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Study Number	B11-1054

FINAL REPORT

TWENTY-EIGHT-DAY REPEATED-DOSE ORAL TOXICITY STUDY OF 13F-SFA IN RATS

April, 2014

Chemicals Evaluation and Research Institute, Japan, Hita

GLP STATEMENT

Chemicals Evaluation and Research Institute, Japan, Hita

Sponsor: DAIKIN INDUSTRIES, LTD.

Title: Twenty-Eight-Day Repeated-Dose Oral Toxicity Study of 13F-SFA in Rats

Study Number: B11-1054

This study was conducted in compliance with the following GLP principles.

OECD Principles of Good Laboratory Practice (November 26, 1997)

I confirmed that this report accurately reflects the raw data obtained and that data of the study have reliability.

Study Director: April 16, 2014

Date

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QUALITY ASSURANCE STATEMENT

1. TITLE

Twenty-Eight-Day Repeated-Dose Oral Toxicity Study of 13F-SFA in Rats

2. SPONSOR

Name: DAIKIN INDUSTRIES, LTD.

Address: 1-1, Nishi Hitotsuya, Settsu-shi, Osaka 566-8585, Japan

3. TESTING FACILITY

Name: Chemicals Evaluation and Research Institute, Japan, Hita (CERI Hita)

Address: 3-822 Ishii-machi, Hita-shi, Oita 877-0061, Japan

4. PURPOSE OF STUDY

A twenty-eight-day repeated-dose oral toxicity study of 13F-SFA in rats was performed in CERI Hita (study number B11-0836, GLP). In this study, the test substance was treated by gavage at the dose level of 0, 5, 25 and 125 mg/kg/day. Kidney weights were increased in the 25 mg/kg and higher dose groups and the histopathological changes were observed in the liver and kidney in the 125 mg/kg group. The No-Observed-Adverse-Effect Level (NOAEL) was estimated to be 5 mg/kg/day (Shiraishi, etc, 2007). The purpose of the present study is to confirm the presence or absence of the toxic effects of the test substance at 30 mg/kg/day and reproducibility the test results.

5. TESTING METHOD

This study was conducted partially in accordance with the "Repeated Dose 28-day Oral Toxicity Study in Rodents" (No. 407, Adopted: October 3, 2008) prescribed in the "OECD Guideline for The Testing of Chemicals".

6. GLP COMPLIANCE

OECD Principles of Good Laboratory Practice (November 26, 1997)

7. ANIMAL WELFARE

This study complied with the guideline for the animal experiment at CERI Hita that was made by reference to the following act and guidelines.

a) Act on Welfare and Management of Animals (Japan, Act Number 105, 1973)

b) Standards Relating to the Care and Management of Laboratory Animals and Relief of Pain (Ministry of the Environment, Japan, 2006)

c) Fundamental Guidelines for Proper Conduct of Animal Experiment and Related Activities in Academic Research Institutions under the jurisdiction of the Ministry of Health, Labour and Welfare (Ministry of Health, Labour and Welfare, Japan, 2006)

d) Fundamental Guidelines for Proper Conduct of Animal Experiment and Related Activities in Academic Research Institutions under the jurisdiction of the Ministry of Agriculture, Forestry and Fisheries (Ministry of Agriculture, Forestry and Fisheries, Japan, 2006)

e) Fundamental Guidelines for Proper Conduct of Animal Experiment and Related Activities in Academic Research Institutions (Ministry of Education, Culture, Sports, Science and Technology, Japan, 2006)

f) Guidelines for Proper Conduct of Animal Experiments (Science Council of Japan, 2006)

8. STUDY SCHEDULE

Study Initiation	November 28, 2013
Animal Receipt	December 3, 2013
Initiation of Examination (Initiation of dosing)	December 10, 2013
Necropsy	January 7, 2014
Termination of Examination (Termination of pathological examination)	March 25, 2014
Study Completion	April 15, 2014

9. STUDY DIRECTOR

Katsumi Miyata Section 2, CERI Hita

10. PERSONNEL CONCERNED WITH STUDY

Study Staff:

(Responsible for the animal examinations: quarantine, acclimation, care and management of animals, preparation and administration of the test substance, general clinical observations, body weight and food consumption measurements)

Pathological Examinations:

(Responsible for the pathological examinations: necropsy, collection of tissues, organ weight measurements and histopathological examinations)

Clinical Examinations:

(Responsible for the clinical examinations: blood chemical examinations)

Chemical Analysis:

(Responsible for the analysis examinations: identity and stability confirmation of the test substance, concentration analysis of the test substance formulations)

Other Staffs:

(Animal examinations)

(Pathological examinations)

Junichi Kikuchi
(Clinical examinations)

11. STORAGE AND RETENTION PERIOD OF RAW DATA AND SPECIMENS

The original study plan and the amendment, the original final report, raw data, study contract documents, test substance information, other record documents, specimens and samples of the test substance is retained in CERI Hita. The retention period is 10 years after the completion of the study. Stability of the test substance during the retention period is not confirmed. After the retention period, any measures (continued storage, disposal or return) are settled upon the consultation with the sponsor.

12. APPROVAL BY STUDY DIRECTOR

Study Director: April 16, 2014

Date

13. SUMMARY

A twenty-eight-day repeated-dose oral toxicity study of 13F-SFA in rats was performed in CERI Hita (study number B11-0836, GLP). In this study, the test substance was treated by gavage at the dose level of 0, 5, 25 and 125 mg/kg/day. Kidney weights were increased in the 25 mg/kg and higher dose groups. The No-Observed-Adverse-Effect Level (NOAEL) was estimated to be 5 mg/kg/day. In the present study the test substance was administered to rats for 28 days to confirm the presence or absence of the toxic effects at 30 mg/kg/day and reproducibility the test results.

The test substance was dissolved with olive oil including 0.5 w/v% polyoxyethylene (20) sorbitan monooleate (Tween80). Dose levels were 0 (olive oil with 0.5 w/v% Tween80), 30 and 120 mg/kg/day. The test substance was administered to five males and five females of Crl:CD(SD) rats in each dose level. All animals were observed for general clinical observation, and their body weights and food consumption were measured. Blood samples were taken at the next day of the last dosing and blood chemical examinations and pathological examinations were performed.

Salivation was transiently observed just after dosing in males of the 30 and 120 mg/kg groups and females of the 120 mg/kg group, or just before dosing in one male of the 120 mg/kg group. Total bilirubin was increased in males of the 120 mg/kg group. Absolute weight of the liver was increased in males and females of the 120 mg/kg group. Relative weight of the liver was increased in males of the 30 and 120 mg/kg groups and females of the 120 mg/kg group. Hypertrophy of the hepatocytes was observed in males and females of the 120 mg/kg group. Kidney weights were increased in females of the 120 mg/kg group without histopathological lesions. The NOAEL of the 13F-SFA was 30 mg/kg/day under the conditions tested since adverse effects were not noted in the 30 mg/kg group.

14. MATERIALS

14.1 TEST SUBSTANCE

a) Name, etc (Information provided by the sponsor)

Chemical name: 3,3,4,4,5,5,6,6,7,7,8,8,8-tridecafluorooctyl=acrylate

Other name: 13F-SFA

CAS number: 17527-29-6

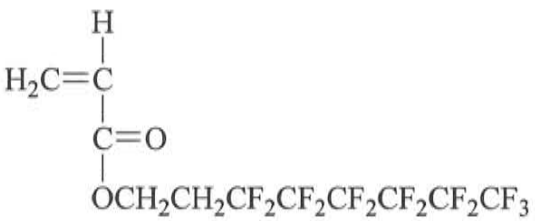
b) Supplier and Lot No. (Information provided by the sponsor)

Supplier: DAIKIN INDUSTRIES, LTD.

Lot number: 6SFAD31003

c) Structural Formula, etc (Information provided by the sponsor)

Structural formula:



Molecular formula: $C_{11}H_7F_{13}O_2$

Molecular weight: 418.15

d) Purity, etc (Information provided by the sponsor)

Purity: 99.8%

Impurities: unknown component 0.2%

The test substance was treated with as 100% in purity.

e) Physicochemical Properties (Information provided by the sponsor)

Appearance at normal temperature: colorless clear liquid

Boiling point: 78°C (8 mmHg)

Solubility to solvent, etc

Solvent	Solubility
Dimethylsulfoxide	Soluble (arbitrary mixable)
Acetone	Soluble (arbitrary mixable)

Density: 1.554 g/cm^3 (25°C)

Hydrolyzability: hydrolyzable

CH_2=CHCOO$CH_2CH_2C_6F_{13}$ → $C_6F_{13}CH_2CH_2OH$ + CH_2=CHCOOH

f) Storage

The test substance was stored in a dark airtight container at room temperature in the cabinet number 1 or 5 in the test substance storage room.

Actual temperature: 17.2-21.2°C (tolerance temperature: 10-30°C)

Storage periods: October 21, 2013 to December 31, 2013 (from receipt of the test substance to the final preparation of the dosing formulations)

g) Identity and Stability under Storage Condition (Fig. 1)

Identity of the test substance was confirmed by comparing of infrared (IR) spectra between measured before the dosing period in CERI Hita and provided by the sponsor. The IR spectrum measured before the dosing period was identical with those provided by the sponsor.

Stability of the test substance during the dosing period was confirmed by comparing the IR spectra between measured before and after the dosing period. The test substance was judged to be stable during the dosing period under the storage condition since there were no differences in the IR spectra before or after the dosing period.

Instrument: IR spectrophotometer FT-720 (HORIBA)

Wave number: 4000 cm^{-1} - 400 cm^{-1}

Pre-Treatment: Potassium bromide neat

h) Handling Precaution

In order to avoid inhalation and contact with the skin and eyes, chemically resistant gloves, a mask, a head cap, safety glasses and a lab coat were worn.

14.2 VEHICLE

a) Name

Olive oil containing 0.5% polyoxyethylene (20) sorbitan monooleate (Tween80)

b) Reason of Selection

In the previous study (study number B11-0836) olive oil was selected. In the present study preparation method of the dosing formulation was confirmed and the test substance dissolved in the same vehicle at concentration of 1.20 w/v%.

c) Manufacturer, Grade, Lot number and Storage of Vehicle.

	Manufacturer	Grade	Lot number	Storage place	Storage temperature
Olive oil	Taisei Pharmaceutical Industries	Japanese pharmacopoeia	308019	Reagent storage room	Room temperature
Tween80	Wako Pure Chemical Industries	Chemistry grade	TLN6911	Refrigerator 7 in the test substance storage room	Refrigeration

14.3 ANIMALS

Crl:CD(SD) rats (SPF) were obtained from Charles River Japan Hino Breeding Center. This strain is established as a laboratory animal and widely used in a general toxicity study, and we have historical data of this strain.

Eighteen males and eighteen females were obtained at 4 weeks old. The animals were quarantined and acclimatized for 6 days under group housing of three or five per cage. No abnormalities were noted in any animals during the quarantine or acclimation periods. The animals were allocated to groups using body weight-stratified randomization using body weights measured on 6 days after reception. The animals were housed individually after grouping. Body weights of each animal were in the range of ±20% of the mean body weights at grouping. The animals not treated were excluded from the study. Clinical conditions and excretions were observed once or more a day from the reception to the start of dosing. The animals were identified by means of a marker on the tail before grouping and ear-tags for after grouping. Cages and racks were identified by individual labels and indications of study number and dose levels, respectively. At the onset of treatment the animals were 5 weeks old with body weight ranges of 125.3-149.1 g and 119.1-131.9 g, for males and females respectively.

