ジクロロメタンのマウスを用いた
吸入による2週間毒性試験報告書

試験番号：0230

APPENDIX

APPENDIXES

APPENDIXES (CONTINUED)

APPENDIX A 1

BODY WEIGHT CHANGES :SUMMARY, MOUSE : MALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
UNIT : g
REPORT TYPE : A1 2
SEX : MALE

BODY WEIGHT CHANGES (SUMMARY)
ALL ANIMALS

Group Name	Administration week-day 0-0	1-1	1-7	2-7
Control	22.8± 0.9	23.6± 0.9	24.6± 1.3	25.7± 1.2
1000ppm	22.5± 1.0	22.9± 1.3	23.7± 1.6	24.6± 1.8
2000ppm	22.4± 0.9	22.6± 0.7	23.9± 1.2	24.5± 1.3
4000ppm	22.6± 0.7	22.8± 0.8	24.4± 1.0	25.2± 1.0
8000ppm	22.7± 0.7	21.9± 0.7**	23.9± 0.9	25.4± 1.6
16000ppm	22.5± 0.8	20.4± 0.6**	21.5± 1.0**	22.4± 1.6**

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

APPENDIX A 2

BODY WEIGHT CHANGES : SUMMARY, MOUSE : FEMALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
UNIT : g
REPORT TYPE : A1 2
SEX : FEMALE

BODY WEIGHT CHANGES (SUMMARY)
ALL ANIMALS

Group Name	Administration week-day 0-0	1-1	1-7	2-7
Control	18.4± 0.8	18.7± 0.8	19.4± 0.9	20.8± 1.3
1000ppm	18.2± 0.6	18.8± 0.5	19.6± 0.6	21.1± 1.1
2000ppm	18.3± 0.7	18.5± 0.4	19.2± 0.5	20.5± 0.7
4000ppm	18.1± 0.7	18.2± 0.8	20.1± 0.7	20.2± 0.7
8000ppm	17.9± 0.4	17.4± 0.3**	19.0± 0.5	19.9± 0.5
16000ppm	17.9± 0.6	14.8± 0.3**	-	-

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

(HAN260)

APPENDIX B 1

FOOD CONSUMPTION CHANGES : SUMMARY, MOUSE : MALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
UNIT : g
REPORT TYPE : A1 2
SEX : MALE

FOOD CONSUMPTION CHANGES (SUMMARY)
ALL ANIMALS

Group Name	Administration week-day(effective) 1-7(7)	2-7(7)
Control	4.4± 0.4	4.3± 0.2
1000ppm	4.2± 0.4	4.2± 0.3
2000ppm	4.3± 0.3	4.3± 0.3
4000ppm	4.3± 0.3	4.2± 0.2
8000ppm	4.0± 0.3*	4.6± 0.5
16000ppm	3.4± 0.2**	4.1± 0.3

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

APPENDIX B 2

FOOD CONSUMPTION CHANGES : SUMMARY, MOUSE : FEMALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
UNIT : g
REPORT TYPE : A1 2
SEX : FEMALE

FOOD CONSUMPTION CHANGES (SUMMARY)
ALL ANIMALS

Group Name	Administration 1-7(7)	week-day(effective) 2-7(7)
Control	3.6± 0.2	3.7± 0.2
1000ppm	3.7± 0.3	3.7± 0.3
2000ppm	3.7± 0.3	3.8± 0.3
4000ppm	3.9± 0.3*	3.5± 0.2
8000ppm	3.5± 0.3	3.6± 0.2
16000ppm	-	-

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

APPENDIX C 1

HEMATOLOGY : SUMMARY, MOUSE : MALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
SAMPLING DATE : 002-7
SEX : MALE REPORT TYPE : A1

HEMATOLOGY (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	RED BLOOD CELL $10^6/\mu\ell$	HEMOGLOBIN g/dℓ	HEMATOCRIT %	MCV f ℓ	MCH p g	MCHC g/dℓ	PLATELET $10^3/\mu\ell$
Control	5	10.19± 0.09	15.5± 0.2	47.1± 0.7	46.2± 0.3	15.3± 0.1	33.0± 0.5	1201± 47
1000ppm	5	10.61± 0.28	16.4± 0.4	49.1± 1.3	46.2± 0.3	15.4± 0.2	33.4± 0.6	1193± 138
2000ppm	5	10.30± 0.45	16.0± 0.7	47.7± 1.7	46.3± 0.7	15.5± 0.2	33.5± 0.4	1126± 88
4000ppm	5	10.52± 0.59	16.2± 0.9	48.9± 2.3	46.5± 0.8	15.4± 0.1	33.1± 0.7	1221± 237
8000ppm	5	10.43± 0.37	16.1± 0.6	48.9± 1.1	46.9± 0.7	15.4± 0.2	32.8± 0.5	1187± 167
16000ppm	5	10.34± 0.39	15.8± 0.3	47.2± 1.2	45.6± 0.7	15.3± 0.4	33.5± 0.5	1422± 81

Significant difference ; * : $P \leqq 0.05$ ** : $P \leqq 0.01$ Test of Dunnett

(HCL070)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
SAMPLING DATE : 002-7
SEX : MALE REPORT TYPE : A1

