ON THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE (SELECTED)

		Male				Female			
Group		Control	750 ppm	1500 ppm	3000 ppm	Control	500 ppm	1500 ppm	4500 ppm
No. of animals examined		<50>	<50>	<50>	<50>	<50>	<50>	<49>	<50>
Nasal cavity	Grade								
Respiratory metaplasia : gland	+	1	10	14	10	3	5	12	14
	2+	2	1	3	0	0	0	0	0
Stomach									
Erosion : forestomach	+	0	0	0	0	0	1	0	4
Ulcer : forestomach	+	1	0	0	3	0	0	0	0
Squamous cell hyperplasia :	+	2	0	1	4	5	7	13	10
forestomach	2+	0	0	0	4	0	2	2	33
	3+	0	0	0	0	0	0	0	2
Hyperplasia : glandular stomach	+	13	9	11	2	0	8	2	0
Mineralization : glandular stomach	+	7	3	3	0	3	7	1	0
Liver									
Clear cell focus	+	4	0	2	0	1	0	1	3
	2+	0	2	1	0	1	0	0	0
Kidney									
Desquamation : pelvis	+	0	0	1	1	2	1	10	4
	2+	0	1	0	2	1	3	4	3
Mineralization : cortex	+	15	5	4	7	0	0	0	1
Testis									
Mineralization	+	16	16	8	7	-	-	-	-
	2+	3	2	0	2	-	-	-	-
Uterus									
Cystic endometrial hyperplasia	+	-	-	-	-	24	23	18	21
	2+	-	-	-	-	6	0	5	8
Brain									
Mineralization	+	37	29	19	21	15	11	15	18
	2+	0	0	0	1	0	0	0	0

Grade + : Slight 2+ : Moderate 3+ : Marked

TABLE 20 CAUSE OF DEATH OF MALE AND FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

Group	Male				Female			
	Control	750 ppm	1500 ppm	3000 ppm	Control	500 ppm	1500 ppm	4500 ppm
No. of dead/moribund animals	<13>	<14>	<12>	<7>	<19>	<26>	<22>	<14>
Hydronephrosis	0	2	2	1	1	1	1	0
Urinary retention	1	0	0	0	0	0	1	0
Urinary system lesions	0	1	0	0	0	0	0	0
Arteritis	0	0	1	0	0	0	0	1
Amyloidosis	0	0	0	0	0	1	0	0
Peritonitis	0	0	0	0	0	0	1	0
Tumor death :								
leukemia	3	6	4	1	11	11	7	3
subcutis	0	0	1	1	0	0	0	1
lung	2	0	3	0	0	0	0	0
liver	6	3	0	1	1	3	2	0
kidney	0	0	0	0	0	2	0	0
pituitary	0	0	1	0	1	1	1	0
ovary	-	-	-	-	0	1	0	0
uterus	-	-	-	-	2	4	7	8
mammary gland	0	0	0	0	2	0	0	0
epididymis	1	1	0	2	-	-	-	-
No microscopical confirmation	0	1	0	1	1	2	2	1

TABLE 21 HISTORICAL CONTROL DATA OF SELECTED NEOPLASTIC LESIONS IN JAPAN BIOASSAY RESEARCH CENTER : Crj: BDF$_1$ MALE MICE

Organs / Tumors	No. of animals examined	No. of animals with bearing tumor	Incidence (%)	Min. - Max. (%)
Lung	<1046>			
Bronchiolar-alveolar adenoma		74	7.1	2 - 18
Bronchiolar-alveolar carcinoma		120	11.5	0 - 24
Lymph node	<1047>			
Malignant lymphoma		111	10.6	2 - 22
Spleen	<1046>			
Malignant lymphoma		43	4.1	0 - 8
Hemangioma		18	1.7	0 - 10
Hemangiosarcoma		26	2.5	0 - 6
Liver	<1047>			
Hepatocellular adenoma		179	17.1	4 - 34
Hepatocellular carcinoma		224	21.4	2 - 42
Hemangioma		14	1.3	0 - 10
Hemangiosarcoma		42	4.0	0 - 12
Histiocytic sarcoma		30	2.9	0 - 8
Stomach	<1046>			
Squamous cell carcinoma		1	0.1	0 - 2