14.4 HOUSING CONDITIONS

The barrier-system animal rooms (quarantine room 1 and animal room 1) were maintained at a stable temperature (21-25°C) and relative humidity (40-70%) with 10-15 air changes per hour and artificial light-dark cycle of 12-12 hours (light on: 7:00 and light off: 19:00). The actual temperature and humidity were 22.5-24.1°C and 48.2-62.8%, respectively. The animals were housed in a hanging stainless steel cage with wire-mesh floor at the size of 260 W×380 D×180 H mm for quarantine and acclimation periods, and a hanging stainless cage with wire-mesh floor at the size of 165 W×300 D×150 H mm for after grouping. Undertrays were changed at the end of the quarantine period and at grouping, and twice a week after grouping and before carrying animals from the animal room on the day of autopsy. Feeders, cages and racks were changed once at grouping.

The animals had free access to a pelleted diet (MF, lot number 131004, Oriental Yeast) and chlorinated Hita city supply by an automatic watering system, maintained at 3-5 ppm of chloric level prepared by adding sodium hypochlorite (Purelox). The diets and housing materials were autoclaved at 121°C for 30 min prior to use. Information of the contaminants in the diets was obtained from supplier and confirmed to meet the requirements in our laboratory which referred to the "Toxic Substances Control Act of US-EPA" (1979). Contaminants in drinking water were analyzed twice a year according to the water regulations of the "Ordinance on drinking water quality standards" (ordinance number 101 of Japan's MHLW). The analytical data of contaminants was in the stated ranges of CERI Hita.

15. METHODS

15.1 DOSE SETTING

In the previous study of 13F-SFA performed in CERI Hita (study number B11-0836, GLP) the test substance was administered at dose levels of 0, 5, 25 and 125 mg/kg/day for 28 days to five male and five female Crl:CD(SD) rats at 5 weeks of age in each dose group During the dosing period general clinical observations, detailed clinical observations, sensorimotor function, body weight and food intakes measurements were performed. After the dosing period urinalyses, blood examinations and pathological examinations were performed. In this study increased aspartate aminotransferase, alanine aminotransferase and alkaline phosphatase, and decreased total cholesterol were noted in males of the 125 mg/kg group. In females of the same dose group, increased γ-glutamyl transpeptidase, triglyceride, albumin, A/G ratio and total bilirubin, and decreased cholinesterase was noted. In the organ weights, relative weights of the liver were increased in male and female of the 125 mg/kg group. In the histopathological examinations, periportal hypertrophy, periportal prominent nucleoli and single cell necrosis of the hapatocytes, and dilatation of the renal tubules were observed in males of the 125 mg/kg group. Diffuse hypertrophy of the hepatocytes and ballooning of the epithelium of the renal tubules were observed in females of the 125 mg/kg group. The test substance was considered to affect the liver and kidney. The incisor was also affected in males and females of the 125 mg/kg group. The No-Observed-Adverse-Effect Level was estimated to be 5 mg/kg/day based on increased relative weights of the kidneys in males of the 25 mg/kg and higher dose groups.

In the present study, dose levels were selected at 120 and 30 mg/kg/day to confirm the presence or absence of the toxic effects at 30 mg/kg/day and reproducibility of the test results.

15.2 GROUP ALLOCATION

Two dose levels for the test substance groups and one vehicle control group were selected. Vehicle control group is showed as the control group from this section.

Group		Dose (mg/kg/day)	Volume (mL/kg)	Concentration of formulation (w/v%)	Number of Animals (Animal Number)	
					Male	Female
Vehicle control		0	10	0	5 (1 - 5)	5 (16 - 20)
Test substance	Low dose	30	10	0.300	5 (6 - 10)	5 (21 - 25)
	High dose	120	10	1.20	5 (11 - 15)	5 (26 - 30)

15.3 FORMULATIONS

a) Preparation of the Vehicle

Tween80 was weighed at final concentration of 0.5 w/v% and dissolved by stirring with a magnetic stirrer while adding olive oil.

b) Preparation and Storage of Formulations

The test substance was weighed and vehicle was added, and mixed with homogenizer (ULTRA-TURRAX T25 basic, IKA WERKE). Vehicle was added to prepare 1.20 w/v% formulation. A part of this formulation was taken and diluted with vehicle to prepare 0.300 w/v% formulation. These formulations were subdivided into plastic containers and stored at a cold place (2-7°C, tolerance temperature 1-10°C, refrigerator number 7 in the test substance preparation room). On each dosing day formulations and vehicle were taken out from the storage place and dosed to the animals. The formulations were used within 8 days after preparation.

No. of preparation and date	Concentrations (w/v%)	Test substance weights and dilution methods	Total volume (mL)
1st December 9, 2013 (Concentration analysis)	1.20 0.300	2.4028 g Diluted 45 mL of 1.20 w/v% with vehicle	200 180
2nd December 16, 2013	1.20 0.300	3.0061 g Diluted 50 mL of 1.20 w/v% with vehicle	250 200
3rd December 24, 2013	1.20 0.300	3.6023 g Diluted 62.5 mL of 1.20 w/v% with vehicle	300 250
4th December 31, 2013	1.20 0.300	3.6033 g Diluted 62.5 mL of 1.20 w/v% with vehicle	300 250

c) Homogeneity and Stability Analyses of the Formulations

In the previous study, stability of the test substance, homogeneity, stability and concentration of the formulation were conformed (study number X18-0836, GLP). Formulations of 1.25 and 0.05 w/v% were homogeneous and stable for 8 days under the storage condition.

d) Concentration Analysis

Concentrations of 1.20 and 0.300 w/v% formulations were analyzed using gas chromatography (GC) immediately after the first preparation. Analytical method was decided according to the result of validation of the analytical method (study number X18-0836, non-GLP). Concentrations of all formulations were confirmed to be within 100±10% of each nominal concentration.

Date of analysis	Nominal concentration	Actual concentration	R.N.
December 9, 2013	1.20 w/v%	1.24 w/v%	103%
	0.300 w/v%	0.305 w/v%	102%

R.N.: Rate to the nominal concentration

1) Pre-Treatment

A sample (n=1) was pre-treated after collection from each formulation at middle layer.

(1) 1.20 w/v% Formulation

Accurate 0.5 mL of the formulation was diluted with ethyl acetate to fill the volume of 20 mL. Accurate 0.5 mL of this solution was diluted with ethyl acetate to fill the volume of 15 mL. This solution was served as the analytical sample (dilution rate: 1200).

(2) 0.300 w/v% Formulation

Accurate 0.5 mL of the formulation was diluted with ethyl acetate to fill the volume of 15 mL. Accurate 0.5 mL of this solution was diluted with ethyl acetate to fill the volume of 5 mL. This solution was served as the analytical sample (dilution rate: 300).

2) Preparation of Standard Solution

The test substance (approx. 0.1 g) was weighed accurately and brought to a volume of 100 mL with ethyl acetate to prepare the approx. 1000 μg/mL standard stock solution. Accurate 2 mL of this was diluted with ethyl acetate to fill the volume of 20 mL to make the approx. 100 μg/mL standard solution. Accurate 2 mL of this was diluted with ethyl acetate to fill the volume of 20 mL to make the approx. 10 μg/mL standard solution.

3) Analytical Conditions

(1) Instruments

Gas chromatograph:	HP6890 Series (Yokogawa Analytical Systems)
Controller:	G1512A (Yokogawa Analytical Systems)
Autosampler:	18596C (Yokogawa Analytical Systems)
Injector:	18593B (Yokogawa Analytical Systems)
Data processor:	GC-Chemstation (Yokogawa Analytical Systems)

(2) Conditions

Column:	HP-1ms (F.T. 0.25 μm, 0.25 mm I.D.×30 m, Agilent Technologies)
Column oven temperature:	100°C
Injection port temperature:	250°C
Carrier gas:	Helium
Control mode:	constant flow
Carrier gas flow rate:	1.0 mL/min
Detector:	FID
Detector temperature:	250°C
Injection method:	split (split ratio 10:1)
Injection volume:	2 μL

4) Data Processing

(1) Detection value and quantitative analytical method

A peak area was used as a detection value.

In the validation of the analytical method, a straight line which passed through the origin of the coordinates was obtained. Therefore concentrations of the analytical samples were determined by a single level calibration.

(2) Calculation of test substance concentration and relative rate

Concentrations of the test substance in formulations (C: w/v%) were rounded off to three significant figures.

$$C = \frac{Cs \times A \times D}{As \times 10000}$$

Cs: test substance concentration in standard solution (μg/mL)

As: detection value of the test substance for standard solution

A: detection value of the test substance for each analytical sample

D: dilution rate in each analytical sample

Relative rates to the nominal concentrations (R.N.: %) were rounded off to three significant figures.

$$\text{R.N.} = \frac{\text{Actual concentration}}{\text{Nominal concentration}} \times 100$$

15.4 ADMINISTRATION

The test substance was administered daily by gavage between 9:00 and 11:34 for 28 days according to the testing method. A syringe (Terumo) connected to a Nelaton catheter (Terumo) was used. Dosing volume was 10 mL/kg based on the latest body weights.

15.5 GENERAL CLINICAL OBSERVATIONS

During the dosing period all animals were observed twice a day (before dosing and after dosing).

15.6 BODY WEIGHT MEASUREMENTS

Body weights were measured using an electric balance (SARTORIUS) for all animals on days 1, 3, 8, 12, 17, 21, 26 and 28 of the dosing period, and on the necropsy days (before carrying animals from the animal room)

15.7 FOOD CONSUMPTION MEASUREMENTS

Food weights were measured using an electric balance (SARTORIUS) for all animals at the group allocation, and on days 1, 3, 8, 15, 22 and 28 of the dosing period. Feeding weights on days 8, 15 and 22 were measured after food was replenished. Mean food consumption per day was calculated from their feeding and remainder weights.

15.8 BLOOD EXAMINATIONS

a) Blood Sampling and Preparation

Blood was collected from the abdominal aorta under isoflurane anesthesia. Blood samples were collected in a glass tube and centrifuged at 3000 r.p.m. for 10 min after overnight fasting (16 to 20 hours).

b) Blood Chemical Examinations

Serum samples were used to examine the following parameters.

Parameters	Methods	Instruments
Aspartate aminotransferase (AST)	UV method (method based on JSCC)	A
Alanine aminotransferase (ALT)	UV method (method based on JSCC)	
Alkaline phosphatase (ALP)	*p*-Nitrophenyl phosphate method	
γ-Glutamyl transpeptidase (γ-GTP)	*L*-γ-glutamyl-3-carboxy-4-nitroanilide method	
Total cholesterol (T-Cho)	COD·ESPAS method	
Triglyceride (TG)	GPO·ESPAS glycerol blocking Method	
Blood urea nitrogen (BUN)	Urease·GlDH method	
Creatinine	Creatininase·F-DAOS method	
Total protein (T-Protein)	Biuret method	
Albumin	Bromocresol green method	
A/G ratio	$\frac{\text{Albumin}}{\text{T - Protein} - \text{Albumin}}$	-
Glucose	Hexokinase·G-6-PDH method	A
Total bilirubin (T-Bil)	Enzyme method	
Total bile acids (TBA)	Enzyme cycling method	
Inorganic phosphorus (IP)	Fiske-Subbarow method	
Calcium (Ca)	OCPC method	
Sodium (Na)	Crown-Ether membrane electrode method	B
Potassium (K)	Crown-Ether membrane electrode method	
Chloride (Cl)	MO membrane method	

A: Clinical chemistry analyzer, Model 7170 (Hitachi)

B: Electrolyte analyzer, PVA-EX II (A&T)

15.9 PATHOLOGICAL EXAMINATIONS

a) Gross Necropsy

All animals were subjected to a detailed gross necropsy after blood sampling and bleeding from the ventral aorta on the next day of last dosing. External surface of the body, all orifices, subcutis, cranial, thoracic, abdominal and pelvic cavities, and their contents were observed.

b) Tissue Collecting and Organ Weight Measurements

The liver and kidneys were removed and their weights were measured using an electric balance (SARTORIUS) for all animals. The kidneys were weighed right and left together. Relative organ weights of these organs were calculated based on the body weights measured on the necropsy day.

c) Histopathological Examinations

The liver and kidneys were preserved in 10% neutralized buffered formalin followed by embedding in paraffin, sectioning and hematoxylin and eosin (HE) staining. Livers in males and females of all groups were examined since treatment related change was suspected in the organ weights. Kidneys in males and females of all groups were examined since treatment related change was suspected in the organ weights of females together with the study objective of this study.