HEMATOLOGY (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	WBC $10^3/\mu\ell$	Differential WBC (%) N-BAND	N-SEG	EOSINO	BASO	MONO	LYMPHO	OTHERS
Control	5	1.70± 0.67	1± 1	10± 1	1± 1	0± 0	2± 1	86± 1	0± 0
1000ppm	5	1.58± 0.74	0± 1	10± 4	1± 1	0± 0	2± 2	87± 4	0± 0
2000ppm	5	2.15± 1.03	1± 1	10± 2	1± 1	0± 0	2± 1	87± 3	0± 1
4000ppm	5	2.19± 1.48	0± 1	13± 7	2± 1	0± 0	2± 1	83± 8	0± 0
8000ppm	5	1.23± 0.52	1± 2	12± 6	1± 1	0± 0	2± 1	85± 5	0± 0
16000ppm	5	0.93± 0.33	0± 0	11± 5	0± 1	0± 0	3± 1	85± 6	0± 0

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

(HCL070)

APPENDIX C 2

HEMATOLOGY : SUMMARY, MOUSE : FEMALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
SAMPLING DATE : 002-7
SEX : FEMALE REPORT TYPE : A1

HEMATOLOGY (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	RED BLOOD CELL $10^6/\mu\ell$	HEMOGLOBIN g/dℓ	HEMATOCRIT %	MCV f ℓ	MCH p g	MCHC g/dℓ	PLATELET $10^3/\mu\ell$
Control	5	10.16± 0.40	15.4± 0.4	46.3± 1.8	45.5± 0.6	15.2± 0.2	33.3± 0.7	922± 129
1000ppm	5	10.47± 0.53	16.0± 0.7	48.4± 2.5	46.2± 0.6	15.3± 0.1	33.2± 0.4	1156± 23**
2000ppm	5	10.54± 0.35	16.2± 0.5	49.0± 1.8	46.5± 0.8	15.4± 0.4	33.0± 0.7	1156± 94**
4000ppm	5	10.63± 0.28	16.2± 0.6	49.1± 1.5	46.2± 0.5	15.3± 0.4	33.0± 0.5	1152± 89**
8000ppm	5	10.42± 0.36	16.0± 0.7	47.7± 2.0	45.8± 0.7	15.3± 0.2	33.4± 0.4	1325± 109**
16000ppm	0	-	-	-	-	-	-	-

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
SAMPLING DATE : 002-7
SEX : FEMALE REPORT TYPE : A1

HEMATOLOGY (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	WBC $10^3/\mu\ell$		Differential WBC (%) N-BAND		N-SEG		EOSINO		BASO		MONO		LYMPHO		OTHERS	
Control	5	1.79±	0.72	0±	0	8±	2	1±	1	0±	0	2±	1	89±	3	0±	0
1000ppm	5	2.25±	1.04	0±	0	8±	2	1±	1	0±	0	2±	1	88±	1	0±	0
2000ppm	5	2.25±	0.78	0±	0	11±	4	1±	1	0±	0	3±	1	85±	5	0±	0
4000ppm	5	1.94±	0.40	1±	1	11±	7	1±	1	0±	0	2±	1	86±	8	0±	0
8000ppm	5	1.82±	0.93	0±	1	9±	4	1±	1	0±	0	2±	1	87±	4	0±	0
16000ppm	0	-		-		-		-		-		-		-		-	

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

(HCL070)

APPENDIX D 1

BIOCHEMISTRY : SUMMARY, MOUSE : MALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
SAMPLING DATE : 003-1
SEX : MALE REPORT TYPE : A1

BIOCHEMISTRY (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	TOTAL PROTEIN g/dℓ	ALBUMIN g/dℓ	A/G RATIO	T-BILIRUBIN mg/dℓ	GLUCOSE mg/dℓ	T-CHOLESTEROL mg/dℓ	GOT IU/ℓ
Control	5	5.2± 0.2	2.8± 0.1	1.2± 0.0	0.30± 0.07	302± 14	95± 10	34± 1
1000ppm	5	5.1± 0.2	2.8± 0.1	1.3± 0.1	0.36± 0.14	270± 57	83± 9	35± 6
2000ppm	5	4.9± 0.2	2.8± 0.1	1.3± 0.1	0.36± 0.08	292± 18	83± 7	35± 4
4000ppm	5	5.1± 0.5	2.8± 0.3	1.2± 0.1	0.36± 0.10	281± 39	89± 17	39± 3
8000ppm	5	5.0± 0.4	2.9± 0.3	1.3± 0.1	0.33± 0.06	305± 22	98± 7	36± 5
16000ppm	5	4.9± 0.2	2.8± 0.1	1.3± 0.0	0.32± 0.04	292± 20	87± 11	36± 5

Significant defference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
SAMPLING DATE : 003-1
SEX : MALE REPORT TYPE : A1

BIOCHEMISTRY (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	GPT IU/ℓ		LDH IU/ℓ		CPK IU/ℓ		UREA NITROGEN mg/dℓ		SODIUM mEq/ℓ		POTASSIUM mEq/ℓ		CHLORIDE mEq/ℓ	
Control	5	13±	1	217±	33	86±	26	19.0±	4.9	153±	2	4.7±	0.1	120±	2
1000ppm	5	13±	1	216±	47	78±	26	20.3±	4.5	153±	1	4.3±	0.5	121±	1
2000ppm	5	13±	2	221±	83	100±	70	21.0±	3.6	152±	1	4.3±	0.5	119±	2
4000ppm	5	15±	2	279±	78	108±	74	21.8±	4.8	154±	2	4.3±	0.6	121±	2
8000ppm	5	14±	6	299±	195	133±	79	18.5±	3.7	155±	1	4.4±	0.3	121±	1
16000ppm	5	13±	1	231±	56	98±	23	16.3±	1.1	154±	1	4.2±	0.3	121±	1

Significant defference : * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

(HCL074)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
SAMPLING DATE : 003-1
SEX : MALE REPORT TYPE : A1