21 carcinogenicity studies examined in Japan Bioassay Research Center were used.
Study No. 0044, 0060, 0062, 0064, 0066, 0068, 0096, 0105, 0116, 0140, 0159, 0163, 0190, 0206, 0211, 0225, 0243, 0270, 0285, 0297, 0319

TABLE 22 HISTORICAL CONTROL DATA OF SELECTED NEOPLASTIC LESIONS IN JAPAN BIOASSAY RESEARCH CENTER : Crj: BDF_1 FEMALE MICE

Organs / Tumors	No. of animals examined	No. of animals with bearing tumor	Incidence (%)	Min. - Max. (%)
Lung	<1048>			
Bronchiolar-alveolar adenoma		42	4.0	0 - 10
Bronchiolar-alveolar carcinoma		32	3.1	0 - 8
Lymph node	<1048>			
Malignant lymphoma		277	26.4	12 - 44
Spleen	<1048>			
Malignant lymphoma		77	7.3	0 - 26
Liver	<1048>			
Hepatocellular adenoma		54	5.2	2 - 10
Hepatocellular carcinoma		26	2.5	0 - 8
Kidney	<1048>			
Renal cell carcinoma		0	0	0
Stomach	<1047>			
Squamous cell papiloma		4	0.4	0 - 2
Pituitary	<1042>			
Adenoma		159	15.3	2 - 34
Adenocarcinoma		6	0.6	0 - 4
Uterus	<1046>			
Endometria stromal polyp		30	2.9	0 - 10
Histiocytic sarcoma		207	19.8	10 - 30

21 carcinogenicity studies examined in Japan Bioassay Research Center were used.
Study No. 0044, 0060, 0062, 0064, 0066, 0068, 0096, 0105, 0116, 0140, 0159, 0163, 0190, 0206, 0211, 0225, 0243, 0270, 0285, 0297, 0319