15.10 STATISTICAL ANALYSIS

Data regarding body weights, food consumption, blood chemical examinations and organ weights were analyzed by the Bartlett's test for homogeneity of variances. When the variances were homogeneous at a significance level of 5%, the Dunnett's test was used. When the variances were not homogeneous, the nonparametric Dunnett's test was used.

16. DEVIATION FROM THE PROTOCOL

There were no deviations from the protocol.

17. RESULT

17.1 GENERAL CLINICAL OBSERVATIONS

(Table 1, Appendix 1)

In males, salivation was transiently observed just after dosing in three of five of the 30 mg/kg group and four of five of the 120 mg/kg group. One animal salivated just before dosing in the 120 mg/kg group on day 11.

In females, salivation was transiently observed just after dosing in three of five of the 120 mg/kg group.

17.2 BODY WEIGHTS

(Fig. 2, Table 2, Appendix 2)

No abnormalities were noted in either sex of any treatment groups.

17.3 FOOD CONSUMPTION

(Fig. 3, Table 3, Appendix 3)

Food consumption was not affected in either sex of any treatment groups.

17.4 BLOOD EXAMINATIONS

(Table 4, Appendix 4)

In males, total bilirubin was increased in the 120 mg/kg group. No abnormal changes were noted in the 30 mg/kg group.

In females, no abnormalities were noted in any treatment groups.

17.5 PATHOLOGICAL EXAMINATIONS

a) Organ Weights (Tables 5 and 6, Appendices 5 and 6)

In males, absolute weight of the liver was increased in the 120 mg/kg group (130% of the control group). Relative weights of the liver were increased in the 30 mg/kg group (114% of the control group) and 120 mg/kg group (125% of the control group).

In females, absolute weight (135% of the control group) and relative weight (136% of the control group) of the liver were increased in the 120 mg/kg group. Relative weights of the kidneys (111% of the control group) were increased in the 120 mg/kg group. Abnormal weight changes were not noted in the 30 mg/kg group.

b) Gross Necropsy (Table 7, Appendix 7)

In males, mottled teeth and enlargement of the liver were observed in each three of five of the 120 mg/kg group.

In females, mottled teeth in three and enlargement of the liver in four were observed in five of the 120 mg/kg group.

c) Histopathological Examinations (Table 8, Appendix 7)

In males, periportal hypertrophy of the hepatocytes was observed in three of five of the 120 mg/kg group. Microgranuloma in the liver was observed in each one of the 30 and 120 mg/kg groups, and solitary cyst in the medulla of the kidney was observed in each one of the control and 120 mg/kg groups without dose-relationship.

In females, diffused hypertrophy of the hepatocytes was observed in four of five of the 120 mg/kg group. Microgranuloma was observed in the liver in one of the 120 mg/kg group as a spontaneous lesion. No abnormal changes were noted in the 30 mg/kg group.

18. DISCUSSION

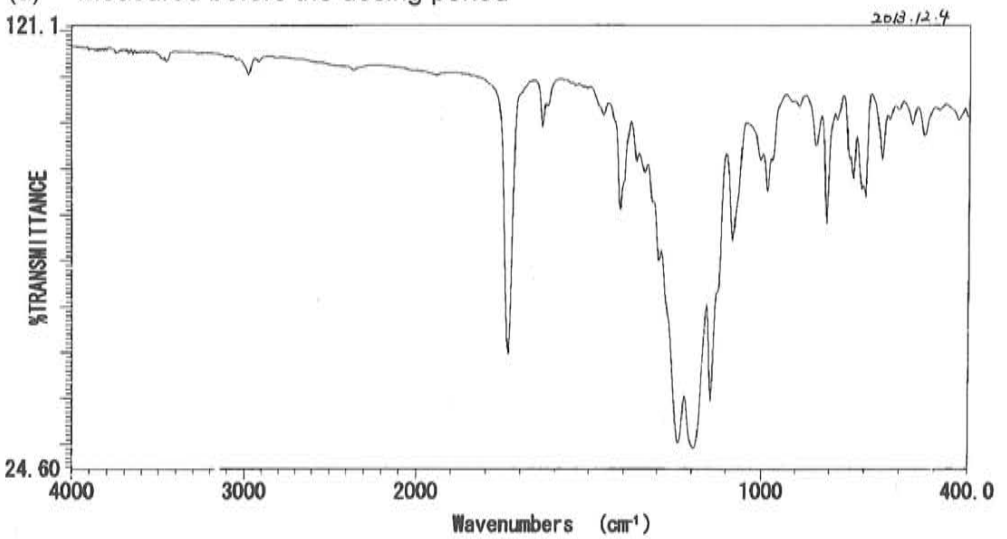
In the previous study, hypertrophy of the hepatocytes and ballooning of the epithelium of the renal tubules were observed in the 125 mg/kg group. The NOAEL was estimated to be 5 mg/kg/day since kidney weights were increased in the 25 mg/kg and higher dose groups.

In the present study, liver weights were increased in the 30 and 120 mg/kg groups, and hypertrophy of the hepatocytes was observed in the 120 mg/kg group; however, no histopathological change was noted in the 30 mg/kg group. Althoguh the kidney weights were increased in the 120 mg/kg group, histopathological changes were not noted at any dose levels. From these results the NOAEL of the 13F-SFA was 30 mg/kg/day since no adverse effects were noted in the 30 mg/kg group.

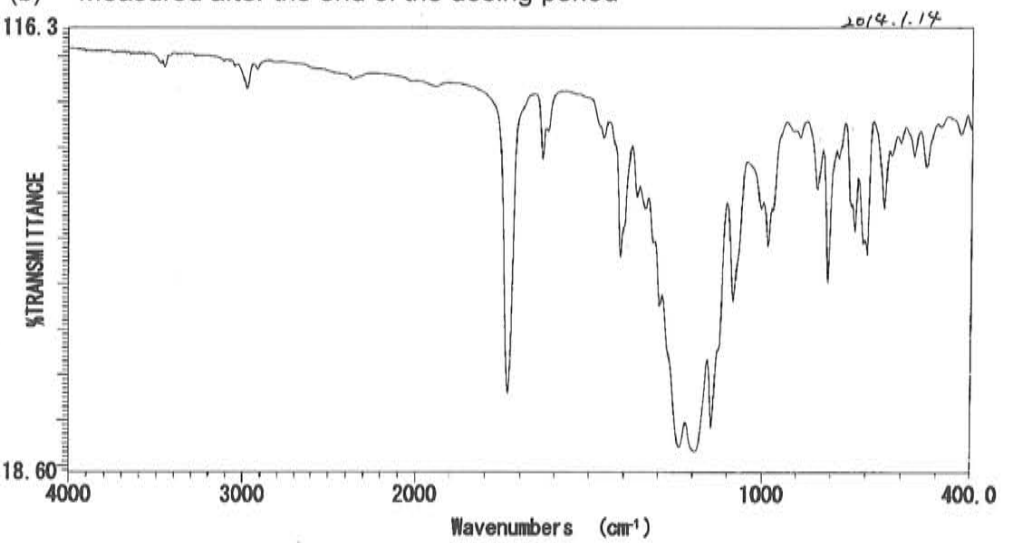
19. REFERENCE

Shiraishi et al, (2007) Twenty-Eight-Day Repeated-Dose Oral Toxicity Study of 13F-SFA in Rats, Final repot (study number B11-0836) unpublished.