BIOCHEMISTRY (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	CALCIUM mg/dℓ		INORGANIC PHOSPHORUS mg/dℓ	
Control	5	9.2±	0.1	9.0±	1.2
1000ppm	5	8.9±	0.4	8.3±	0.9
2000ppm	5	9.0±	0.2	8.6±	2.1
4000ppm	5	9.3±	0.7	9.4±	1.2
8000ppm	5	8.9±	0.4	10.0±	1.4
16000ppm	5	9.0±	0.5	9.5±	0.9

Significant defference : * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

APPENDIX D 2

BIOCHEMISTRY : SUMMARY, MOUSE : FEMALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
SAMPLING DATE : 003-1
SEX : FEMALE REPORT TYPE : A1

BIOCHEMISTRY (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	TOTAL PROTEIN g/dℓ	ALBUMIN g/dℓ	A/G RATIO	T-BILIRUBIN mg/dℓ	GLUCOSE mg/dℓ	T-CHOLESTEROL mg/dℓ	GOT IU/ℓ
Control	5	5.1± 0.2	3.0± 0.1	1.5± 0.1	0.38± 0.08	297± 66	81± 5	42± 6
1000ppm	5	5.2± 0.3	3.2± 0.2	1.6± 0.2	0.33± 0.11	278± 47	79± 12	45± 4
2000ppm	5	5.3± 0.1	3.2± 0.1	1.6± 0.1	0.27± 0.20	253± 29	72± 4	47± 6
4000ppm	5	5.3± 0.3	3.3± 0.2	1.6± 0.1	0.29± 0.12	268± 37	81± 2	46± 10
8000ppm	5	5.2± 0.5	3.2± 0.3	1.5± 0.1	0.36± 0.15	286± 31	89± 9	42± 4
16000ppm	0	-	-	-	-	-	-	-

Significant defference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

(HCL074)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
SAMPLING DATE : 003-1
SEX : FEMALE REPORT TYPE : A1

BIOCHEMISTRY (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	GPT IU/ℓ		LDH IU/ℓ		CPK IU/ℓ		UREA NITROGEN mg/dℓ		SODIUM mEq/ℓ		POTASSIUM mEq/ℓ		CHLORIDE mEq/ℓ	
Control	5	13±	3	293±	161	120±	62	18.7±	1.6	153±	2	4.6±	0.9	120±	2
1000ppm	5	16±	5	260±	37	89±	47	19.4±	3.3	156±	4	4.5±	1.0	122±	2
2000ppm	5	17±	3	329±	99	162±	45	17.8±	1.9	154±	2	5.1±	1.2	121±	3
4000ppm	5	17±	2	304±	65	108±	43	16.6±	2.1	156±	3	4.7±	0.5	122±	2
8000ppm	5	14±	4	317±	109	122±	66	15.9±	2.0	155±	2	4.7±	1.0	121±	2
16000ppm	0	-		-		-		-		-		-		-	

Significant defference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

(HCL074)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
SAMPLING DATE : 003-1
SEX : FEMALE REPORT TYPE : A1

BIOCHEMISTRY (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	CALCIUM mg/dℓ	INORGANIC PHOSPHORUS mg/dℓ
Control	5	8.9± 0.5	8.3± 1.3
1000ppm	5	9.1± 0.7	7.7± 2.1
2000ppm	5	9.1± 0.4	10.3± 2.1
4000ppm	5	9.2± 0.3	9.0± 2.4
8000ppm	5	8.9± 0.5	8.4± 0.9
16000ppm	0	-	-

Significant defference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

(HCL074)

APPENDIX E 1

GROSS FINDINGS : SUMMARY, MOUSE : MALE : SACRIFICED ANIMALS

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : MALE

GROSS FINDINGS (SUMMARY)
SACRIFICED ANIMALS (2W)

Organ	Findings	Group Name / NO. of Animals	Control 10 (%)	1000ppm 10 (%)	2000ppm 10 (%)	4000ppm 10 (%)
spleen	black zone		0 (0)	1 (10)	1 (10)	0 (0)
kidney	hydronephrosis		0 (0)	0 (0)	0 (0)	1 (10)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : MALE

GROSS FINDINGS (SUMMARY)
SACRIFICED ANIMALS (2W)

Organ	Findings	Group Name NO. of Animals	8000ppm 10 (%)	16000ppm 5 (%)
spleen	black zone		1 (10)	0 (0)
kidney	hydronephrosis		0 (0)	0 (0)

APPENDIX E 2

GROSS FINDINGS : SUMMARY, MOUSE : FEMALE : SACRIFICED ANIMALS

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : FEMALE

GROSS FINDINGS (SUMMARY)
SACRIFICED ANIMALS (2W)

Organ	Findings	Group Name NO. of Animals	Control 10 (%)	1000ppm 10 (%)	2000ppm 10 (%)	4000ppm 10 (%)
spleen	black zone		1 (10)	1 (10)	0 (0)	0 (0)

STUDY NO. : 0Z30
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : FEMALE

GROSS FINDINGS (SUMMARY)
SACRIFICED ANIMALS (2W)

Organ	Findings	Group Name	8000ppm	16000ppm
		NO. of Animals	10 (%)	0 (%)
spleen	black zone		1 (10)	- (-)