FIGURES

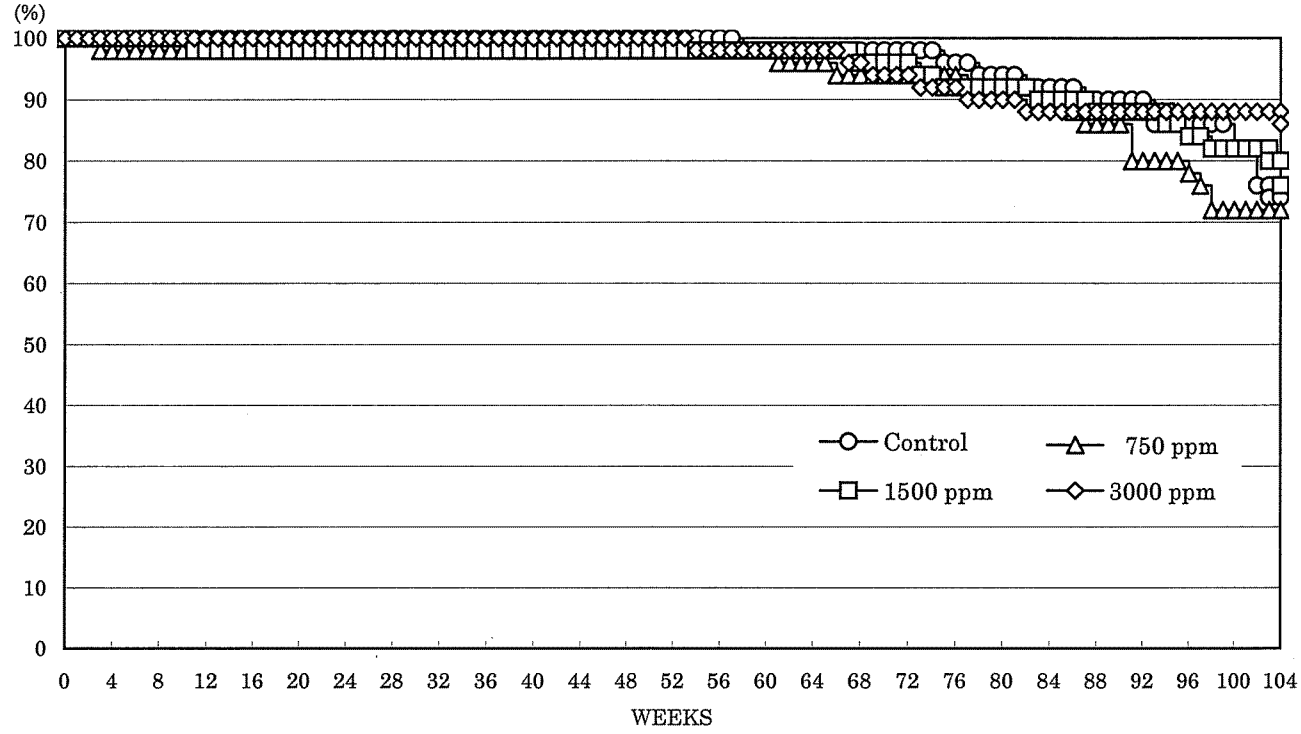


FIGURE 1 SURVIVAL RATE OF MALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