(a) Measured before the dosing period



(b) Measured after the end of the dosing period



(c) Provided by the sponsor

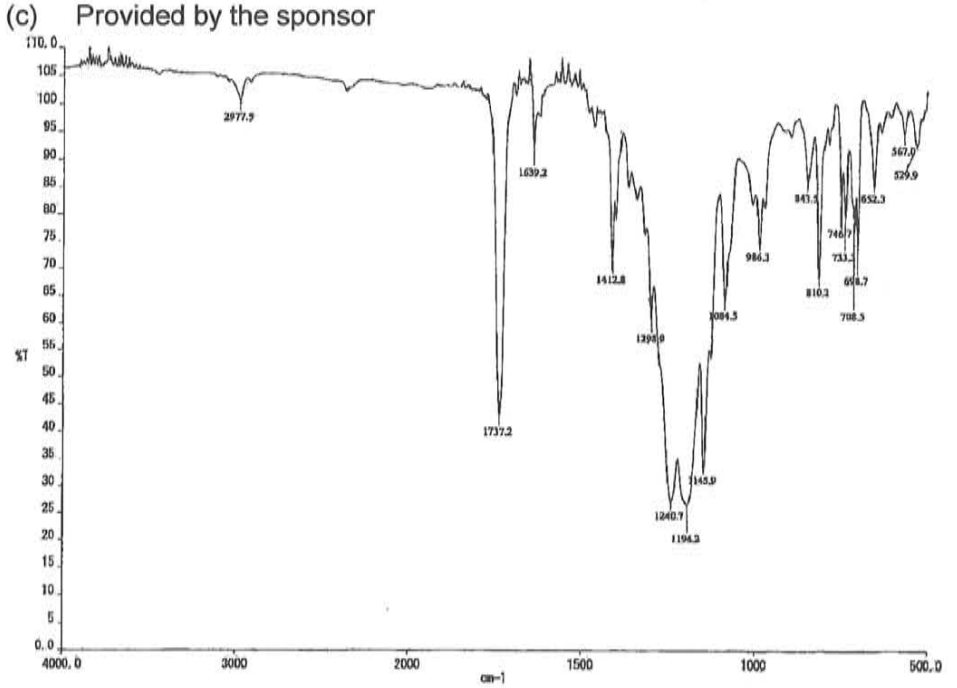


Fig. 1 IR spectrum of the test substance

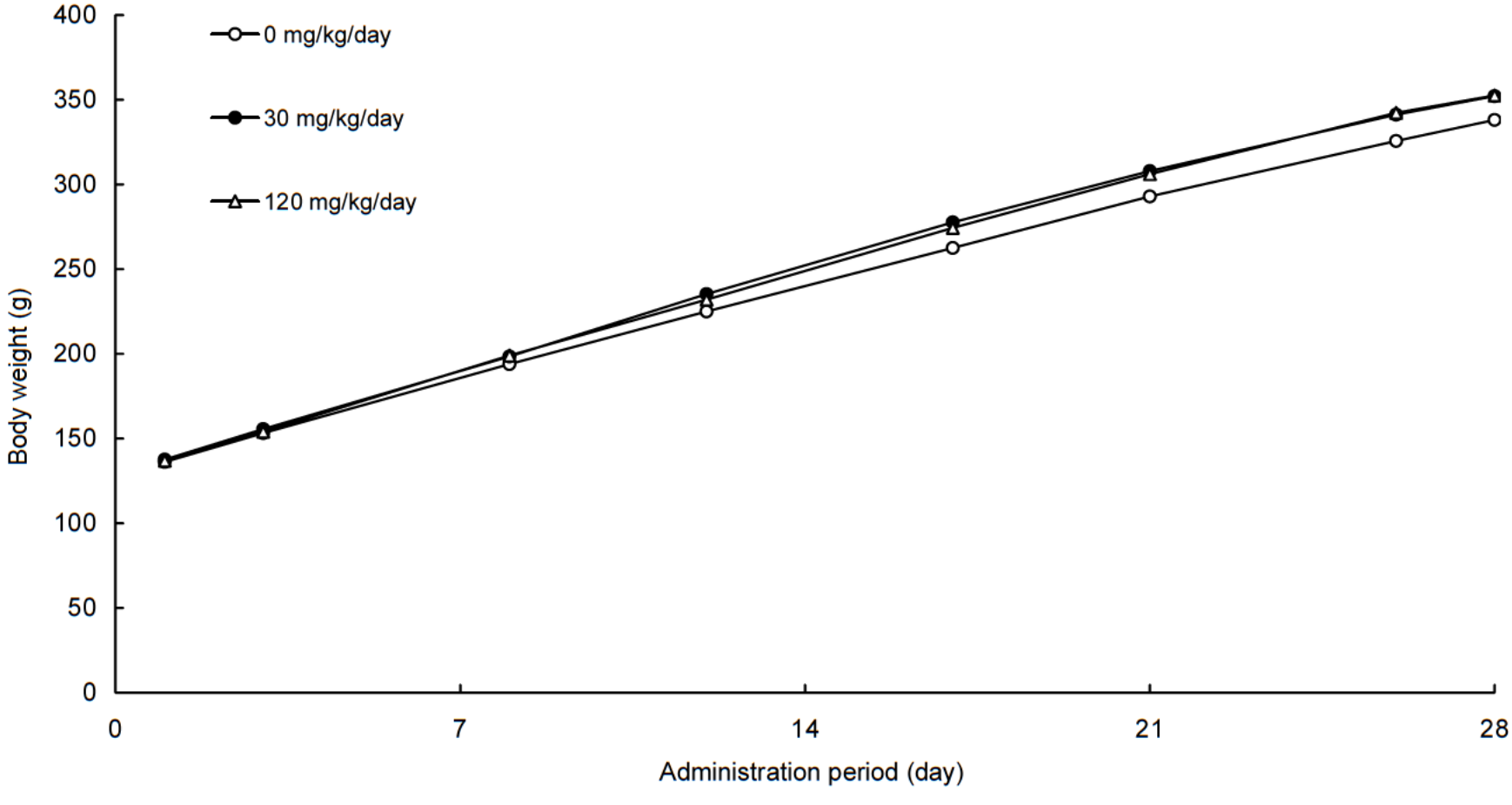


Fig. 2-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Body weights: Male

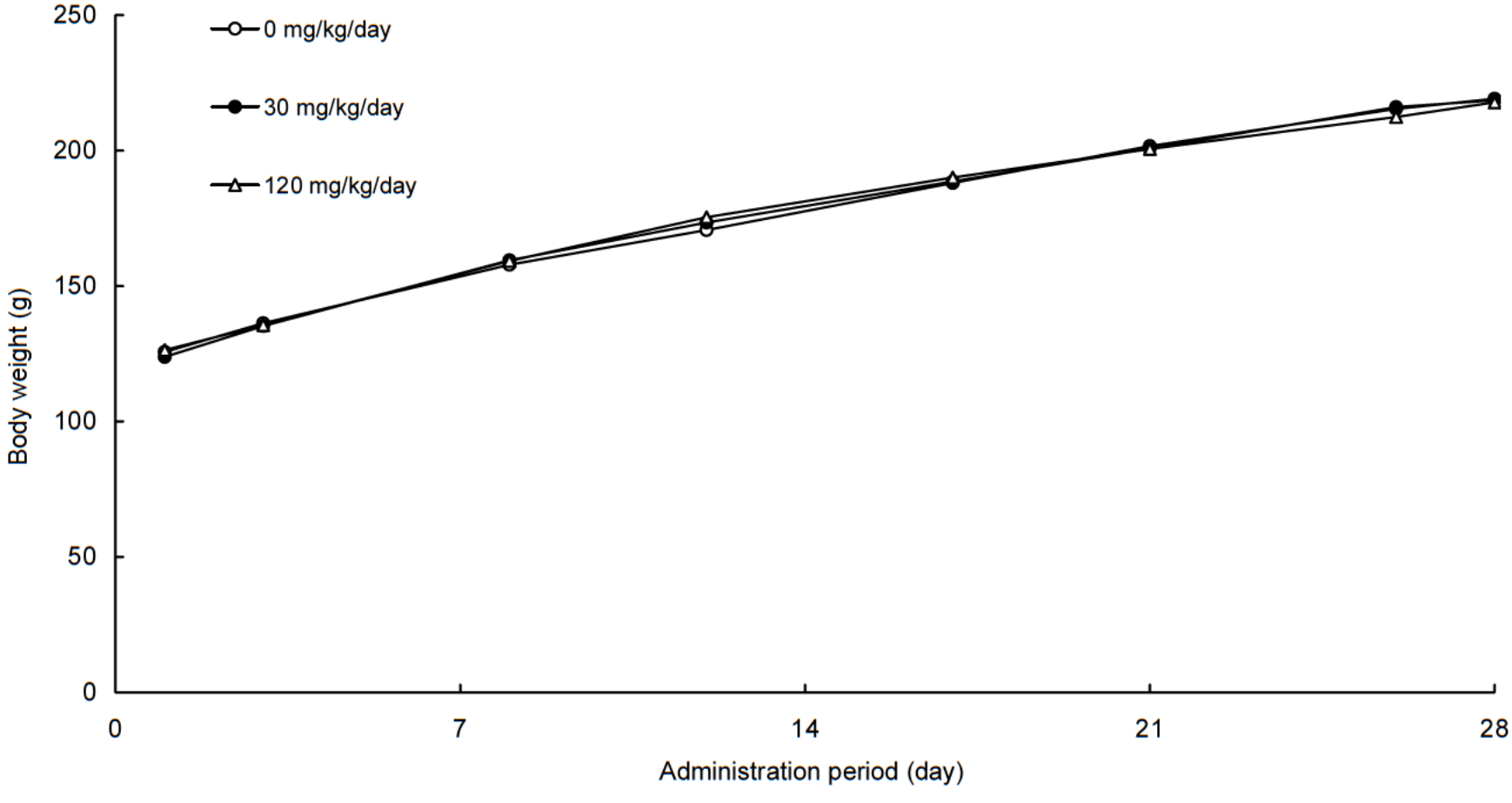


Fig. 2-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Body weights: Female

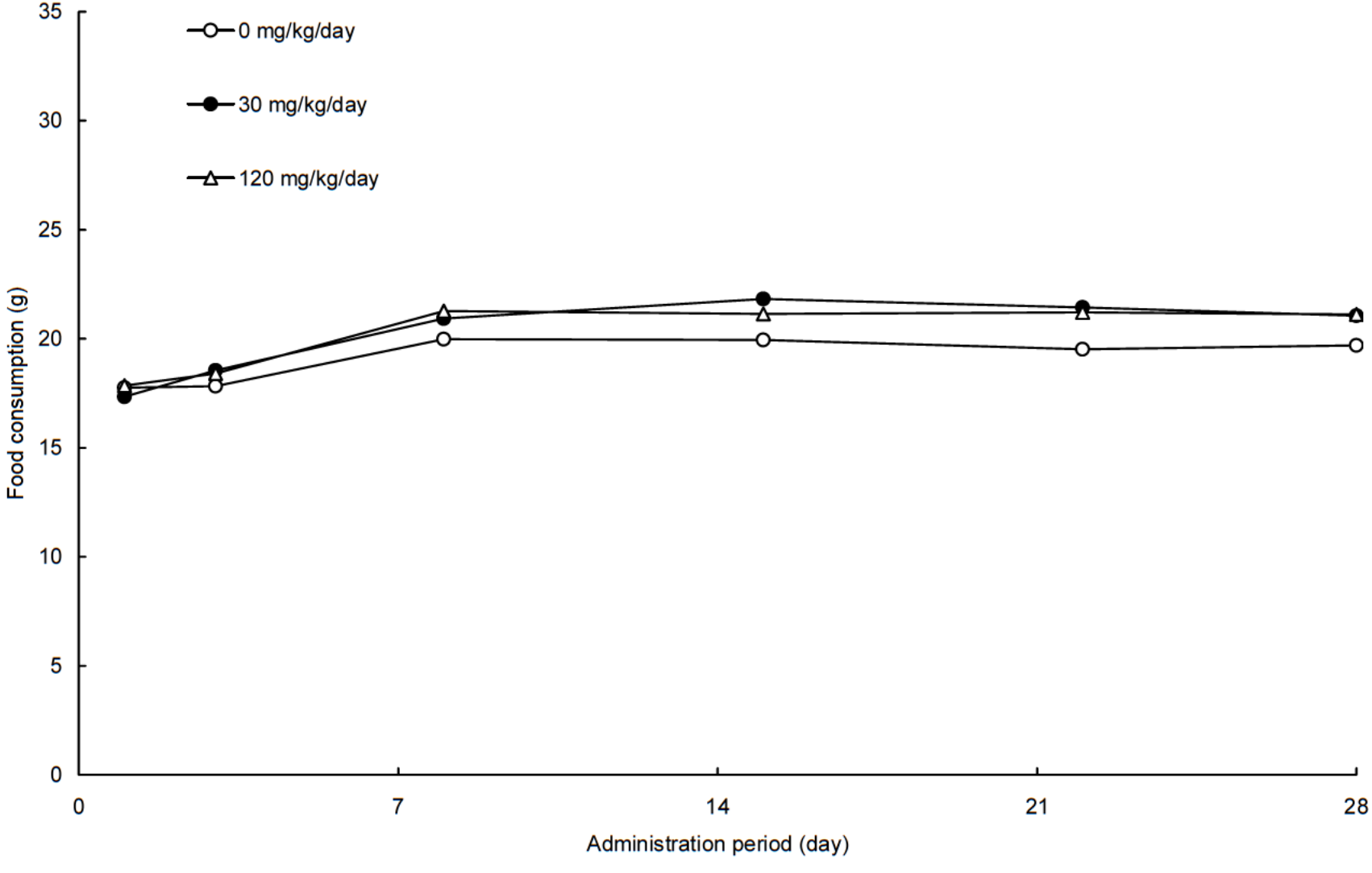


Fig. 3-1 Twenty-eight-day repeated-dose oral toxcity study in rats
Food consumption: Male

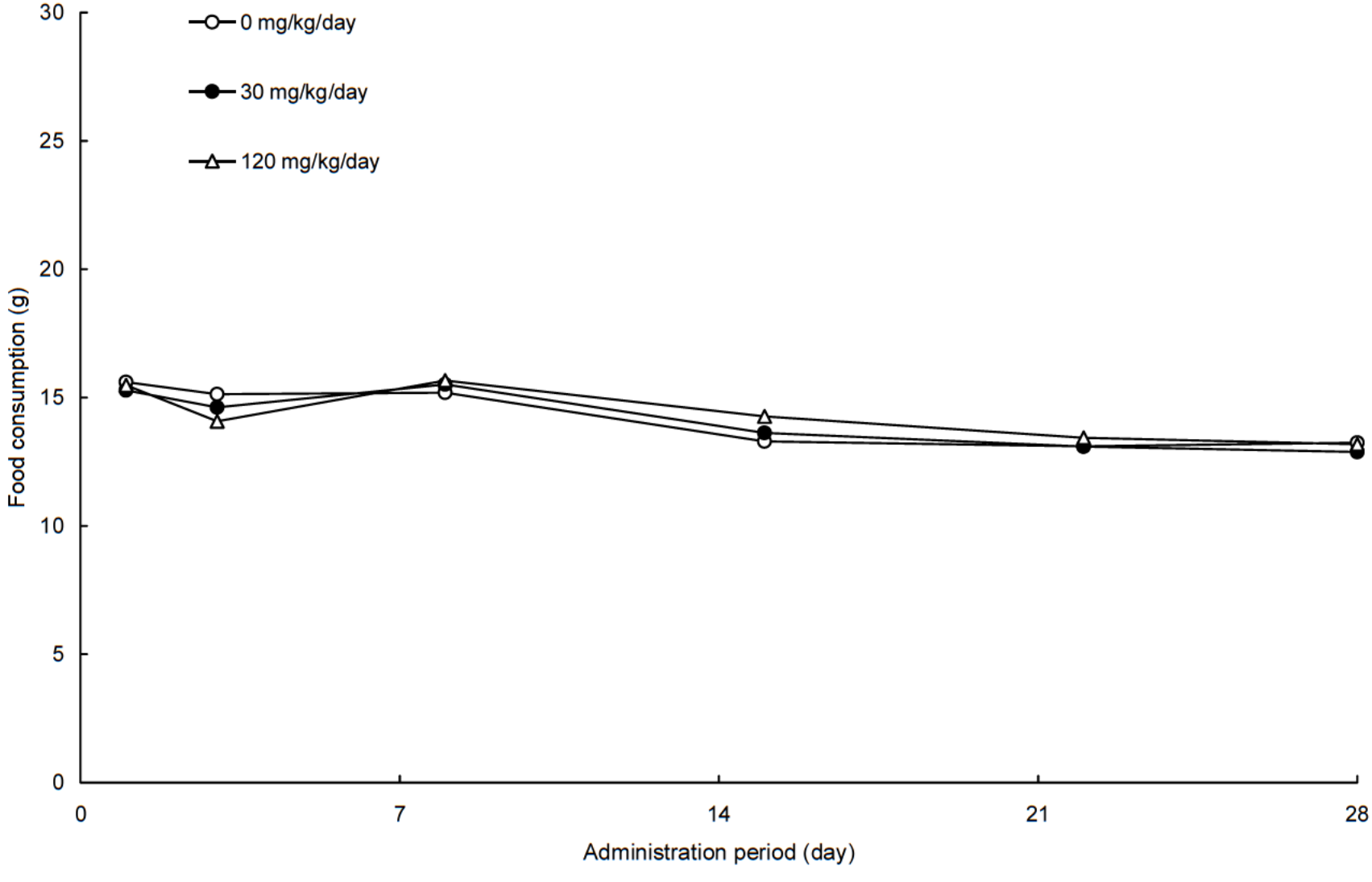


Fig. 3-2 Twenty-eight-day repeated-dose oral toxcity study in rats
Food consumption: Female