APPENDIX F 1

ORGAN WEIGHT, ABSOLUTE : SUMMARY, MOUSE : MALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : MALE
UNIT: g

ORGAN WEIGHT:ABSOLUTE (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	Body Weight	THYMUS	ADRENALS	TESTES	HEART	LUNGS
Control	5	25.4± 1.7	0.059± 0.007	0.008± 0.003	0.170± 0.021	0.136± 0.006	0.138± 0.014
1000ppm	5	23.9± 1.6	0.042± 0.015	0.009± 0.003	0.168± 0.021	0.126± 0.014	0.128± 0.013
2000ppm	5	24.5± 1.1	0.038± 0.005	0.008± 0.002	0.165± 0.018	0.127± 0.009	0.133± 0.008
4000ppm	5	25.2± 1.1	0.042± 0.008	0.009± 0.002	0.166± 0.029	0.135± 0.017	0.149± 0.012
8000ppm	5	25.0± 2.0	0.036± 0.008	0.008± 0.003	0.169± 0.007	0.120± 0.010	0.150± 0.017
16000ppm	5	22.4± 1.6	0.021± 0.002**	0.008± 0.001	0.148± 0.005	0.121± 0.005	0.150± 0.016

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : MALE
UNIT: g

ORGAN WEIGHT:ABSOLUTE (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	KIDNEYS	SPLEEN	LIVER	BRAIN
Control	5	0.372± 0.025	0.054± 0.004	1.296± 0.130	0.440± 0.020
1000ppm	5	0.379± 0.021	0.047± 0.012	1.179± 0.171	0.438± 0.015
2000ppm	5	0.383± 0.027	0.049± 0.005	1.255± 0.079	0.446± 0.019
4000ppm	5	0.459± 0.179	0.054± 0.019	1.299± 0.113	0.440± 0.019
8000ppm	5	0.384± 0.029	0.046± 0.006	1.370± 0.187	0.443± 0.008
16000ppm	5	0.374± 0.027	0.036± 0.003**	1.345± 0.083	0.446± 0.013

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

APPENDIX F 2

ORGAN WEIGHT, ABSOLUTE : SUMMARY, MOUSE : FEMALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : MALE
UNIT: %

ORGAN WEIGHT:RELATIVE (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	Body Weight (g)	THYMUS	ADRENALS	TESTES	HEART	LUNGS
Control	5	25.4± 1.7	0.233± 0.036	0.030± 0.010	0.671± 0.091	0.537± 0.018	0.543± 0.062
1000ppm	5	23.9± 1.6	0.174± 0.051*	0.036± 0.009	0.703± 0.096	0.524± 0.034	0.535± 0.027
2000ppm	5	24.5± 1.1	0.157± 0.025**	0.033± 0.010	0.671± 0.068	0.520± 0.032	0.543± 0.050
4000ppm	5	25.2± 1.1	0.167± 0.028*	0.037± 0.007	0.656± 0.095	0.536± 0.058	0.594± 0.061
8000ppm	5	25.0± 2.0	0.143± 0.029**	0.032± 0.011	0.680± 0.059	0.481± 0.022	0.599± 0.050
16000ppm	5	22.4± 1.6	0.095± 0.009**	0.036± 0.005	0.661± 0.030	0.543± 0.027	0.669± 0.055**

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

(HCL042)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : MALE
UNIT: %

ORGAN WEIGHT:RELATIVE (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	KIDNEYS	SPLEEN	LIVER	BRAIN
Control	5	1.464± 0.080	0.213± 0.012	5.093± 0.334	1.734± 0.105
1000ppm	5	1.585± 0.094	0.193± 0.040	4.912± 0.480	1.836± 0.092
2000ppm	5	1.560± 0.068	0.200± 0.021	5.124± 0.389	1.819± 0.082
4000ppm	5	1.823± 0.713	0.216± 0.077	5.155± 0.338	1.749± 0.077
8000ppm	5	1.538± 0.060	0.183± 0.025	5.461± 0.337	1.777± 0.122
16000ppm	5	1.668± 0.077	0.161± 0.016	6.004± 0.123**	1.993± 0.090**

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

APPENDIX G 1

ORGAN WEIGHT, RELATIVE : SUMMARY, MOUSE : MALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : FEMALE
UNIT: g

ORGAN WEIGHT:ABSOLUTE (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	Body Weight	THYMUS	ADRENALS	OVARIES	HEART	LUNGS
Control	5	20.7± 1.8	0.067± 0.014	0.011± 0.003	0.015± 0.005	0.110± 0.012	0.126± 0.014
1000ppm	5	21.3± 1.2	0.079± 0.010	0.011± 0.002	0.016± 0.003	0.113± 0.007	0.136± 0.009
2000ppm	5	20.3± 0.8	0.074± 0.010	0.010± 0.001	0.014± 0.003	0.117± 0.007	0.138± 0.006
4000ppm	5	20.7± 0.7	0.065± 0.007	0.011± 0.002	0.012± 0.004	0.110± 0.011	0.133± 0.019
8000ppm	5	20.0± 0.7	0.044± 0.003**	0.010± 0.002	0.015± 0.006	0.101± 0.005	0.136± 0.013
16000ppm	0	-	-	-	-	-	-

Significant difference : * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

(HCL040)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : FEMALE
UNIT: g

ORGAN WEIGHT:ABSOLUTE (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	KIDNEYS	SPLEEN	LIVER	BRAIN
Control	5	0.270± 0.021	0.058± 0.006	0.972± 0.116	0.440± 0.010
1000ppm	5	0.268± 0.014	0.056± 0.009	1.037± 0.132	0.439± 0.008
2000ppm	5	0.257± 0.012	0.057± 0.002	1.055± 0.070	0.441± 0.012
4000ppm	5	0.260± 0.015	0.057± 0.008	1.045± 0.075	0.440± 0.021
8000ppm	5	0.266± 0.018	0.046± 0.006	1.103± 0.039	0.438± 0.010
16000ppm	0	-	-	-	-