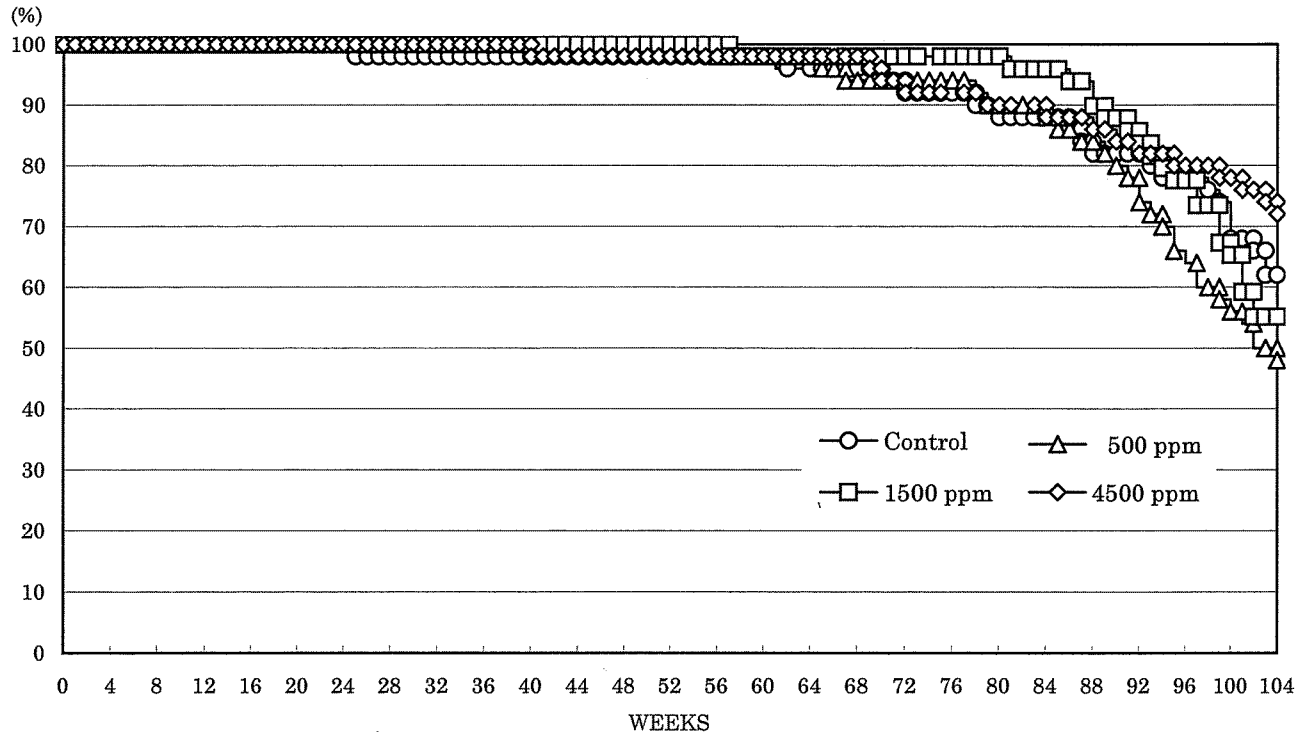


FIGURE 2 SURVIVAL RATE OF FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

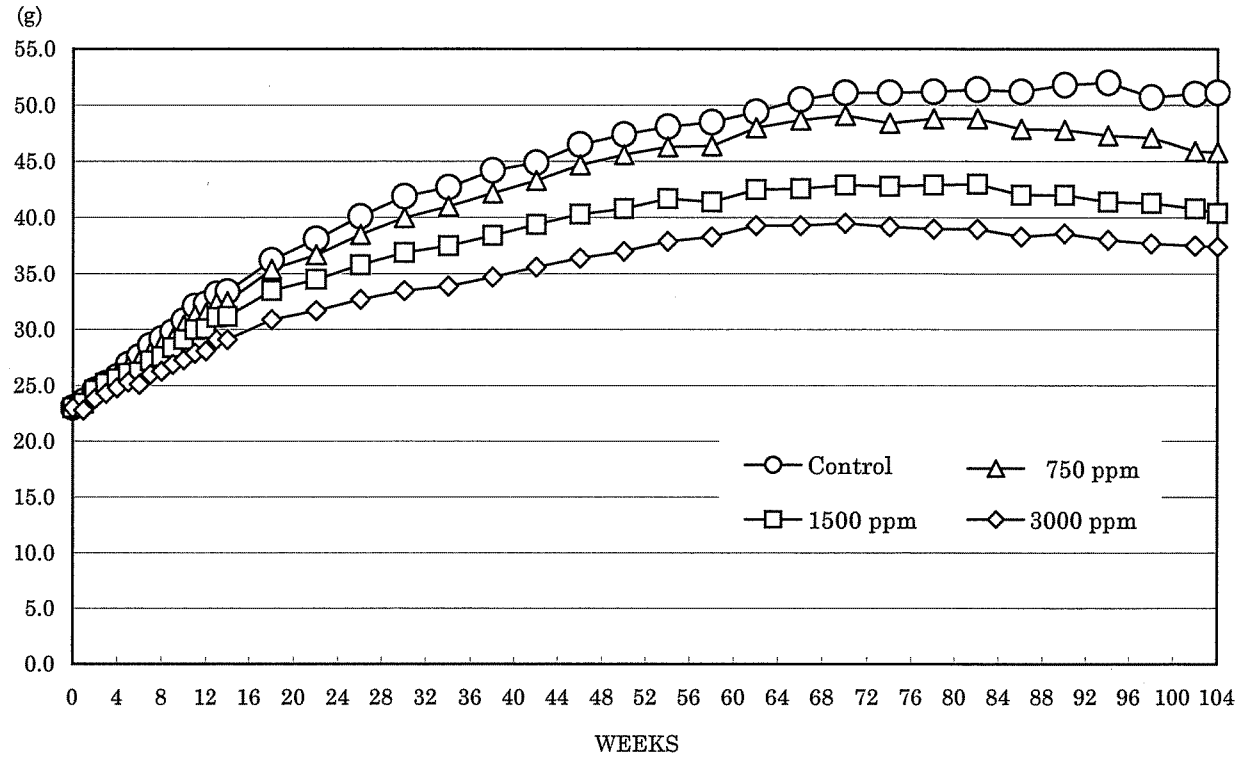


FIGURE 3 BODY WEIGHT CHANGES OF MALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

