

**UNIVERSITY OF VETERINARY SCIENCE
DEPARTMENT OF PHARMACOLOGY & TOXICOLOGY
H-1078 Budapest, István u. 2.**

FINAL REPORT

**ACUTE ORAL TOXICITY STUDY
of AVEMAR
IN MOUSE**

CODE: 9901

**BUDAPEST
1999**

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RESPONSIBLE PERSONS

STUDY DIRECTOR

01.06.1999

Date

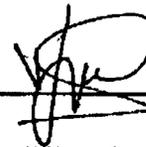


József LEHEL
DVM, Ph.D.
senior researcher

**HEAD OF THE
DEPARTMENT**

01.06.1999

Date

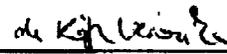


Prof. Gábor SEMJÉN
DVM, Ph.D.
full professor

**QUALITY
ASSURANCE UNIT**

03.06.1999

Date



Veronika RÁTZ
DVM

QUALITY ASSURANCE STATEMENT

I declare that the final report of "Acute oral toxicity study of AVEMAR in mouse" (code: 9901) is based on correct experimental data and the written results and data in the final report are in accordance with experimental results.

Date of inspections	Report to	
	the study director	the head of the department
26th March, 1999	26th March, 1999	28th March, 1999
19th April, 1999	20th April, 1999	-
21st April, 1999	22nd April, 1999	-
21st April, 1999	22nd April, 1999	-
6th May, 1999	7th May, 1999	7th May, 1999
21st May, 1999	26th May, 1999	-
1st June, 1999	2nd June, 1999	-

Budapest, 3rd June, 1999

de Kijf Veronika
Rátz Veronika
DVM
Quality Assurance

SUMMARY

Title of the study: ACUTE ORAL TOXICITY STUDY OF AVEMAR IN MOUSE

Name of substance: AVEMAR

Animal species: CD-1 mouse

Sex: equally of both sexes

Number of animal: 10/dose and sex

Mode of treatment: orally

Duration of treatment: once application

Dose: 2000 mg/kg b.w. (40 mg/20 g b.w.)

Volume: 0.5 ml/20 g b.w.

Excipient: distilled water

Post-treatment period: 14 days

Examined parameters: Clinical symptoms
Lethality
Body weight
Necropsy

Results: There were no lethality and abnormal clinical signs neither in control group nor in the treated group (AVEMAR, dose: 2000 mg/kg b.w.) of both sexes.

The LD₅₀ value of AVEMAR in male and female mice is

LD₅₀ > 2000 mg/kg

1. GENERAL INFORMATION

1.1. TITLE OF THE STUDY

Acute oral toxicity study of AVEMAR in mouse.

1.2. OBJECT OF THE STUDY

The aim of this study was to determine the acute oral LD₅₀ value of the test substance in mouse and to get information about toxic characteristics of the test substance and to the subacute toxicity study.

1.3. TYPE OF THE STUDY

Preclinical safety study was performed according to the GLP as described by regulation No P-44-1992 of the National Institute of Pharmacy (OGYI) and complying with the Good Laboratory Practice for Testing of Chemicals ENV/MC/CHEM (98)17.

The study was carried out with limit-test according to the OECD Guidelines for Testing of Chemicals (No. 401, adopted: 24th February, 1987).

1.4. INSTITUTION PERFORMING THE STUDY

University of Veterinary Science, Department of Pharmacology & Toxicology
H-1078 Budapest, István u. 2.

1.5. SPONSOR

BIROMEDICINA Research, Development and Commercial Co.
H-1088 Budapest, Puskin utca 4.

2. TEST AND CONTROL SUBSTANCE

2.1. TEST SUBSTANCE

Name of the substance:	AVEMAR
Manufacturer:	Biomedicina Research, Development and Commercial Co.
Batch number:	00799115
Analytical examination:	identified with 15th February, 1999
Physical characteristic	brown granules
Storage condition:	storage at refrigerator (0-20 °C)
Safety regulation:	there is no special regulation
Main pharmacological effect:	immunostimulant
Expiry date:	February, 2001

2.1.1. Chemical analysis

Analytical examination of the test substance was performed prior to the study by the Sponsor.

2.2. CONTROL SUBSTANCE

Name of the substance:	distilled water
Manufacturer:	Pharmafontana Gyógyszerészeti Rt.
Batch number:	19990414
Number of the quality certificate:	133/1999.04.16.
Storage condition:	at room temperature
Safety regulation:	no special regulation is required
Expiry date:	21st April, 1999

2.3. DOSAGE FORM

The test substance was dissolved in distilled water.