Significant difference ; * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

APPENDIX G 2

ORGAN WEIGHT, RELATIVE : SUMMARY, MOUSE : FEMALE

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : FEMALE
UNIT: %

ORGAN WEIGHT:RELATIVE (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	Body Weight (g)	THYMUS	ADRENALS	OVARIES	HEART	LUNGS
Control	5	20.7± 1.8	0.326± 0.053	0.052± 0.016	0.071± 0.021	0.534± 0.065	0.611± 0.070
1000ppm	5	21.3± 1.2	0.371± 0.056	0.052± 0.009	0.076± 0.015	0.532± 0.060	0.641± 0.053
2000ppm	5	20.3± 0.8	0.367± 0.058	0.051± 0.005	0.068± 0.016	0.575± 0.037	0.680± 0.034
4000ppm	5	20.7± 0.7	0.313± 0.028	0.053± 0.010	0.059± 0.023	0.530± 0.041	0.644± 0.095
8000ppm	5	20.0± 0.7	0.222± 0.011**	0.048± 0.011	0.076± 0.031	0.506± 0.025	0.681± 0.063
16000ppm	0	-	-	-	-	-	-

Significant difference : * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

(HCL042)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : FEMALE
UNIT: %

ORGAN WEIGHT:RELATIVE (SUMMARY)
SURVIVAL ANIMALS (2)

Group Name	NO. of Animals	KIDNEYS	SPLEEN	LIVER	BRAIN
Control	5	1.307± 0.039	0.283± 0.032	4.699± 0.333	2.139± 0.143
1000ppm	5	1.260± 0.070	0.263± 0.034	4.855± 0.404	2.067± 0.132
2000ppm	5	1.268± 0.053	0.281± 0.017	5.199± 0.354	2.173± 0.066
4000ppm	5	1.256± 0.064	0.274± 0.041	5.050± 0.243	2.127± 0.092
8000ppm	5	1.330± 0.098	0.232± 0.028	5.525± 0.188**	2.193± 0.061
16000ppm	0	-	-	-	-

Significant difference : * : P ≦ 0.05 ** : P ≦ 0.01 Test of Dunnett

APPENDIX H 1

HISTOLOGICAL FINDINGS : NON-NEOPLASTIC LESIONS : SUMMARY

MOUSE : MALE : DEAD AND MORIBUND ANIMALS

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : MALE

HISTOLOGICAL FINDINGS :NON-NEOPLASTIC LESIONS (SUMMARY)
DEAD AND MORIBUND ANIMALS (0- 2W)

Organ	Findings	Group Name	Control				1000ppm				2000ppm				4000ppm			
		No. of Animals on Study	0				0				0				0			
		Grade	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
			(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)
[Respiratory system]																		
lung			< 0>				< 0>				< 0>				< 0>			
	hemorrhage		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)

Grade 1 : Slight 2 : Moderate 3 : Marked 4 : Severe
< a > a : Number of animals examined at the site
b b : Number of animals with lesion
(c) c : b / a * 100

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : MALE

HISTOLOGICAL FINDINGS :NON-NEOPLASTIC LESIONS (SUMMARY)
DEAD AND MORIBUND ANIMALS (0- 2W)

Organ	Findings	Group Name	8000ppm				16000ppm			
		No. of Animals on Study	0				1			
		Grade	1	2	3	4	1	2	3	4
			(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)
[Respiratory system]										
lung			< 0>				< 1>			
	hemorrhage		-	-	-	-	1	0	0	0
			(-)	(-)	(-)	(-)	(100)	(0)	(0)	(0)

Grade 1 : Slight 2 : Moderate 3 : Marked 4 : Severe
< a > a : Number of animals examined at the site
b b : Number of animals with lesion
(c) c : b / a * 100

APPENDIX H 2

HISTOLOGICAL FINDINGS : NON-NEOPLASTIC LESIONS : SUMMARY

MOUSE : MALE : SACRIFICED ANIMALS

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : MALE

HISTOLOGICAL FINDINGS :NON-NEOPLASTIC LESIONS (SUMMARY)
SACRIFICED ANIMALS (2W)

Organ	Findings	Group Name	Control				1000ppm				2000ppm				4000ppm			
		No. of Animals on Study	2				2				2				2			
		Grade	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
			(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)
[Hematopoietic system]																		
spleen			< 2>				< 2>				< 2>				< 2>			
	melanin		0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
			(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)

Grade 1 : Slight 2 : Moderate 3 : Marked 4 : Severe
< a > a : Number of animals examined at the site
b b : Number of animals with lesion
(c) c : b / a * 100

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : MALE

HISTOLOGICAL FINDINGS :NON-NEOPLASTIC LESIONS (SUMMARY)
SACRIFICED ANIMALS (2W)

Organ	Findings	Group Name	8000ppm				16000ppm			
		No. of Animals on Study	2				1			
		Grade	1	2	3	4	1	2	3	4
			(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)
[Hematopoietic system]										
spleen										
	melanin		< 2>				< 1>			
			1	0	0	0	0	0	0	0
			(50)	(0)	(0)	(0)	(0)	(0)	(0)	(0)

Grade 1 : Slight 2 : Moderate 3 : Marked 4 : Severe
< a > a : Number of animals examined at the site
b b : Number of animals with lesion
(c) c : b / a * 100