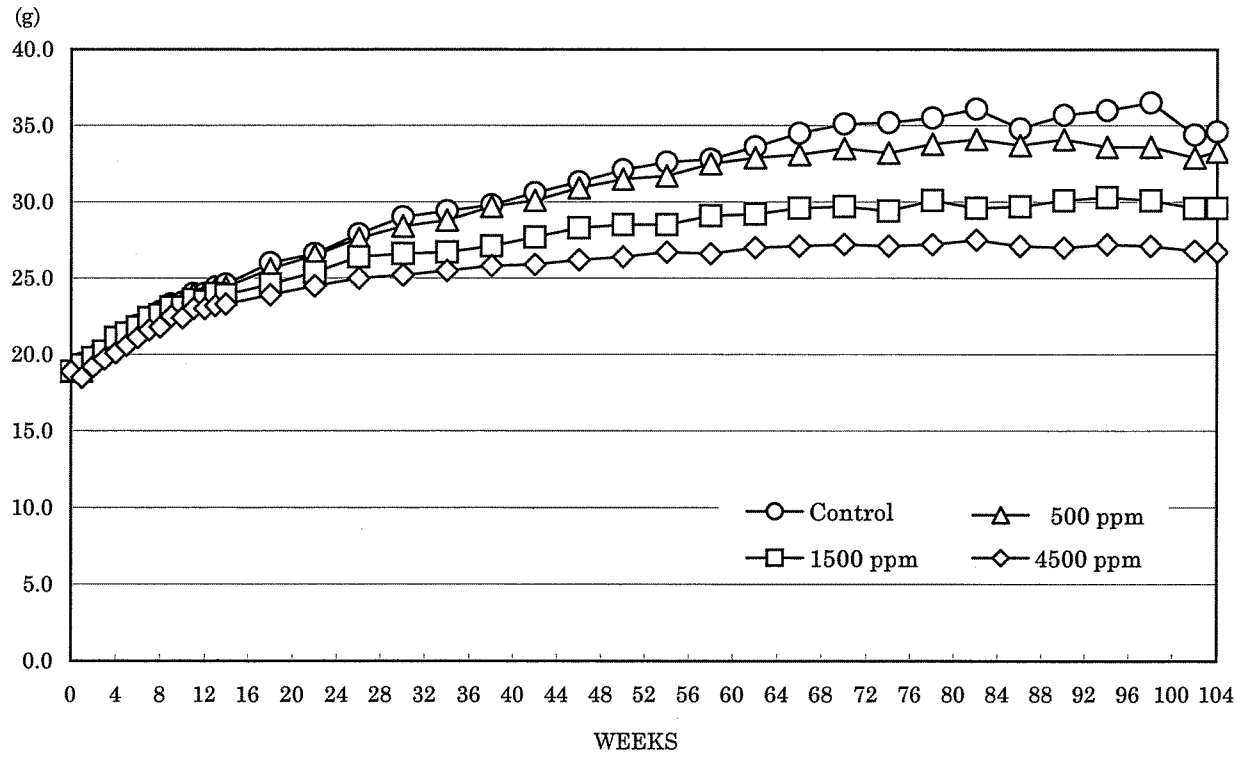


FIGURE 4 BODY WEIGHT CHANGES OF FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