2.3.1. Preparation of the test substance

For the treatment the test substance was prepared immediately before the application (it was no longer than 4 hours) in a calibrated flask. 2 g from the test substance was measured and then distilled water was added to end-volume (25 ml).

2.3.2. Stability control of the test substance

Stability control was not performed, complying with the prescription of the preparation of the test substance.

3. TEST SYSTEM

3.1. ANIMALS

Species/Strain:	mouse/CD-1
Age at start of the examination:	5 weeks
Body weight at start of the examination:	
Males:	22.4-27.4 g
Females:	19.6-23.7 g
Number of animals ordered:	60
Number of animals involved in the study:	
Males:	20
Females:	20

3.1.1. Supplier

Charles River Hungary Ltd.
Commercial Office, H-1078 Budapest, István u. 11.
Arrival of the animals: 12nd April, 1999

3.1.2. Hygienic stage

Hygienic status: SPF

3.2. REASON FOR THE SELECTION OF ANIMALS

The mouse as the test system was selected according to the request of the Sponsor.

3.3. IDENTIFICATION AND HOUSING OF ANIMALS

The animals were identified by ear numbering technique and 5 animals was housed in every box. The code of the study, the animals' identification number, the exact time of the treatment was written onto the box where the animals were kept.

3.4. KEEPING CONDITIONS

Mode of the keeping:	conventional
Type of animal box:	I. type polypropylene
Size of box	
height:	13 cm
length:	42 cm
width:	15 cm
Number of animal per box:	5
Animal room:	rodent room

3.4.1. Environmental conditions

Air exchange:	15 times/hour
Temperature:	22 ± 3 °C
Relative humidity:	30-80 %
Lighting:	artificial, 12-hour light-dark cycles

The temperature and the relative humidity was continuously recorded by thermohygrograph.

3.4.2. Feed

The animals were fed ad libitum with Charles River VRF1 autoclaved mouse and rat diet.

3.4.2.1. Manufacturer

Altromin GmbH, 32791 Germany Lange Str. 42.

3.4.2.2. Identification

The diet was identified according to the manufacturing date.

Lot number of the diet: 1343.

3.4.2.3. Chemical analysis

Regularly controlled by the Manufacturer.

Number of the examination certificate: 30316/99-ALTR. (02.03.99),
Z01315/99 (27.01.99).

3.4.2.4. Microbiological controlling

Regularly controlled by the Manufacturer.

3.4.2.5. Feeding

Ad libitum.

3.4.3. Drinking

During the study the animals were consumed tap water ad libitum via drinking bottles.

3.4.3.1. Chemical analysis

Chemical analysis of the drinking water was controlled yearly by ÁNTSZ.

3.4.3.2. Microbiological controlling

Microbiological controlling of the drinking water was controlled yearly by ÁNTSZ.

3.4.4. Litter

During the study the animals were kept on litter of low germ.

3.4.4.1. Manufacturer

JRS Faserstoff-Werke, 73494 Germany, Ellwangen-Holzmühle

3.4.4.2. Microbiological controlling

Regularly controlled by the Manufacturer.

3.5. ACCLIMATIZATION PERIOD

The animals were observed for a week (7 days) prior to the initiation of the study. Only apparently healthy animals, free from abnormal clinical signs, were used in the study.

3.6. RANDOMIZATION

The randomization of the animals was made 3 days before beginning of the treatment by a randomization computer program based on the individual body weight of animals.

Date of randomization: 19th April, 1999

4. STUDY DESIGN

4.1. DOSES, GROUP DIVISION

Group	Dose mg/kg b.w.	No. of animal		Identification No.	
		male	female	male	female
Control (D ₀)	0	10	10	1-10	11-20
AVEMAR (D ₁)	2000	10	10	21-30	31-40

4.2. REASON FOR DOSE SELECTION

Based on discussion with the Sponsor the test substance was considered to be nontoxic, so the limit-test was adopted. Accordingly, the dose was 2000 mg/kg b.w., and it was given orally. The Sponsor did not want pilot study. We used distilled water as control substance, the dosage rate of which was 0.5 ml/20g b.w.

5. TREATMENT

5.1. MODE OF APPLICATION

Orally, via orogastric tube.

5.2. REASON FOR APPLICATION ROUTE

According to the request of the Sponsor.

5.3. FREQUENCY AND DURATION OF THE TREATMENT

Single administration.