APPENDIX H 3

HISTOLOGICAL FINDINGS : NON-NEOPLASTIC LESIONS : SUMMARY

MOUSE : FEMALE : DEAD AND MORIBUND ANIMALS

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : FEMALE

HISTOLOGICAL FINDINGS :NON-NEOPLASTIC LESIONS (SUMMARY)
DEAD AND MORIBUND ANIMALS (0- 2W)

Organ	Findings	Group Name	Control				1000ppm				2000ppm				4000ppm			
		No. of Animals on Study	0				0				0				0			
		Grade	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
			(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)
[Digestive system]																		
liver			< 0>				< 0>				< 0>				< 0>			
	fatty change:central		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)

Grade 1 : Slight 2 : Moderate 3 : Marked 4 : Severe
< a > a : Number of animals examined at the site
b b : Number of animals with lesion
(c) c : b / a * 100

Organ	Findings	Group Name	8000ppm				16000ppm			
		No. of Animals on Study	0				2			
		Grade	1	2	3	4	1	2	3	4
			(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)
[Digestive system]										
liver			< 0>				< 2>			
	fatty change:central		-	-	-	-	1	0	0	0
			(-)	(-)	(-)	(-)	(50)	(0)	(0)	(0)

Grade 1 : Slight 2 : Moderate 3 : Marked 4 : Severe
< a > a : Number of animals examined at the site
b b : Number of animals with lesion
(c) c : b / a * 100

APPENDIX H 4

HISTOLOGICAL FINDINGS : NON-NEOPLASTIC LESIONS : SUMMARY

MOUSE : FEMALE : SACRIFICED ANIMALS

(2-WEEK STUDY)

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : FEMALE

HISTOLOGICAL FINDINGS :NON-NEOPLASTIC LESIONS (SUMMARY)
SACRIFICED ANIMALS (2W)

Organ	Findings	Group Name	Control				1000ppm				2000ppm				4000ppm			
		No. of Animals on Study	2				2				2				2			
		Grade	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
			(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)
[Urinary system]																		
kidney																		
	basophilic change		< 2>				< 2>				< 2>				< 2>			
			0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
			(0)	(0)	(0)	(0)	(50)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)	(0)

Grade 1 : Slight 2 : Moderate 3 : Marked 4 : Severe
< a > a : Number of animals examined at the site
b b : Number of animals with lesion
(c) c : b / a * 100

STUDY NO. : 0230
ANIMAL : MOUSE BDF1
REPORT TYPE : A1
SEX : FEMALE

HISTOLOGICAL FINDINGS :NON-NEOPLASTIC LESIONS (SUMMARY)
SACRIFICED ANIMALS (2W)

Organ	Findings	Group Name	8000ppm				16000ppm			
		No. of Animals on Study	2				0			
		Grade	1	2	3	4	1	2	3	4
			(%)	(%)	(%)	(%)	(%)	(%)	(%)	(%)
[Urinary system]										
kidney			< 2>				< 0>			
	basophilic change		0	0	0	0	-	-	-	-
			(0)	(0)	(0)	(0)	(-)	(-)	(-)	(-)

Grade 1 : Slight 2 : Moderate 3 : Marked 4 : Severe
< a > a : Number of animals examined at the site
b b : Number of animals with lesion
(c) c : b / a * 100

APPENDIX I 1

IDENTITY OF DICHLOROMETHANE IN THE 2-WEEK INHALATION STUDY

IDENTITY OF DICHLOROMETHANE IN THE 2-WEEK INHALATION STUDY

Lot No. APR5259

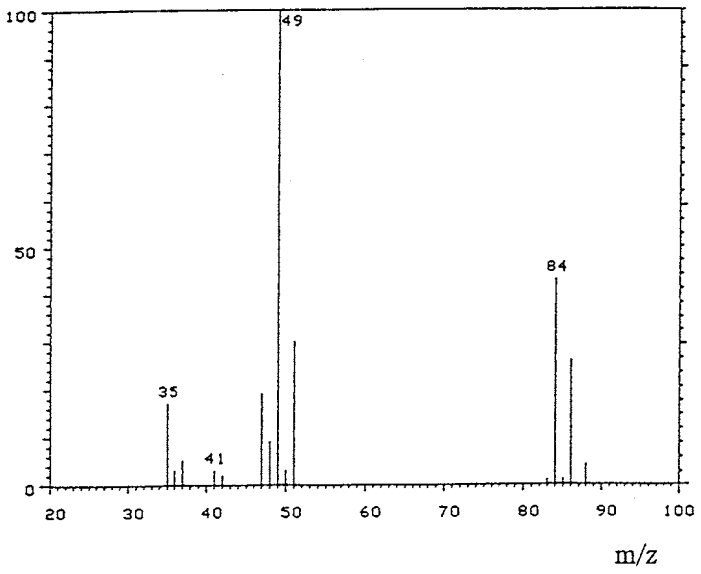
1. Spectral data

Mass Spectrometry

Instrument : Hitachi M-80B Mass Spectrometer

Ionization : EI(Electron Ionization)

Ionization Voltage : 70eV



Mass Spectrum of Test Substance

Results: The mass spectrum was consistent with literature spectrum.