Table 1 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of general clinical observations

Sex	Signs	Administration period		
	mg/kg/day	0	30	120
Male		ss 5[a)]	ss 5	ss 5
	No abnormalities detected	5	2	1
	Salivation		3	4
Female		ss 5[a)]	ss 5	ss 5
	No abnormalities detected	5	5	2
	Salivation			3

(R), Recovery.
ss, scheduled sacrifice animal.
a) Number of animals examined.

Table 2-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of body weights (g) : Male

	Dose	Main group	Main group	Main group
	(mg/kg/day)	0	30	120
Administration period (day)	1	136.20 ±6.89 (5)	137.56 ±6.08 (5)	136.90 ±8.06 (5)
	3	153.22 ±10.98 (5)	155.62 ±9.02 (5)	154.02 ±10.40 (5)
	8	193.92 ±18.73 (5)	198.42 ±15.12 (5)	199.04 ±15.15 (5)
	12	225.00 ±25.54 (5)	235.30 ±20.32 (5)	232.02 ±20.03 (5)
	17	262.48 ±30.89 (5)	277.66 ±24.96 (5)	274.34 ±27.40 (5)
	21	292.90 ±34.81 (5)	307.90 ±29.37 (5)	306.06 ±34.21 (5)
	26	325.62 ±37.78 (5)	341.20 ±35.27 (5)	342.24 ±41.24 (5)
	28	338.08 ±40.34 (5)	352.10 ±33.96 (5)	352.52 ±44.48 (5)

Values are shown as Mean ± S.D..

Figure(s) in parentheses indicate number of animals used for the statistical analysis.

* Significantly different from vehicle control at P<0.05.

** Significan ly different from vehicle control at P<0.01.

Table 2-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of body weights (g) : Female

	Dose	Main group	Main group	Main group
	(mg/kg/day)	0	30	120
Administration period (day)	1	125.68 ±4.93 (5)	123.84 ±3.57 (5)	126.34 ±4.53 (5)
	3	136.28 ±6.31 (5)	135.22 ±5.06 (5)	135.60 ±5.28 (5)
	8	157.86 ±5.99 (5)	159.48 ±7.80 (5)	159.24 ±7.18 (5)
	12	170.66 ±8.83 (5)	173.42 ±9.57 (5)	175.38 ±10.51 (5)
	17	188.02 ±10.91 (5)	188.60 ±9.68 (5)	190.00 ±10.83 (5)
	21	201.64 ±12.23 (5)	200.76 ±11.15 (5)	200.58 ±10.76 (5)
	26	215.34 ±13.30 (5)	216.04 ±12.01 (5)	212.36 ±15.01 (5)
	28	219.10 ±12.68 (5)	218.54 ±14.12 (5)	217.78 ±19.44 (5)

Values are shown as Mean ± S.D..

Figure(s) in parentheses indicate number of animals used for the statistical analysis.

* Significantly different from vehicle control at P<0.05.

** Significan ly different from vehicle control at P<0.01.

Table 3 Twenty-eight-day repeated-dose oral toxicity study in rats

Summary of food consumption (g/rat/day)

Sex	Dose (mg/kg/day)	Number of animals	Administration period 1	3	8	15	22	28 (days)
Male	0	5	17.75 ± 1.73	17.82 ± 3.05	19.98 ± 2.46	19.95 ± 3.03	19.52 ± 2.30	19.70 ± 2.12
	30	5	17.34 ± 1.35	18.55 ± 2.11	20.93 ± 2.25	21.83 ± 2.41	21.44 ± 2.13	21.06 ± 2.14
	120	5	17.85 ± 2.67	18.41 ± 2.42	21.27 ± 2.68	21.14 ± 3.10	21.21 ± 3.61	21.12 ± 3.71
Female	0	5	15.60 ± 1.12	15.13 ± 1.07	15.19 ± 1.32	13.29 ± 1.47	13.10 ± 0.89	13.24 ± 1.03
	30	5	15.28 ± 1.92	14.62 ± 1.58	15.51 ± 1.08	13.62 ± 0.64	13.09 ± 1.02	12.88 ± 1.31
	120	5	15.48 ± 0.52	14.08 ± 1.66	15.66 ± 1.38	14.27 ± 0.76	13.43 ± 1.18	13.18 ± 1.71

Mean±S.D.

* Significantly different from vehicle control at P<0.05.

** Significantly different from vehicle control at P<0.01.

Table 4-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of blood chemical examinations:Male

Items	Dose (mg/kg/day)	Main groups 0	30	120
AST	(IU/L)	56.0 ±13.4 (5)	49.8 ±14.1 (5)	51.4 ±7.6 (5)
ALT	(IU/L)	19.0 ±4.6 (5)	16.4 ±6.2 (5)	19.8 ±3.3 (5)
ALP	(IU/L)	579.8 ±128.2 (5)	524.6 ±220.6 (5)	543.8 ±109.4 (5)
γ-GTP	(IU/L)	0.66 ±0.11 (5)	0.66 ±0.38 (5)	0.88 ±0.26 (5)
T-Cho	(mg/dL)	46.0 ±8.9 (5)	32.6 ±11.8 (5)	40.2 ±5.5 (5)
TG	(mg/dL)	77.0 ±24.5 (5)	68.2 ±38.4 (5)	59.0 ±18.6 (5)
BUN	(mg/dL)	7.26 ±1.60 (5)	8.20 ±0.98 (5)	8.26 ±1.99 (5)
Creatinine	(mg/dL)	0.250 ±0.023 (5)	0.226 ±0.018 (5)	0.238 ±0.019 (5)
T-Protein	(g/dL)	4.42 ±0.52 (5)	4.12 ±0.91 (5)	4.08 ±0.41 (5)
Albumin	(g/dL)	2.14 ±0.34 (5)	1.96 ±0.43 (5)	1.94 ±0.18 (5)
A/G ratio	(-)	0.936 ±0.111 (5)	0.914 ±0.096 (5)	0.908 ±0.061 (5)
Glucose	(mg/dL)	141.8 ±8.2 (5)	130.4 ±8.0 (5)	134.8 ±15.4 (5)
T-Bil	(mg/dL)	0.060 ±0.007 (5)	0.064 ±0.009 (5)	0.082 * ±0.019 (5)
TBA	(μmol/L)	26.00 ±16.38 (5)	12.24 ±2.53 (5)	16.62 ±5.63 (5)
IP	(mg/dL)	7.64 ±0.47 (5)	7.60 ±0.33 (5)	7.70 ±0.39 (5)
Ca	(mg/dL)	8.90 ±0.52 (5)	8.20 ±0.83 (5)	8.58 ±0.49 (5)
Na	(mEq/L)	144.2 ±0.8 (5)	144.2 ±0.8 (5)	142.8 ±1.5 (5)
K	(mEq/L)	4.24 ±0.11 (5)	4.24 ±0.21 (5)	4.22 ±0.15 (5)
Cl	(mEq/L)	106.82 ±1.61 (5)	106.92 ±1.05 (5)	106.82 ±1.25 (5)

Values are shown as Mean ± S.D..

Figure(s) in parentheses indicate number of animals used for the statistical analysis.

* Significantly different from vehicle control at P<0.05.

** Significantly different from vehicle control at P<0.01.

Table 4-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of blood chemical examinations:Female

Items	Dose (mg/kg/day)	Main groups 0	30	120
AST	(IU/L)	54.6 ±11.1 (5)	55.4 ±5.7 (5)	49.8 ±4.9 (5)
ALT	(IU/L)	15.6 ±4.9 (5)	15.4 ±2.1 (5)	16.6 ±3.8 (5)
ALP	(IU/L)	296.8 ±84.2 (5)	282.0 ±57.3 (5)	327.8 ±105.1 (5)
γ-GTP	(IU/L)	0.86 ±0.30 (5)	0.88 ±0.33 (5)	1.54 ±0.92 (5)
T-Cho	(mg/dL)	51.2 ±11.2 (5)	50.8 ±8.7 (5)	56.2 ±16.2 (5)
TG	(mg/dL)	28.8 ±25.6 (5)	23.2 ±10.1 (5)	22.2 ±5.1 (5)
BUN	(mg/dL)	11.08 ±1.69 (5)	11.24 ±0.90 (5)	8.96 ±1.61 (5)
Creatinine	(mg/dL)	0.248 ±0.016 (5)	0.266 ±0.009 (5)	0.252 ±0.039 (5)
T-Protein	(g/dL)	4.98 ±0.79 (5)	4.80 ±0.35 (5)	4.32 ±0.53 (5)
Albumin	(g/dL)	2.46 ±0.34 (5)	2.44 ±0.17 (5)	2.16 ±0.29 (5)
A/G ratio	(-)	0.984 ±0.062 (5)	1.036 ±0.038 (5)	0.998 ±0.048 (5)
Glucose	(mg/dL)	116.8 ±20.1 (5)	130.4 ±19.5 (5)	121.6 ±26.9 (5)
T-Bil	(mg/dL)	0.066 ±0.011 (5)	0.068 ±0.020 (5)	0.080 ±0.010 (5)
TBA	(μmol/L)	17.26 ±3.11 (5)	20.10 ±21.24 (5)	11.78 ±4.93 (5)
IP	(mg/dL)	6.44 ±0.32 (5)	6.90 ±0.43 (5)	6.46 ±0.36 (5)
Ca	(mg/dL)	9.24 ±0.48 (5)	8.86 ±0.32 (5)	8.58 ±0.52 (5)
Na	(mEq/L)	143.2 ±0.8 (5)	142.4 ±0.9 (5)	142.4 ±0.5 (5)
K	(mEq/L)	4.32 ±0.20 (5)	4.04 ±0.24 (5)	4.10 ±0.20 (5)
Cl	(mEq/L)	109.08 ±1.46 (5)	108.60 ±1.30 (5)	108.88 ±0.52 (5)

Values are shown as Mean ± S.D..

Figure(s) in parentheses indicate number of animals used for the statistical analysis.

* Significantly different from vehicle control at P<0.05.

** Significantly different from vehicle control at P<0.01.

Table 5-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of absolute organ weights: Male

Items	Dose (mg/kg/day)	0	30	120
Liver	(g)	9.484 ±1.821 (5)	11.150 ±1.195 (5)	12.284 * ±1.937 (5)
Kidneys	(g)	2.302 ±0.320 (5)	2.530 ±0.210 (5)	2.666 ±0.287 (5)
Final body weight	(g)	315.92 ±35.00 (5)	326.88 ±32.92 (5)	328.32 ±40.48 (5)

Values are shown as Mean ± S.D..