5.4. VOLUME

40 mg/20 g b.w./0.5 ml.

5.5. PROCESS OF THE TREATMENT

Based on the request of the Sponsor the animals were not starved before the treatment.

The animals were fixed with hand. The mice were treated (between 9 and 11 hours) with the dose of the test substance (see 4.1.) once orally via orogastric tube. In the control group (D0) we used distilled water at a dose of 0.5 ml/20 g b.w.

Date of treatment: 21st April, 1999

6. POST-TREATMENT PERIOD

The animals were observed for 14 days after the treatment.

Duration of post-treatment period: 22th April - 5th May, 1999

7. EXAMINED PARAMETERS

7.1. MEASURE OF BODY WEIGHT

The animals were weighed at the arrival, on the day of randomization, before the treatment, and 24 hours after the treatment.

At the 24-hour measurement we observed loss of weight in a few animals, therefore we continued the daily weighing of all animals until increase of body weight.

The animals were also weighed on the day 7 and 14 of the post-treatment period, and before of pathological examination.

7.2. GENERAL STATUS, BEHAVIOUR AND CLINICAL SYMPTOMS

The clinical signs were observed during the first 6 hours after treatment. During this period the animals were deprived from food and only drinking water was given ad libitum.

During the post-treatment period the mice were observed twice daily.

The type, intensity, time of appearance and duration of clinical signs were recorded.

The observation included: the state of the skin, fur, eyes and mucous membranes, respiratory function, circulation, autonomic nervous function, somatomotor activity, trembling, salivation, diarrhoea, and behaviour of animal.

7.3. LETHALITY

The lethality was recorded in the first 6 hours after the treatment and in the post-treatment period twice daily.

7.4. PATHOLOGICAL EXAMINATION

At the end of the post-treatment period all animals were exterminated by intraperitoneal application of diluted Nembutal Injection, and postmortem examination was performed.

Date of necropsy: 6th May, 1999

8. STATISTICAL ANALYSIS

During the randomization we used the t-test for the significance analysis. The results of statistical analysis was not significant ($P < 0.05$), so we did not carry out further statistical test.

The calculation of the average and standard deviation value of the body weight data was done on a Pentium-100MHz-640 KB computer. The statistical analysis of the data was made by GraphPAD InStat computer program. The calculation of difference between group was performed by Paired two-tailed t-test.

9. PROCEDURES

The examinations were performed according to the prescriptions of the Standard Operation Procedures of the Department of Pharmacology & Toxicology of The University of Veterinary Science.

10. ARCHIVATION

The study plan, the documents and any information in connection with the study, and the final report will be stored for 10 years in the Archives of the Department of Pharmacology & Toxicology. The control sample of the test substance will be stored until the end of expiry date + 1 year. After archivation time the archived documents will be given to the Sponsor.

11. DEVIATION FROM THE PROTOCOL

In the SOP of euthanasia of experimental animals (SOP No. TOX-ÁLT-011) being dose (0.2 ml/20 g b.w.) of diluted Nembutal Injection (1:9 ratio) was suitable for only deep anaesthesia, therefore we used twofold dose, and dilution of 1:1 ratio, respectively.

12. RESULTS

12.1. Clinical symptoms

There were no abnormal clinical signs neither in control group nor in the treated group (AVEMAR, dose: 2000 mg/kg b.w.) of both sexes after the treatment and during the post-treatment period (Table 1-2).

12.2. Lethality

There was no lethality neither in control group nor in the treated group (AVEMAR, dose: 2000 mg/kg b.w.) after the treatment and during the post-treatment period (Table 1-2).

12.3. Body weight

The animals were weighed individually after the treatment at 24 hours. During the 24-hour weighing we observed loss of body weight either in control group

(male: No. 1; female: No. 12) or in the treated group (male: No. 23, 26; female: No. 32), therefore we carried on the daily weighing of all animals until increase of body weight.

The body weight of male No. 1 (control), male No. 26 and female No. 32 (treated) became regulated by the day 2 after the treatment. The body weight of male No. 23 treated with AVEMAR (2000 mg/kg b.w.) and that of the female No. 12 was even higher than prior to medication on day 4 and 5 after treatment, respectively.

In the remaining animals loss of body weight was not observed at the 24-hour measuring, however there was a fluctuation in body weight of both control and treated groups throughout the observation period.

The animals were also weighed on the day 7 and 14 of the post-treatment period. The weighing data are listed in Table 3-6.

There was no significant difference between the two groups of both sexes in different weighing time.