Determined Values Fragment Peak(m/z)	Literature Values* Fragment Peak(m/z)
35	35
49	49
84	84

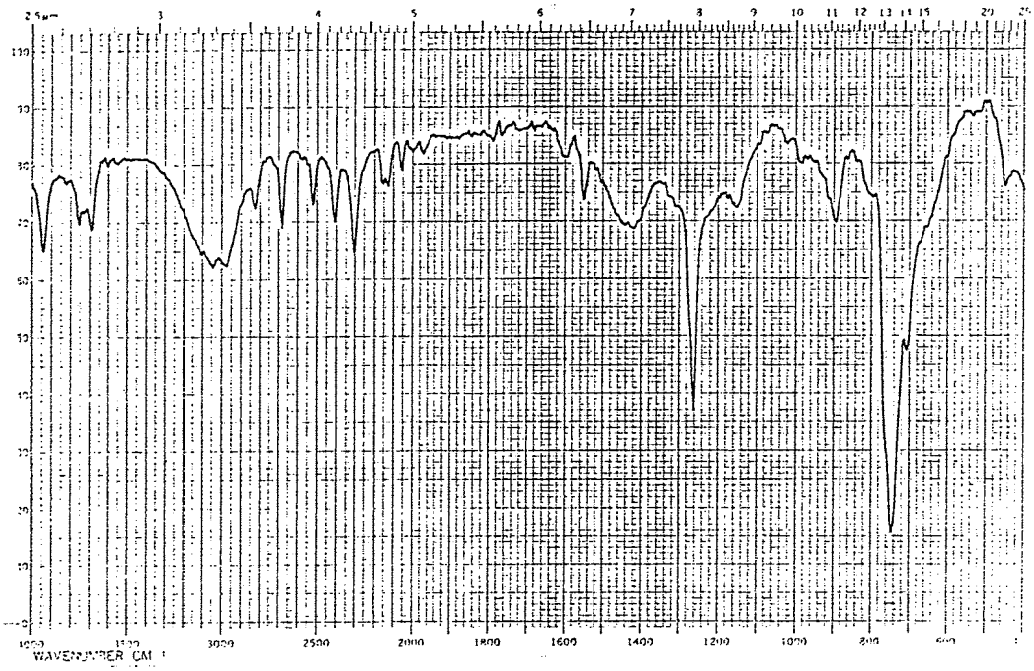
(*EPA/NIH Mass Spectral Data Base (1978) Vol. 1, p. 33.)

Infrared Spectrometry

Instrument : Hitachi 270-30 Infrared Spectrometer

Cell : KBr Liquid Cell

Slit : Medium



Infrared Spectrum of Test Substance

Results: The infrared spectrum was consistent with literature spectrum.

Determined Values Wave Number(cm^{-1})	Literature Values* Wave Number(cm^{-1})
430～ 480	
650～ 840	650～ 850
870～ 940	870～ 940
970～1000	970～1000
1120～1180	1130～1180
1200～1340	1200～1350
1370～1500	1380～1500
1530～1570	1540～1570
1580～1630	1580～1630
2040～2090	2050～2090
2100～2190	2120～2190
2250～2360	2280～2370
2380～2460	2400～2460
2500～2550	2500～2560
2650～2730	2650～2730
2800～2860	2800～2860
2900～3200	2900～3200
3650～3730	3670～3750
3730～3800	3750～3800
3900～4000	3900～4000

(*Performed by the WAKO PURE CHEMICAL INDUSTRIES, LTD.)

2. Conclusions: The test substance was identified as dichloromethane, by the mass spectrum and the infrared spectrum.

APPENDIX I 2

STABILITY OF DICHLOROMETHANE IN THE 2-WEEK INHALATION STUDY

CONCENTRATION OF DICHLOROMETHANE IN INHALATION CHAMBER

Group Name	Concentration(ppm) Mean ± S.D.
Control	0.0 ± 0.0
1,000ppm	1,007.8 ± 12.6
2,000ppm	2,010.3 ± 13.8
4,000ppm	4,027.3 ± 29.8
8,000ppm	7,955.6 ± 149.2
16,000ppm	16,014.1 ± 113.9

APPENDIX J 1

CONCENTRATION OF DICHLOROMETHANE IN THE INHALATION CHAMBER OF THE 2-WEEK INHALATION STUDY

STABILITY OF DICHLOROMETHANE IN THE 2-WEEK INHALATION STUDY

Lot No. APR5259

1. Sample: This lot was used from 1993.4.27 to 1993.5.10. Test substance was stored in a dark place at room temperature.

2. Infrared Spectrometry

Instrument : Hitachi 270-30 Infrared Spectrometer

Cell : KBr Liquid Cell

Slit : Medium

Results: The result of infrared spectrum did not change when before and after the lot of study.

1993.04.07(date analyzed)	1993.05.12(date analyzed)
Wave Number(cm^{-1})	Wave Number(cm^{-1})
430～ 480	430～ 480
650～ 840	650～ 840
870～ 940	870～ 940
970～1000	970～1000
1120～1180	1120～1180
1200～1340	1200～1340
1370～1500	1370～1500
1530～1570	1530～1570
1580～1630	1580～1630
2040～2090	2040～2090
2100～2190	2100～2190
2250～2360	2250～2360
2380～2460	2380～2460
2500～2550	2500～2550
2650～2730	2650～2730
2800～2860	2800～2860
2900～3200	2900～3200
3650～3730	3650～3730
3730～3800	3730～3800
3900～4000	3900～4000

3. Gas Chromatography

Instrument : Hewlett Packard 5890A Gas Chromatograph

Column : Methyl Silicone(0.2 mmϕ × 50 m)

Column Temperature : 60 °C

Flow Rate : 1 ml/min

Detector : FID(Flame Ionization Detector)

Injection Volume : 1 μl

Results: Gas chromatography indicated one major peak (peak No.1) and one impurity (peak No.2 < 1% of total area) analyzed at 1993.4.7 and one major peak (peak No.1) and one impurity (peak No.2 < 1% of total area) analyzed at 1993.5.12. No new trace impurity peak in the test substance analyzed at 1993.5.12 was detected.