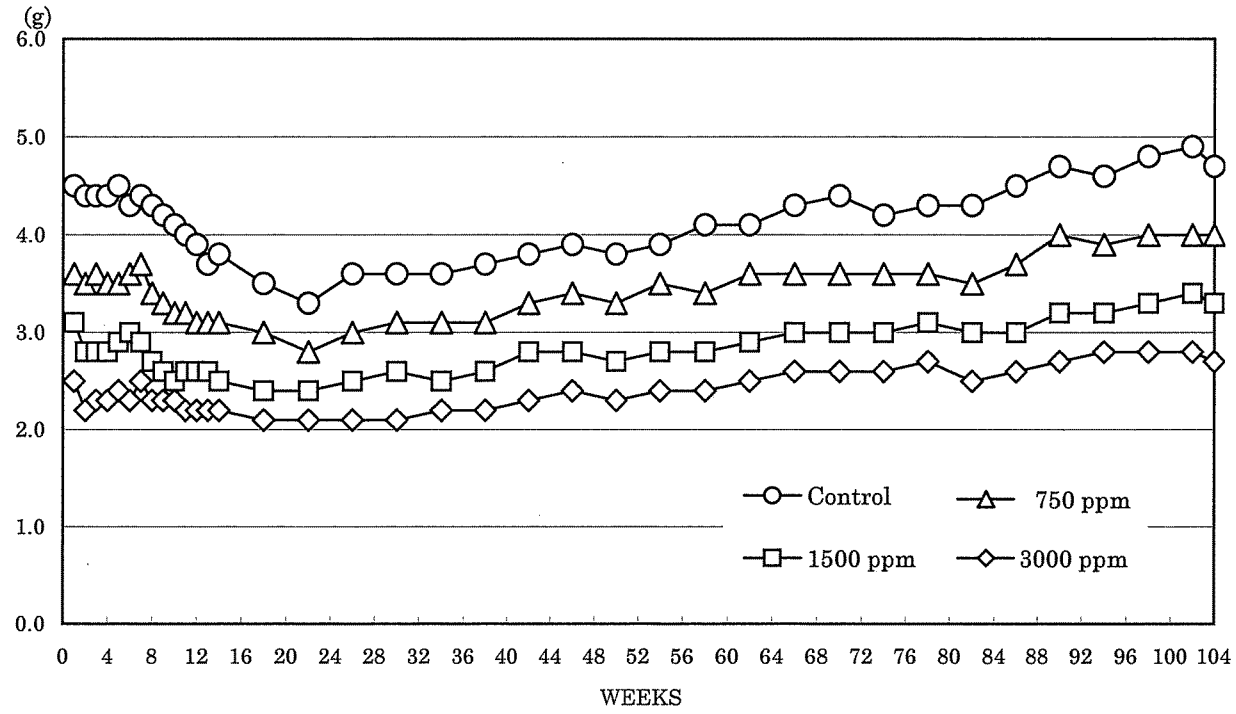


FIGURE 5 WATER CONSUMPTION CHANGES OF MALE MICE IN THE 2-YEARDRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

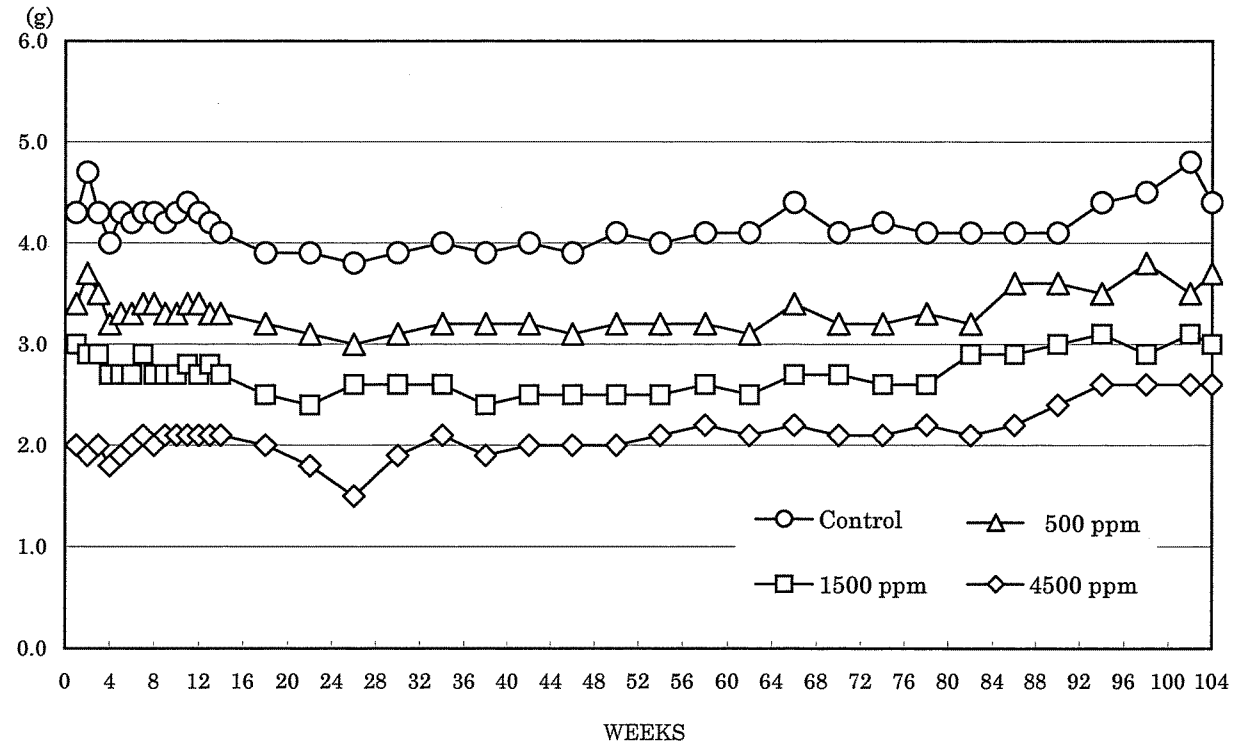