Figure(s) in parentheses indicate number of animals used for the statistical analysis.

* Significantly different from vehicle control at P<0.05.

** Significantly different from vehicle control at P<0.01.

Table 5-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of absolute organ weights: Female

Items	Dose (mg/kg/day)	0	30	120
Liver	(g)	6.032 ±0.509 (5)	6.594 ±0.555 (5)	8.166 ** ±1.033 (5)
Kidneys	(g)	1.614 ±0.214 (5)	1.604 ±0.061 (5)	1.780 ±0.121 (5)
Final body weight	(g)	206.60 ±12.52 (5)	206.44 ±12.92 (5)	206.02 ±15.95 (5)

Values are shown as Mean ± S.D..

Figure(s) in parentheses indicate number of animals used for the statistical analysis.

* Significantly different from vehicle control at P<0.05.

** Significantly different from vehicle control at P<0.01.

Table 6-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of relative organ weights: Male

Items	Dose (mg/kg/day)	0	30	120
Liver	(g/100g)	2.982 ±0.248 (5)	3.414 * ±0.214 (5)	3.732 ** ±0.283 (5)
Kidneys	(g/100g)	0.728 ±0.069 (5)	0.778 ±0.077 (5)	0.816 ±0.065 (5)
Final body weight	(g)	315.92 ±35.00 (5)	326.88 ±32.92 (5)	328.32 ±40.48 (5)

Values are shown as Mean ± S.D..

Figure(s) in parentheses indicate number of animals used for the statistical analysis.

* Significantly different from vehicle control at P<0.05.

** Significantly different from vehicle control at P<0.01.

Table 6-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of relative organ weights: Female

Items	Dose (mg/kg/day)	0	30	120
Liver	(g/100g)	2.918 ±0.107 (5)	3.192 ±0.155 (5)	3.954 ** ±0.299 (5)
Kidneys	(g/100g)	0.780 ±0.058 (5)	0.778 ±0.047 (5)	0.864 * ±0.042 (5)
Final body weight	(g)	206.60 ±12.52 (5)	206.44 ±12.92 (5)	206.02 ±15.95 (5)

Values are shown as Mean ± S.D..

Figure(s) in parentheses indicate number of animals used for the statistical analysis.

* Significantly different from vehicle control at P<0.05.

** Significantly different from vehicle control at P<0.01.

Table 7-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of macroscopic examinations

Findings	Male			
	0	30	120	(mg/kg/day)
	ss	ss	ss	
	5[a)]	5	5	
No abnormalities detected	5	5	0	
Oral cavity				
Mottled teeth	0	0	3	
Liver				
Enlargement	0	0	3	

ss: scheduled sacrifice animal.

a) Number of animals examined.

Table 7-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of macroscopic examinations

Findings	Female			
	0	30	120	(mg/kg/day)
	ss	ss	ss	
	5[a)]	5	5	
No abnormalities detected	5	5	0	
Oral cavity				
Mottled teeth	0	0	3	
Liver				
Enlargement	0	0	4	

ss: scheduled sacrifice animal.

a) Number of animals examined.

Table 8-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of histopathological examinations

Findings	Grade	Male			
		0	30	120	(mg/kg/day)
		ss	ss	ss	
		5[a)]	5	5	
Liver					
No abnormalities detected		5/5[b)]	4/5	1/5	
Hypertrophy of hepatocytes, periportal	+	0/5	0/5	3/5	
Microgranuloma	+	0/5	1/5	1/5	
Kidney					
No abnormalities detected		4/5	---	4/5	
Solitary cyst in medulla	+	1/5	---	1/5	

ss: scheduled sacrifice animal.
a) Number of animals autopsied.
b) Number of animals affected / Number of animals examined.
---: Not examined.
+: slight.

Table 8-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Summary of histopathological examinations

Findings	Grade	Female			
		0	30	120	(mg/kg/day)
		ss	ss	ss	
		5[a)]	5	5	
Liver					
No abnormalities detected		5/5[b)]	5/5	1/5	
Hypertrophy of hepatocytes, diffuse	+	0/5	0/5	4/5	
Microgranuloma	+	0/5	0/5	1/5	
Kidney					
No abnormalities detected		5/5	---	5/5	

ss: scheduled sacrifice animal.
a) Number of animals autopsied.
b) Number of animals affected / Number of animals examined.
---: Not examined.
+: slight.

Appendix 1-1 Twenty-eight-day repeated-dose oral toxicity study in rats
General clinical observations of individual animals
0 mg/kg/day

Signs	Sex	Administration period 1	2	3	4	(week)
No abnormalities detected	Male	1[a)], 2, 3, 4, 5	1, 2, 3, 4, 5	1, 2, 3, 4, 5	1, 2, 3, 4, 5	
	Female	16, 17, 18, 19, 20	16, 17, 18, 19, 20	16, 17, 18, 19, 20	16, 17, 18, 19, 20	

a) Animal number.

Appendix 1-2 Twenty-eight-day repeated-dose oral toxicity study in rats
General clinical observations of individual animals
30 mg/kg/day

Signs	Sex	Administration period 1	2	3	4	(week)
No abnormalities detected	Male	6[a)], 7, 8, 9, 10	6, 7, 8, 9, 10	6, 7	6, 7, 8, 10	
	Female	21, 22, 23, 24, 25	21, 22, 23, 24, 25	21, 22, 23, 24, 25	21, 22, 23, 24, 25	
Salivation (disappeared within 15 or 30 minutes after dosing)	Male			8, 9, 10	9	
	Female					

a) Animal number.

Appendix 1-3 Twenty-eight-day repeated-dose oral toxicity study in rats
General clinical observations of individual animals
120 mg/kg/day

Signs	Sex	Administration period 1 (week)	2 (week)	3 (week)	4 (week)
No abnormalities detected	Male	11[a)], 12, 13, 14, 15	12	11, 12	11, 12
	Female	26, 27, 28, 29, 30	26, 27, 30	27, 28, 29, 30	27, 28, 30
Salivation (disappeared within 15 or 30 minutes after dosing)	Male		11, 13, 14, 15	13, 14, 15	13, 14, 15
	Female		28, 29	26	26, 29
Salivation (just before dosing)	Male		13		
	Female				

a) Animal number.

Appendix 2-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Body weights of individual animals (g) : Male

Group		Main group				
Dose (mg/kg/day)		0				
	Animal No.	1	2	3	4	5
Administration period (day)	1	125.3	133.4	141.3	140.2	140.8
	3	135.6	151.4	162.1	154.4	162.6
	8	167.9	185.4	209.7	192.6	214.0
	12	189.8	210.3	244.3	227.8	252.8
	17	222.0	241.4	290.9	265.3	292.8
	21	247.2	269.3	320.4	296.9	330.7
	26	276.2	305.7	353.0	321.7	371.5
	28	286.6	315.7	364.3	334.2	389.6

Appendix 2-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Body weights of individual animals (g) : Male

Group		Main group				
Dose (mg/kg/day)				30		
	Animal No.	6	7	8	9	10
Administration period (day)	1	132.3	133.3	134.4	141.5	146.3
	3	147.3	147.6	153.0	163.5	166.7
	8	185.4	182.6	196.0	214.2	213.9
	12	217.0	215.8	231.5	261.4	250.8
	17	254.7	257.6	267.1	307.3	301.6
	21	279.9	288.0	292.2	340.7	338.7
	26	310.9	316.5	319.3	381.9	377.4
	28	320.7	330.9	331.9	396.2	380.8

Appendix 2-3 Twenty-eight-day repeated-dose oral toxicity study in rats
Body weights of individual animals (g) : Male

Group		Main group				
Dose (mg/kg/day)		120				
	Animal No.	11	12	13	14	15
Administration period (day)	1	126.8	134.8	135.4	138.4	149.1
	3	139.7	152.8	151.2	158.2	168.2
	8	175.7	194.8	200.2	213.9	210.6
	12	200.8	228.9	231.7	252.5	246.2
	17	233.5	275.1	266.5	306.2	290.4
	21	255.0	310.4	294.2	345.3	325.4
	26	280.6	348.5	328.4	390.6	363.1
	28	286.1	363.8	335.1	403.6	374.0

Appendix 2-4 Twenty-eight-day repeated-dose oral toxicity study in rats
Body weights of individual animals (g) : Female

Group		Main group				
Dose (mg/kg/day)		0				
	Animal No.	16	17	18	19	20
Administration period (day)	1	119.1	124.7	123.5	131.2	129.9
	3	129.6	135.2	131.2	141.1	144.3
	8	154.1	153.4	153.3	166.1	162.4
	12	170.8	165.2	161.2	184.5	171.6
	17	189.5	179.2	175.2	201.9	194.3
	21	204.8	191.3	187.8	218.0	206.3
	26	216.8	203.8	202.4	235.2	218.5
	28	221.6	210.2	207.0	239.3	217.4

Appendix 2-5 Twenty-eight-day repeated-dose oral toxicity study in rats
Body weights of individual animals (g) : Female

Group		Main group				
Dose (mg/kg/day)				30		
	Animal No.	21	22	23	24	25
Administration period (day)	1	119.3	122.1	124.2	124.6	129.0
	3	127.6	134.5	141.6	135.4	137.0
	8	146.9	160.4	166.8	158.4	164.9
	12	157.6	176.4	180.5	171.8	180.8
	17	173.3	192.4	189.3	188.2	199.8
	21	182.4	206.2	199.6	203.9	211.7
	26	196.7	228.3	216.2	215.7	223.3
	28	197.3	234.5	216.7	216.5	227.7

Appendix 2-6 Twenty-eight-day repeated-dose oral toxicity study in rats
Body weights of individual animals (g) : Female

Group		Main group				
Dose (mg/kg/day)		120				
	Animal No.	26	27	28	29	30
Administration period (day)	1	123.4	122.3	123.5	131.9	130.6
	3	127.9	137.4	133.5	137.1	142.1
	8	154.9	161.2	151.0	159.1	170.0
	12	172.5	174.4	165.4	171.4	193.2
	17	192.4	192.8	176.9	182.7	205.2
	21	207.5	202.6	186.7	192.9	213.2
	26	215.6	223.8	195.2	198.4	228.8
	28	220.0	237.8	192.0	204.8	234.3

Appendix 3-1 Twenty-eight-day repeated-dose oral toxicity study in rats

Food consumption of individual animals (g/rat/day)

Sex	Dose (mg/kg/day)	Animal No.	Administration period						
			1	3	8	15	22	28	(days)
Male	0	1	16.32	13.06	16.52	16.17	17.45	17.73	
		2	16.68	18.37	18.70	17.16	16.71	17.65	
		3	20.69	19.94	22.05	22.33	21.88	20.31	
		4	17.61	16.93	20.15	21.71	20.44	20.08	
		5	17.46	20.80	22.50	22.38	21.10	22.75	
	30	6	15.89	16.68	19.23	19.62	20.66	19.98	
		7	17.42	16.97	18.15	20.07	19.08	18.93	
		8	16.54	17.45	21.11	21.11	20.09	19.66	
		9	17.37	20.45	23.47	25.56	23.38	23.64	
		10	19.46	21.20	22.69	22.77	23.98	23.08	
	120	11	15.56	15.60	17.45	17.06	16.59	16.15	
		12	16.51	18.45	21.66	21.40	22.34	22.82	
		13	15.98	16.46	20.11	19.73	18.93	18.44	
		14	19.36	20.22	24.60	25.50	26.05	25.25	
		15	21.82	21.31	22.53	22.03	22.12	22.94	

Appendix 3-2 Twenty-eight-day repeated-dose oral toxicity study in rats

Food consumption of individual animals (g/rat/day)