12.4. Pathological examination

In the pathological examination, which was done at the end of the 14-day post-treatment period, there was no macroscopic lesion in the animals (Table 7-8).

12.5. Evaluation of the results

The dose of 2000 mg/kg b.w. of AVEMAR did not cause lethality and abnormal clinical signs in male and female mice.

The experienced reduction of body weight in a few animals which was also observed in the control group, did not assign to the effect of the test substance.

The oral LD₅₀ value of AVEMAR in male and female mice is:

LD₅₀ > 2000 mg/kg

Budapest, 3rd June, 1999


József Lehel, DVM,
Ph.D.
Study Director

Table 1

CLINICAL SYMPTOMS AND LETHALITY DURING THE EXAMINATION IN MALES

Identifi- cation No.	Dose	*After treatment	Days of post-treatment period													
			1	2	3	4	5	6	7	8	9	10	11	12	13	14
1	Control	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
2		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
3		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
4		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
5		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
6	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
7	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
8	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
9	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
10	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
21	2000 mg/kg	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
22		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
23		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
24		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
25		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
26	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø		
27	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø		
28	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø		
29	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø		
30	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø		

* = During the first 6 hours after the treatment
 NS=No clinical signs Ø=There was no lethality

Table 2

CLINICAL SYMPTOMS AND LETHALITY DURING THE EXAMINATION IN FEMALES

Identi- fication No.	Dose	*After treatment	Days of post-treatment period													
			1	2	3	4	5	6	7	8	9	10	11	12	13	14
11	Control	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
12		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
13		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
14		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
15		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
16		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
17		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
18		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
19		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
20		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø
31	2000 mg/kg	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
32		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
33		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
34		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
35		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	
36	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø		
37	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø		
38	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø		
39	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø		
40	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø	Ø		

* = During the first 6 hours after the treatment
 NS=No clinical signs Ø=There was no lethality

Table 3

INDIVIDUAL BODY WEIGHT OF MALES DURING THE EXAMINATION

Identification No.	Dose	BODY WEIGHT (g)							
		Before treatment	After treatment						
			24 h	day 2	day 3	day 4	day 5	day 7	day 14
1	Control	22.4	22.2	22.9	22.5	23.2	23.9	24.5	26.7
2		24.6	25.7	26.2	25.3	26.6	27.4	28.4	30.3
3		23.7	23.7	24.2	24.1	24.6	25.3	26.2	29.5
4		22.9	23.7	23.7	23.1	24.0	24.8	25.6	27.9
5		27.2	28.5	28.9	29.2	29.9	30.9	31.8	34.7
6		22.9	24.2	24.0	23.9	24.7	25.3	26.4	29.7
7		24.8	25.4	25.4	25.4	26.1	26.6	27.0	30.2
8		24.9	25.8	25.8	25.5	26.1	27.1	27.6	30.2
9		25.0	26.6	26.9	27.1	28.3	29.0	30.2	32.8
10		25.7	26.5	26.8	26.5	27.5	28.4	29.5	30.9
21	2000 mg/kg	23.5	23.8	23.9	23.8	24.4	24.9	26.4	28.1
22		26.6	27.2	27.4	26.7	27.5	28.2	29.9	33.4
23		23.5	22.9	23.3	23.4	24.0	26.2	25.5	28.4
24		25.6	26.2	26.7	26.2	26.9	27.3	27.8	30.2
25		26.7	26.9	27.6	27.3	28.2	28.9	30.4	33.1
26		27.4	26.9	28.4	28.0	28.8	29.8	31.5	34.1
27		22.8	23.0	23.4	22.8	23.7	24.6	25.4	28.2
28		25.6	26.0	26.3	26.2	27.1	28.3	29.1	31.4
29		25.1	25.2	25.6	25.4	26.3	27.3	28.6	31.9
30		23.3	24.0	24.2	23.7	24.2	25.2	26.3	29.0