FIGURE 6 WATER CONSUMPTION CHANGES OF FEMALE MICE IN THE 2-YEARDRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

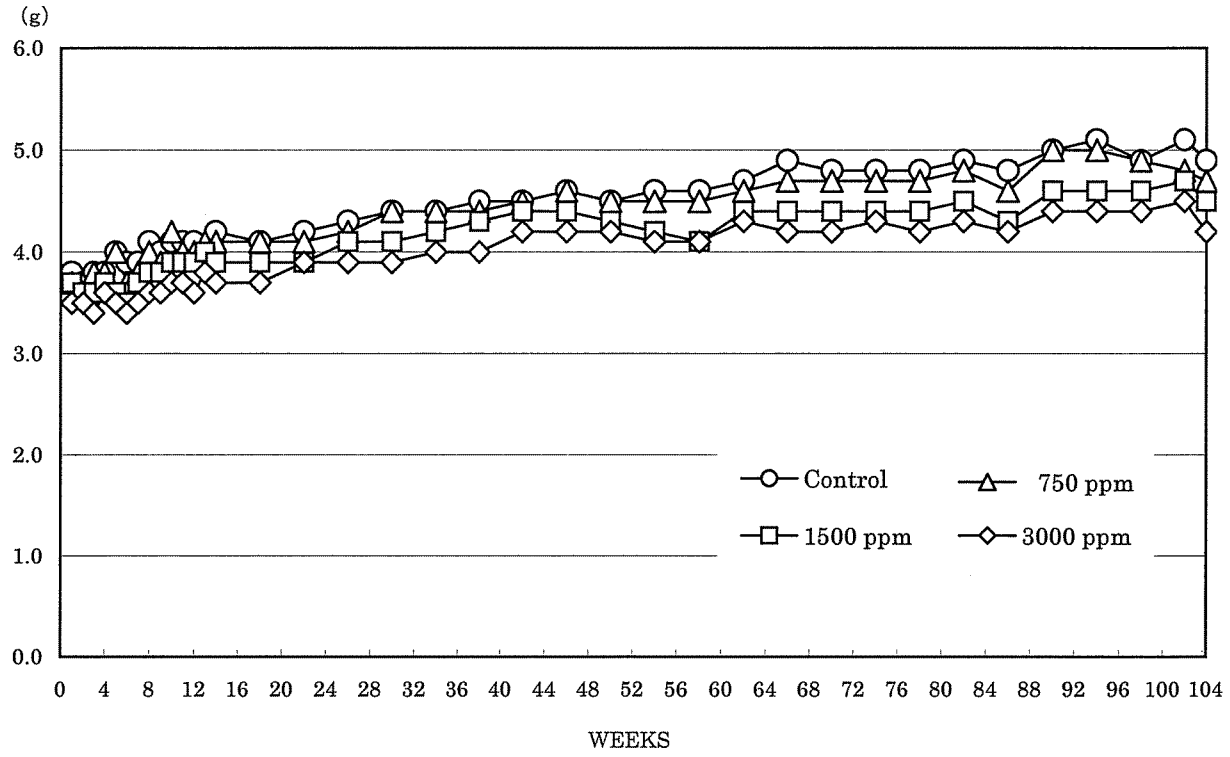


FIGURE 7 FOOD CONSUMPTION CHANGES OF MALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