Sex	Dose (mg/kg/day)	Animal No.	Administration period						
			1	3	8	15	22	28	(days)
Female	0	16	14.07	14.15	14.94	12.65	12.47	12.00	
		17	15.93	14.74	13.31	12.05	12.01	12.77	
		18	14.96	14.21	14.82	12.19	13.06	13.17	
		19	16.98	16.32	16.70	15.50	13.77	14.80	
		20	16.08	16.22	16.16	14.06	14.17	13.45	
	30	21	13.44	12.97	14.25	12.55	11.69	10.87	
		22	14.81	14.69	15.82	14.10	14.07	14.51	
		23	15.37	16.87	17.14	14.13	12.53	13.09	
		24	18.48	15.26	14.98	13.57	14.06	13.21	
		25	14.30	13.30	15.37	13.73	13.08	12.73	
	120	26	14.90	11.93	14.56	14.21	14.41	11.99	
		27	15.37	15.59	15.20	13.64	14.45	15.85	
		28	15.11	13.35	14.49	13.66	11.72	11.52	
		29	15.94	13.62	16.27	14.36	12.73	12.84	
		30	16.10	15.92	17.76	15.50	13.84	13.72	

Appendix 4-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Blood chemical data of individual animals:Male

Group		Main group				
Dose(mg/kg/day)		0				
Items	Animal No.	1 ss	2 ss	3 ss	4 ss	5 ss
AST	(IU/L)	70	71	48	48	43
ALT	(IU/L)	23	24	13	19	16
ALP	(IU/L)	787	573	452	587	500
γ-GTP	(IU/L)	0.7	0.6	0.5	0.7	0 8
T-Cho	(mg/dL)	59	44	46	34	47
TG	(mg/dL)	85	83	61	46	110
BUN	(mg/dL)	8.7	8.9	7.4	5.3	6 0
Creatinine	(mg/dL)	0 25	0.26	0.26	0.21	0.27
T-Protein	(g/dL)	4.9	5.0	4.3	3.8	4.1
Albumin	(g/dL)	2.6	2.4	2.0	1.8	1 9
A/G ratio	(-)	1.13	0.92	0.87	0.90	0.86
Glucose	(mg/dL)	149	151	141	136	132
T-Bil	(mg/dL)	0 06	0.06	0.06	0.07	0.05
TBA	(μmol/L)	55.1	21.1	15.8	19.5	18 5
IP	(mg/dL)	7.9	8.2	7.8	7.1	7 2
Ca	(mg/dL)	9.5	9.4	8.6	8.3	8.7
Na	(mEq/L)	144	143	145	144	145
K	(mEq/L)	4.3	4.2	4.2	4.4	4.1
Cl	(mEq/L)	108.2	104.5	107.6	105.8	108 0

ss: scheduled sacrifice animal.

Appendix 4-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Blood chemical data of individual animals:Male

Group		Main group				
Dose(mg/kg/day)		30				
Items	Animal No.	6 ss	7 ss	8 ss	9 ss	10 ss
AST	(IU/L)	46	46	37	46	74
ALT	(IU/L)	15	13	11	16	27
ALP	(IU/L)	348	400	412	574	889
γ-GTP	(IU/L)	0.3	0.5	0.5	0.7	1 3
T-Cho	(mg/dL)	19	34	25	35	50
TG	(mg/dL)	21	88	42	71	119
BUN	(mg/dL)	9.1	7.7	9.4	7.2	7.6
Creatinine	(mg/dL)	0 20	0.23	0.22	0.23	0.25
T-Protein	(g/dL)	3.5	3.9	3.5	4.0	5.7
Albumin	(g/dL)	1.6	1.9	1.8	1.8	2.7
A/G ratio	(-)	0 84	0.95	1.06	0.82	0.90
Glucose	(mg/dL)	129	121	142	126	134
T-Bil	(mg/dL)	0 06	0.06	0.06	0.06	0.08
TBA	(μmol/L)	8.3	13.8	13.8	11.1	14 2
IP	(mg/dL)	7.1	7.9	7.6	7.5	7 9
Ca	(mg/dL)	7.7	8.2	8.0	7.5	9.6
Na	(mEq/L)	143	144	144	145	145
K	(mEq/L)	4.1	4.2	4.0	4.4	4 5
Cl	(mEq/L)	106.4	108.3	105.6	107.6	106.7

ss: scheduled sacrifice animal.

Appendix 4-3 Twenty-eight-day repeated-dose oral toxicity study in rats
Blood chemical data of individual animals:Male

Group		Main group				
Dose(mg/kg/day)		120				
Items	Animal No.	11 ss	12 ss	13 ss	14 ss	15 ss
AST	(IU/L)	53	59	44	43	58
ALT	(IU/L)	19	24	15	21	20
ALP	(IU/L)	681	470	453	471	644
γ-GTP	(IU/L)	0.9	0.6	1.3	0.8	0 8
T-Cho	(mg/dL)	42	36	46	33	44
TG	(mg/dL)	75	28	57	70	65
BUN	(mg/dL)	10.3	10.0	6.2	8.6	6 2
Creatinine	(mg/dL)	0 27	0.23	0.22	0.23	0.24
T-Protein	(g/dL)	3.8	4.4	3.6	4.0	4.6
Albumin	(g/dL)	1.9	2.0	1.7	1.9	2 2
A/G ratio	(-)	1 00	0.83	0.89	0.90	0.92
Glucose	(mg/dL)	120	144	141	152	117
T-Bil	(mg/dL)	0.11	0.09	0.08	0.06	0.07
TBA	(μmol/L)	19.0	24.1	14.3	8.9	16 8
IP	(mg/dL)	7.7	7.7	7.1	7.8	8 2
Ca	(mg/dL)	8.2	8.6	8.2	8.5	9.4
Na	(mEq/L)	145	142	143	141	143
K	(mEq/L)	4.0	4.2	4.2	4.4	4 3
Cl	(mEq/L)	108.3	106.5	107.3	104.9	107.1

ss: scheduled sacrifice animal.

Appendix 4-4 Twenty-eight-day repeated-dose oral toxicity study in rats
Blood chemical data of individual animals:Female

Group		Main group				
Dose(mg/kg/day)		0				
Items	Animal No.	16 ss	17 ss	18 ss	19 ss	20 ss
AST	(IU/L)	41	67	45	60	60
ALT	(IU/L)	8	18	14	17	21
ALP	(IU/L)	254	419	197	280	334
γ-GTP	(IU/L)	0.6	0.9	0.5	1.2	1.1
T-Cho	(mg/dL)	39	47	58	45	67
TG	(mg/dL)	11	73	21	12	27
BUN	(mg/dL)	10.2	11.3	13.3	8.8	11 8
Creatinine	(mg/dL)	0 24	0.26	0.24	0.23	0.27
T-Protein	(g/dL)	3.7	5.1	4.9	5.4	5 8
Albumin	(g/dL)	1.9	2.5	2.5	2.6	2 8
A/G ratio	(-)	1 06	0.96	1.04	0.93	0.93
Glucose	(mg/dL)	98	144	117	128	97
T-Bil	(mg/dL)	0 06	0.08	0.07	0.05	0.07
TBA	(μmol/L)	14.9	20.9	13.3	19.2	18 0
IP	(mg/dL)	6.3	6.2	6.3	6.4	7 0
Ca	(mg/dL)	8.4	9.3	9.5	9.5	9 5
Na	(mEq/L)	143	143	144	144	142
K	(mEq/L)	4.3	4.0	4.5	4.3	4 5
Cl	(mEq/L)	109.1	108.0	110.8	110.2	107 3

ss: scheduled sacrifice animal.

Appendix 4-5 Twenty-eight-day repeated-dose oral toxicity study in rats
Blood chemical data of individual animals:Female

Group		Main group				
Dose(mg/kg/day)		30				
Items	Animal No.	21 ss	22 ss	23 ss	24 ss	25 ss
AST	(IU/L)	62	54	47	55	59
ALT	(IU/L)	14	15	14	15	19
ALP	(IU/L)	324	341	288	261	196
γ-GTP	(IU/L)	1.0	0.7	0.6	1.4	0.7
T-Cho	(mg/dL)	46	50	45	47	66
TG	(mg/dL)	35	32	12	15	22
BUN	(mg/dL)	12.8	10.7	11.2	10.9	10.6
Creatinine	(mg/dL)	0 26	0.28	0.26	0.27	0.26
T-Protein	(g/dL)	4.8	4.6	4.7	4.5	5.4
Albumin	(g/dL)	2.5	2.3	2.4	2.3	2.7
A/G ratio	(-)	1 09	1.00	1.04	1.05	1.00
Glucose	(mg/dL)	105	139	151	115	142
T-Bil	(mg/dL)	0.10	0.05	0.05	0.07	0.07
TBA	(μmol/L)	57.7	8.6	7.7	15.6	10 9
IP	(mg/dL)	7.1	7.1	6.2	6.8	7 3
Ca	(mg/dL)	8.9	8.9	8.4	8.8	9 3
Na	(mEq/L)	143	141	143	142	143
K	(mEq/L)	4.2	3.9	3.7	4.1	4 3
Cl	(mEq/L)	108.6	106.6	110.2	108.6	109 0

ss: scheduled sacrifice animal.

Appendix 4-6 Twenty-eight-day repeated-dose oral toxicity study in rats
Blood chemical data of individual animals:Female

Group		Main group				
Dose(mg/kg/day)		120				
Items	Animal No.	26 ss	27 ss	28 ss	29 ss	30 ss
AST	(IU/L)	54	44	53	45	53
ALT	(IU/L)	20	13	20	12	18
ALP	(IU/L)	341	400	206	238	454
γ-GTP	(IU/L)	1.4	0.7	1.4	1.1	3.1
T-Cho	(mg/dL)	69	43	71	35	63
TG	(mg/dL)	21	16	21	23	30
BUN	(mg/dL)	10.9	6.9	8.7	10.2	8.1
Creatinine	(mg/dL)	0 30	0.20	0.24	0.24	0.28
T-Protein	(g/dL)	4.9	3.7	4.8	3.9	4 3
Albumin	(g/dL)	2.5	1.8	2.4	2.0	2.1
A/G ratio	(-)	1 04	0.95	1.00	1.05	0.95
Glucose	(mg/dL)	88	121	155	104	140
T-Bil	(mg/dL)	0 07	0.07	0.08	0.09	0.09
TBA	(μmol/L)	11.2	20.0	8.9	7.2	11.6
IP	(mg/dL)	6.2	6.1	6.8	6.9	6 3
Ca	(mg/dL)	8.7	7.9	9.3	8.3	8.7
Na	(mEq/L)	142	142	142	143	143
K	(mEq/L)	4.2	4.0	4.2	4.3	3 8
Cl	(mEq/L)	109.2	108.7	108.3	109.6	108.6

ss: scheduled sacrifice animal.

Appendix 5-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Absolute organ weights of individual animals: Male

Dose	(mg/kg/day)	0				
Items	Animal No.	1 ss	2 ss	3 ss	4 ss	5 ss
Liver	(g)	7.35	8.20	10.50	9.43	11.94
Kidneys	(g)	1.91	2.07	2.30	2.67	2.56
Final body weight	(g)	269.2	298.6	338.6	313.7	359.5

ss: scheduled sacrifice animal.

Appendix 5-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Absolute organ weights of individual animals: Male

Dose	(mg/kg/day)	30				
Items	Animal No.	6 ss	7 ss	8 ss	9 ss	10 ss
Liver	(g)	10.71	9.54	10.83	12.24	12.43
Kidneys	(g)	2.68	2.28	2.33	2.62	2.74
Final body weight	(g)	294.4	308.5	307.4	368.7	355.4

ss: scheduled sacrifice animal.

Appendix 5-3 Twenty-eight-day repeated-dose oral toxicity study in rats
Absolute organ weights of individual animals: Male

Dose	(mg/kg/day)	120				
Items	Animal No.	11 ss	12 ss	13 ss	14 ss	15 ss
Liver	(g)	9.17	12.14	13.11	14.41	12.59
Kidneys	(g)	2.29	2.60	2.60	2.76	3.08
Final body weight	(g)	267.4	337.8	314.7	375.9	345.8

ss: scheduled sacrifice animal.