Table 4

INDIVIDUAL BODY WEIGHT OF FEMALES DURING THE EXAMINATION

Identification No.	Dose	BODY WEIGHT (g)							
		Before treatment	After treatment						
			24 h	day 2	day 3	day 4	day 5	day 7	day 14
11	Control	21.5	22.5	22.2	21.6	22.1	22.6	22.7	24.2
12		20.0	19.7	19.2	19.2	19.9	21.0	20.7	23.0
13		21.9	21.9	21.9	21.7	22.4	22.3	22.8	24.5
14		20.9	21.3	22.2	22.0	22.5	22.0	23.2	24.9
15		21.3	22.9	23.4	22.8	22.4	23.0	23.8	25.9
16		22.1	22.9	23.2	22.8	22.2	23.0	24.1	25.3
17		20.8	21.1	21.2	20.8	21.4	22.6	22.4	23.6
18		20.5	21.2	21.5	20.4	20.5	21.6	21.6	23.2
19		23.7	25.5	24.8	24.0	24.9	25.9	25.6	30.2
20		21.5	21.9	21.6	20.8	21.5	22.3	22.3	25.7
31	2000 mg/kg	20.8	21.8	20.5	20.2	20.8	21.6	21.2	23.8
32		21.4	21.2	21.7	21.5	22.1	22.5	22.0	23.2
33		22.3	23.9	23.0	22.5	23.7	24.2	24.2	26.5
34		21.1	21.9	22.2	22.4	21.9	22.5	23.7	24.5
35		22.4	23.6	22.4	22.2	23.1	24.5	24.3	26.0
36		20.3	20.6	20.5	20.3	21.3	20.7	21.8	23.7
37		21.3	22.3	22.3	22.4	21.9	22.3	23.5	24.1
38		19.6	19.7	19.5	19.5	20.3	19.8	21.0	21.9
39		20.4	20.9	21.1	21.0	21.5	20.9	22.2	24.3
40		21.6	22.3	22.9	21.6	22.0	22.7	23.9	25.9

Table 5

AVERAGE BODY WEIGHT OF MALES DURING THE EXAMINATION

Dose	Before treatment	BODY WEIGHT (g)						
		24 h	day 2	day 3	day 4	day 5	day 7	day 14
CONTROL								
X	24.41	25.23	25.48	25.26	26.10	26.87	27.72	30.23
± SD	1.47	1.82	1.82	2.00	2.07	2.15	2.26	2.25
2000 mg/kg								
X	25.01	25.21	25.68	25.35	26.11	27.07	28.09	30.78
± SD	1.64	1.66	1.88	1.81	1.89	1.79	2.15	2.31

There was no significant difference ($P < 0.05$) between the two groups in the different weighing time.

Table 6

AVERAGE BODY WEIGHT OF FEMALES DURING THE EXAMINATION

Dose	Before treatment	BODY WEIGHT (g)						
		24 h	day 2	day 3	day 4	day 5	day 7	day 14
CONTROL								
X	21.42	22.09	22.12	21.61	21.98	22.63	22.92	25.05
± SD	1.02	1.54	1.49	1.39	1.35	1.30	1.37	2.07
2000 mg/kg								
X	21.12	21.82	21.61	21.36	21.86	22.17	22.78	24.39
± SD	0.88	1.30	1.17	1.07	1.00	1.48	1.27	1.41

There was no significant difference ($P < 0.05$) between the two groups in the different weighing time.

Table 7

PATHOLOGICAL FINDINGS OF MALES

Identification No.	Dose	Pathological changes
1	Control	there was no macroscopic lesion
2		there was no macroscopic lesion
3		there was no macroscopic lesion
4		there was no macroscopic lesion
5		there was no macroscopic lesion
6		there was no macroscopic lesion
7		there was no macroscopic lesion
8		there was no macroscopic lesion
9		there was no macroscopic lesion
10		there was no macroscopic lesion
21	2000 mg/kg	there was no macroscopic lesion
22		there was no macroscopic lesion
23		there was no macroscopic lesion
24		there was no macroscopic lesion
25		there was no macroscopic lesion
26		there was no macroscopic lesion
27		there was no macroscopic lesion
28		there was no macroscopic lesion
29		there was no macroscopic lesion
30		there was no macroscopic lesion

Table 8

PATHOLOGICAL FINDINGS OF FEMALES

Identification No.	Dose	Pathological changes
11	Control	there was no macroscopic lesion
12		there was no macroscopic lesion
13		there was no macroscopic lesion
14		there was no macroscopic lesion
15		there was no macroscopic lesion
16		there was no macroscopic lesion
17		there was no macroscopic lesion
18		there was no macroscopic lesion
19		there was no macroscopic lesion
20		there was no macroscopic lesion
31	2000 mg/kg	there was no macroscopic lesion
32		there was no macroscopic lesion
33		there was no macroscopic lesion
34		there was no macroscopic lesion
35		there was no macroscopic lesion
36		there was no macroscopic lesion
37		there was no macroscopic lesion
38		there was no macroscopic lesion
39		there was no macroscopic lesion
40		there was no macroscopic lesion