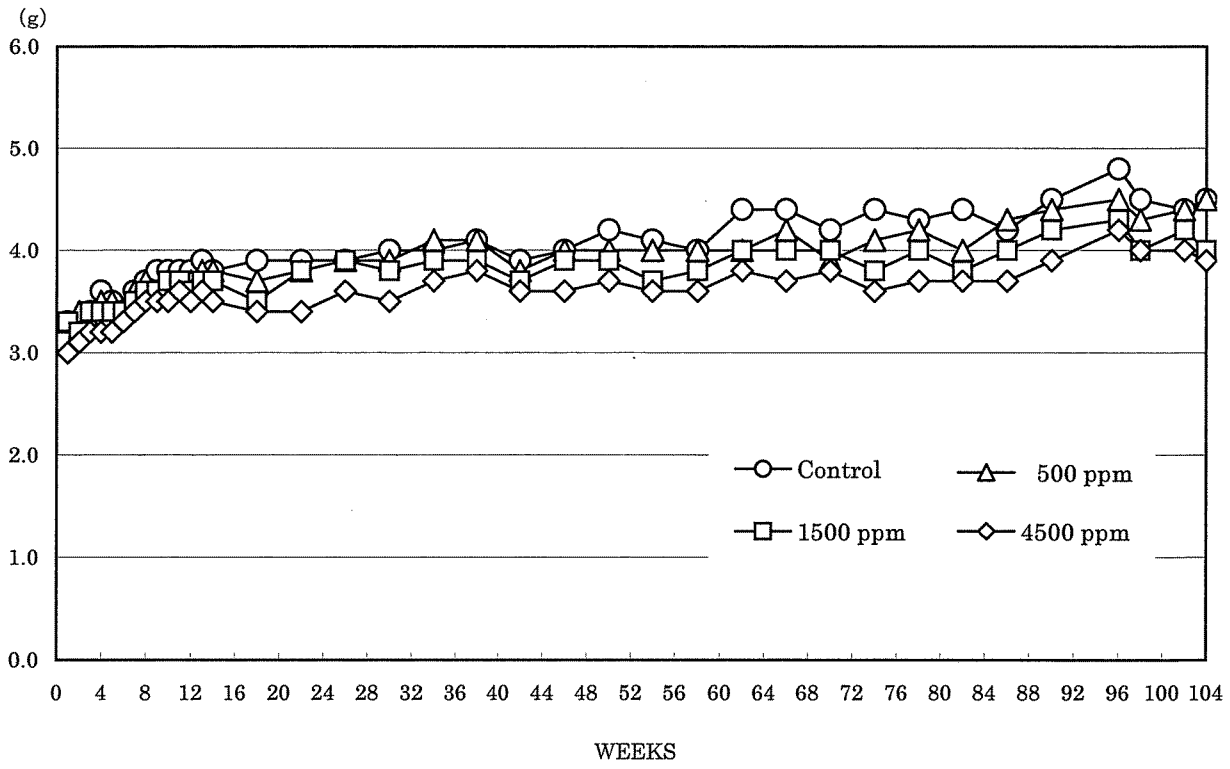


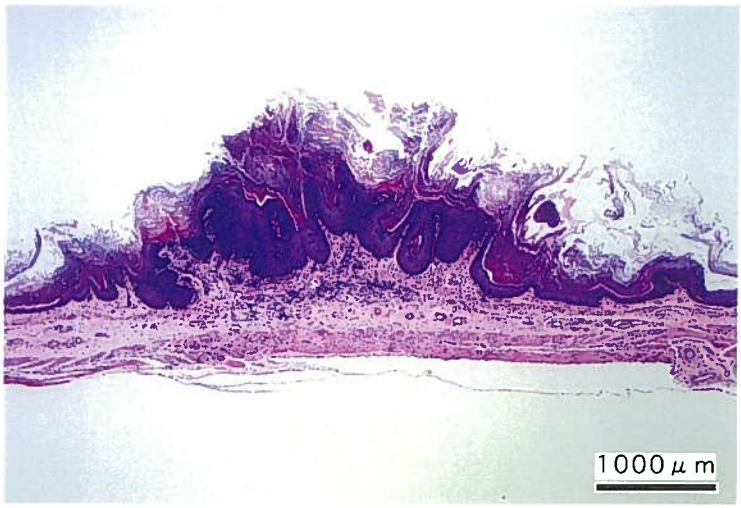
FIGURE 8 FOOD CONSUMPTION CHANGES OF FEMALE MICE IN THE 2-YEAR DRINKING WATER STUDY OF 2-HYDROXYETHYL ACRYLATE

PHOTOGRAPHS

PHOTOGRAPH 1 FORESTOMACH : SQUAMOUS CELL HYPERPLASIA
MOUSE, FEMALE, 4500 ppm, ANIMAL NO. 0348-2324 (H&E)

PHOTOGRAPH 2 KIDNEY : DESQUAMATION : PELVIS,
MOUSE, FEMALE, 1500 ppm, ANIMAL NO. 0348-2232 (H&E)

PHOTOGRAPH 3 NASAL CAVITY : RESPIRATORY METAPLASIA : GLAND,
MOUSE, MALE, 1500 ppm, ANIMAL NO. 0348-1222 (H&E)



PHOTOGRAPH 1

PHOTOGRAPH 2

PHOTOGRAPH 3