Appendix 5-4 Twenty-eight-day repeated-dose oral toxicity study in rats
Absolute organ weights of individual animals: Female

Dose	(mg/kg/day)	0				
Items	Animal No.	16 ss	17 ss	18 ss	19 ss	20 ss
Liver	(g)	6.02	5.47	5.60	6.65	6.42
Kidneys	(g)	1.57	1.44	1.46	1.97	1.63
Final body weight	(g)	210.5	195.6	193.7	224.5	208.7

ss: scheduled sacrifice animal.

Appendix 5-5 Twenty-eight-day repeated-dose oral toxicity study in rats
Absolute organ weights of individual animals: Female

Dose	(mg/kg/day)	30				
Items	Animal No.	21 ss	22 ss	23 ss	24 ss	25 ss
Liver	(g)	5.68	6.83	7.12	6.51	6.83
Kidneys	(g)	1.53	1.55	1.64	1.67	1.63
Final body weight	(g)	185.7	218.0	205.9	206.0	216.6

ss: scheduled sacrifice animal.

Appendix 5-6 Twenty-eight-day repeated-dose oral toxicity study in rats
Absolute organ weights of individual animals: Female

Dose	(mg/kg/day)	120				
Items	Animal No.	26 ss	27 ss	28 ss	29 ss	30 ss
Liver	(g)	7.95	8.77	7.68	6.87	9.56
Kidneys	(g)	1.70	1.98	1.69	1.72	1.81
Final body weight	(g)	206.9	221.8	185.9	194.4	221.1

ss: scheduled sacrifice animal.

Appendix 6-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Relative organ weights of individual animals: Male

Dose	(mg/kg/day)	0				
Items	Animal No.	1 ss	2 ss	3 ss	4 ss	5 ss
Liver	(g/100g)	2.73	2.75	3.10	3.01	3.32
Kidneys	(g/100g)	0.71	0.69	0.68	0.85	0.71
Final body weight	(g)	269.2	298.6	338.6	313.7	359.5

ss: scheduled sacrifice animal.

Appendix 6-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Relative organ weights of individual animals: Male

Dose	(mg/kg/day)			30		
Items	Animal No.	6 ss	7 ss	8 ss	9 ss	10 ss
Liver	(g/100g)	3.64	3.09	3.52	3.32	3.50
Kidneys	(g/100g)	0.91	0.74	0.76	0.71	0.77
Final body weight	(g)	294.4	308.5	307.4	368.7	355.4

ss: scheduled sacrifice animal.

Appendix 6-3 Twenty-eight-day repeated-dose oral toxicity study in rats
Relative organ weights of individual animals: Male

Dose	(mg/kg/day)			120		
Items	Animal No.	11 ss	12 ss	13 ss	14 ss	15 ss
Liver	(g/100g)	3.43	3.59	4.17	3.83	3.64
Kidneys	(g/100g)	0.86	0.77	0.83	0.73	0.89
Final body weight	(g)	267.4	337.8	314.7	375.9	345.8

ss: scheduled sacrifice animal.

Appendix 6-4 Twenty-eight-day repeated-dose oral toxicity study in rats
Relative organ weights of individual animals: Female

Dose	(mg/kg/day)	0				
Items	Animal No.	16 ss	17 ss	18 ss	19 ss	20 ss
Liver	(g/100g)	2.86	2.80	2.89	2.96	3.08
Kidneys	(g/100g)	0.75	0.74	0.75	0.88	0.78
Final body weight	(g)	210.5	195.6	193.7	224.5	208.7

ss: scheduled sacrifice animal.

Appendix 6-5 Twenty-eight-day repeated-dose oral toxicity study in rats
Relative organ weights of individual animals: Female

Dose	(mg/kg/day)			30		
Items	Animal No.	21 ss	22 ss	23 ss	24 ss	25 ss
Liver	(g/100g)	3.06	3.13	3.46	3.16	3.15
Kidneys	(g/100g)	0.82	0.71	0.80	0.81	0.75
Final body weight	(g)	185.7	218.0	205.9	206.0	216.6

ss: scheduled sacrifice animal.

Appendix 6-6 Twenty-eight-day repeated-dose oral toxicity study in rats
Relative organ weights of individual animals: Female

Dose	(mg/kg/day)	120				
Items	Animal No.	26 ss	27 ss	28 ss	29 ss	30 ss
Liver	(g/100g)	3.84	3.95	4.13	3.53	4.32
Kidneys	(g/100g)	0.82	0.89	0.91	0.88	0.82
Final body weight	(g)	206.9	221.8	185.9	194.4	221.1

ss: scheduled sacrifice animal.

Appendix 7-1 Twenty-eight-day repeated-dose oral toxicity study in rats
Pathological findings of individual animals

Sex	Dose (mg/kg/day)	Animal No.	Fate	Macroscopic findings	Histopathological findings[a)]
Male	0	1	ss	No abnormalities detected	No abnormalities detected
		2	ss	No abnormalities detected	No abnormalities detected
		3	ss	No abnormalities detected	Kidney Solitary cyst in medulla +
		4	ss	No abnormalities detected	No abnormalities detected
		5	ss	No abnormalities detected	No abnormalities detected

a) Organs/tissues examined as follows: liver and kidneys.
ss: scheduled sacrifice animal.
+: slight.

Appendix 7-2 Twenty-eight-day repeated-dose oral toxicity study in rats
Pathological findings of individual animals

Sex	Dose (mg/kg/day)	Animal No.	Fate	Macroscopic findings	Histopathological findings[a)]
Male	30	6	ss	No abnormalities detected	Liver Microgranuloma +
		7	ss	No abnormalities detected	No abnormalities detected
		8	ss	No abnormalities detected	No abnormalities detected
		9	ss	No abnormalities detected	No abnormalities detected
		10	ss	No abnormalities detected	No abnormalities detected

a) Organs/tissues examined as follows: liver.
ss: scheduled sacrifice animal.
+: slight.

Appendix 7-3 Twenty-eight-day repeated-dose oral toxicity study in rats
Pathological findings of individual animals

Sex	Dose (mg/kg/day)	Animal No.	Fate	Macroscopic findings	Histopathological findings[a)]
Male	120	11	ss	Oral cavity Mottled teeth (upper and lower incisors)	Incisor Not examined Liver Hypertrophy of hepatocytes, periportal + Kidney Solitary cyst in medulla +
		12	ss	Oral cavity Mottled teeth (upper incisors) Liver Enlargement	Incisor Not examined Liver Hypertrophy of hepatocytes, periportal +
		13	ss	Liver Enlargement	Liver Hypertrophy of hepatocytes, periportal +
		14	ss	Liver Enlargement	Liver Microgranuloma +
		15	ss	Oral cavity Mottled teeth (upper and lower incisors)	No abnormalities detected Incisor Not examined

a) Organs/tissues examined as follows: liver and kidneys.
ss: scheduled sacrifice animal.
+: slight.

Appendix 7-4 Twenty-eight-day repeated-dose oral toxicity study in rats
Pathological findings of individual animals

Sex	Dose (mg/kg/day)	Animal No.	Fate	Macroscopic findings	Histopathological findings[a)]
Female	0	16	ss	No abnormalities detected	No abnormalities detected
		17	ss	No abnormalities detected	No abnormalities detected
		18	ss	No abnormalities detected	No abnormalities detected
		19	ss	No abnormalities detected	No abnormalities detected
		20	ss	No abnormalities detected	No abnormalities detected

a) Organs/tissues examined as follows: liver and kidneys.
ss: scheduled sacrifice animal.

Appendix 7-5 Twenty-eight-day repeated-dose oral toxicity study in rats
Pathological findings of individual animals

Sex	Dose (mg/kg/day)	Animal No.	Fate	Macroscopic findings	Histopathological findings[a)]
Female	30	21	ss	No abnormalities detected	No abnormalities detected
		22	ss	No abnormalities detected	No abnormalities detected
		23	ss	No abnormalities detected	No abnormalities detected
		24	ss	No abnormalities detected	No abnormalities detected
		25	ss	No abnormalities detected	No abnormalities detected

a) Organs/tissues examined as follows: liver.
ss: scheduled sacrifice animal.

Appendix 7-6 Twenty-eight-day repeated-dose oral toxicity study in rats
Pathological findings of individual animals

Sex	Dose (mg/kg/day)	Animal No.	Fate	Macroscopic findings	Histopathological findings[a)]
Female	120	26	ss	Oral cavity Mottled teeth (upper incisors) Liver Enlargement	Incisor Not examined Liver Hypertrophy of hepatocytes, diffuse + Microgranuloma +
		27	ss	Oral cavity Mottled teeth (upper and lower incisors) Liver Enlargement	Incisor Not examined Liver Hypertrophy of hepatocytes, diffuse +
		28	ss	Liver Enlargement	Liver Hypertrophy of hepatocytes, diffuse +
		29	ss	Oral cavity Mottled teeth (upper incisors)	No abnormalities detected Incisor Not examined
		30	ss	Liver Enlargement	Liver Hypertrophy of hepatocytes, diffuse +

a) Organs/tissues examined as follows: liver and kidneys.
ss: scheduled sacrifice animal.
+: slight.

QUALITY ASSURANCE STATEMENT

Chemicals Evaluation and Research Institute, Japan, Hita

Sponsor: DAIKIN INDUSTRIES, LTD.

Title: Twenty-Eight-Day Repeated-Dose Oral Toxicity Study of 13F-SFA in Rats

Study Number: B11-1054

I assure that the final report accurately describes the test methods and procedures, and that the reported results accurately reflect the raw data of this study. The inspections of the study were carried out and the results were reported to the Study Director and the Test Facility Management by Quality Assurance Unit as follows.

Item of inspection	Date of inspection	Date of report
Study plan	November 29, 2013	November 29, 2013
Assessment of response (study plan)	December 2, 2013	December 2, 2013
Stability of test substance	December 4, 2013	December 4, 2013
Preparation of test substance formulations	December 9, 2013	December 10, 2013
Concentration analysis of test substance formulations	December 9, 2013	December 10, 2013
Body weight measurement	December 10, 2013	December 10, 2013
Administration and clinical observations	December 10, 2013	December 10, 2013
Study plan amendment (No.1)	January 6, 2014	January 6, 2014
Blood sampling	January 7, 2014	January 7, 2014
Dissection, necropsy and organ weight measurements	January 7, 2014	January 7, 2014
Blood chemistry	January 7, 2014	January 7, 2014
Clinical chemistry data	March 4, 2014	March 4, 2014
Re-inspection of clinical chemistry data	March 5, 2014	March 5, 2014
Animal data	March 28, 2014	March 28, 2014
Pathological data	March 31, 2014	April 1, 2014
Test substance and housing condition data	April 11, 2014	April 11, 2014
Raw data and draft final report	April 11, 2014	April 11, 2014
Re-inspection of raw data and draft final report	April 14, 2014	April 14, 2014
Raw data and draft final report (No.2)	April 14, 2014	April 14, 2014
Final report	April 16, 2014	April 16, 2014

The inspection result of following item was reported to the Study Director and the Test Facility Management based on the report of facility-based inspection and/or process-based inspection relevant to this study type and timeframe.

Item of inspection	Date of inspection	Date of report
Animal receipt	November 19, 2013	April 16, 2014
Quarantine and acclimatization	November 19, 2013	April 16, 2014
Animal management	November 19, 2013	April 16, 2014
Allocation and animal identification	October 3, 2013	April 16, 2014
Food intake measurement	November 6, 2013	April 16, 2014
Pathological preparation	November 5, 6, 7, 8 and 15, 2013	April 16, 2014
Pathological preparation	January 20, 21, 27 and 30, 2014	April 16, 2014
Microscopic examinations	February 25, 2014	April 16, 2014

Date: April 16, 2014

Quality Assurance Manager :