Date (date analyzed)	Peak No.	Retention Time(min)	Area Count
1993.04.07	1	3.303	65203
	2	3.41	8
1993.05.12	1	3.305	64019
	2	3.407	10

4. Conclusions: The test substance was stable for about 5 weeks in a dark place at room temperature.

APPENDIX J 2

ENVIRONMENTAL CONDITIONS OF INHALATION CHAMBER IN THE 2-WEEK INHALATION STUDY OF DICHLOROMETHANE

ENVIRONMENTAL CONDITIONS OF INHALATION CHAMBER IN THE 2-WEEK INHALATION STUDY OF DICHLOROMETHANE

Group Name	Temperature(°C) Mean ± S.D.	Humidity(%) Mean ± S.D.	Ventilation Rate(L/min) Mean ± S.D.	Room Air Change(time/h) Mean
Control	21.9 ± 0.1	55.2 ± 0.6	103.8 ± 0.9	12.0
1,000ppm	21.9 ± 0.1	58.1 ± 0.6	103.9 ± 0.8	12.0
2,000ppm	21.6 ± 0.1	56.6 ± 0.5	103.1 ± 0.8	11.9
4,000ppm	21.8 ± 0.1	55.4 ± 0.4	104.6 ± 0.9	12.1
8,000ppm	22.8 ± 0.2	53.9 ± 0.6	104.1 ± 1.0	12.0
16,000ppm	21.6 ± 0.2	53.8 ± 0.9	104.3 ± 1.0	12.0

APPENDIX K 1

METHODS FOR HEMATOLOGY AND BIOCHEMISTRY IN THE 2-WEEK INHALATION STUDY OF DICHLOROMETHANE

METHODS FOR HEMATOLOGY AND BIOCHEMISTRY IN THE 2-WEEK INHALATION STUDY OF DICHLOROMETHANE

Item	Method
Hematology	
Red blood cell (RBC)	Light scattering method [1]
Hemoglobin (Hgb)	Cyanmethemoglobin method [1]
Hematocrit (Hct)	Calculated as RBC × MCV/10 [1]
Mean corpuscular volume (MCV)	Light scattering method [1]
Mean corpuscular hemoglobin (MCH)	Calculated as Hgb/RBC × 10 [1]
Mean corpuscular hemoglobin concentration (MCHC)	Calculated as Hgb/Hct × 100 [1]
Platelet	Light scattering method [1]
White blood cell (WBC)	Light scattering method [1]
Differential WBC	Pattern recognition method [2] (May-Grunwald-Giemsa staining)
Biochemistry	
Total protein (TP)	Biuret method [3]
Albumin (Alb)	BCG method [3]
A/G ratio	Calculated as Alb/(TP-Alb) [3]
T-bilirubin	Michaelson method [3]
Glucose	Enzymatic method (HK·G-6-PDH) [3]
T-cholesterol	Enzymatic method (CEH·COD·POD) [3]
Glutamic oxaloacetic transaminase (GOT)	UV·Rate method [3]
Glutamic pyruvic transaminase (GPT)	UV·Rate method [3]
Lactate dehydrogenase (LDH)	UV·Rate method [3]
Creatine phosphokinase (CPK)	UV·Rate method [3]
Urea nitrogen	Enzymatic method (Urease·GLDH) [3]
Sodium	Flame photometry [4]
Potassium	Flame photometry [4]
Chloride	Coulometric titration [4]
Calcium	OCPC method [3]
Inorganic phosphorus	Enzymatic method (SPL·PGM·G-6-PDH) [3]

1) Automatic blood cell analyzer (Technicon H·1 : Technicon Instruments Corporation,USA)

2) Automatic blood cell differential analyzer (Hitachi 8200 : Hitachi,Ltd.,Japan)

3) Automatic analyzer (Hitachi 705 : Hitachi,Ltd.,Japan)

4) Flame photometer (Hitachi 750 : Hitachi,Ltd.,Japan)

APPENDIX K 2

UNISTS AND DECIMAL PLACEFOR HEMAYOLOGY AND BIOCHEMISTRY IN THE 2-WEEK INHALATION STUDY OF DICHLOROMETHANE

UNITS AND DECIMAL PLACE FOR HEMATOLOGY AND BIOCHEMISTRY IN THE 2-WEEK INHALATION STUDY OF DICHLOROMETHANE

Item	Unit	Decimal place
Hematology		
Red blood cell (RBC)	$\times 10^6 / \mu L$	2
Hemoglobin	g/dL	1
Hematocrit	%	1
Mean corpuscular volume (MCV)	fL	1
Mean corpuscular hemoglobin (MCH)	pg	1
Mean corpuscular hemoglobin concentration (MCHC)	g/dL	1
Platelet	$\times 10^3 / \mu L$	0
White blood cell (WBC)	$\times 10^3 / \mu L$	2
Differential WBC	%	0
Biochemistry		
Total protein	g/dL	1
Albumin	g/dL	1
A/G ratio	-	1
T-bilirubin	mg/dL	2
Glucose	mg/dL	0
T-cholesterol	mg/dL	0
Glutamic oxaloacetic transminase (GOT)	IU/L	0
Glutamic pyruvic transaminase (GPT)	IU/L	0
Lactate dehydrogenase (LDH)	IU/L	0
Creatine phosphokinase (CPK)	IU/L	0
Urea nitrogen	mg/dL	1
Sodium	mEq/L	0
Potassium	mEq/L	1
Chloride	mEq/L	0
Calcium	mg/dL	1
Inorganic phosphorus	mg/dL	1