SC-18862
52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

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October 10, 1972

Pathology—Toxicology
Project No. 856ot70

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INTRODUCTION

In this toxicity study SC-l8862, a nutritive artificial sweetening agent, was administered orally in the milk formula to infant Rhesus monkeys for 52 consecutive weeks. SC-18862 is a dipeptide and is split to its constituent moieties by peptidases in the digestive tract.

This study was designed to determine the adverse effects, If any, of SC-18862 ingestion on the neonatal Rhesus monkey, and also whether all such effects were identical in nature and magnitude to those produced by an equimolar molar quantity of L-phenylalanine.[1]

A research project involving repeated daily oral administration of any agent to a sizable population of baby monkeys, commencing at birth and continuing uninterrupted throughout the 1st year of life, is a major undertaking fraught with hazard, even for the partially initiated.

Thus, this study was performed at the Primate Research Center, Madison, Wisconsin under the direction of the late Dr. Harry A. Waisman, Prof. of Pediatrics and Director, Joseph P. Kennedy Memorial Laboratories. His established expertise in research involving phenylalanine and the neonatal Rhesus monkey was invaluable, and his unfortunate demise necessitated revision of the initial objectives of this study. This report does provide valuable physical examination and clinical laboratory data enabling comparison of SC-l8862 with known effects of L-phenylalanine.

METHODS

Material evaluated

SC-l8862 is a fine white powder with the chemical name L-aspartyl, L-phenylalanine methyl ester. Three lots (74O2C, 75060B, 74060) were used throughout this study. These lots contained from 0.2 to 1% of SC-l9l92 (Diketopiperazine; DKP), a conversion product of SC-18862.

Animals, housing and diet

Infant Rhesus monkeys (Macaca mulatta) from full-term, normal pregnancies were separated from their mothers within 6 hours after birth and transferred to individual heated cages.

During the first 24 hours of life, the infants were fed a 10% glucose solution at four-hour intervals; during the second day, this diet was supplemented with equal volumes of a commercial milk preparation (Similac, Ross Laboratories, Columbus, Ohio; Control diet, CD). Thereafter, the infants were fed CD ad libitum at four hour intervals until they were placed on the experimental liquid formula.

During the training period, the infant was gently wrapped in a cloth diaper and held while fed from a toy nursing bottle and nipple. Four feedings per day was the preferred number for this experiment. Later, between days 12 and 30, the animals were weaned and fed from a small cup: on or after day 31 they were fed from a large cup.

Compound administration.

Similac formula was supplemented with SC-18862 on a "phenylalanine equivalent" basis: 1.83 g L-aspartyl, L-phenylalanine methyl ester contains 1.0 g L-phenylalanine. The SC-18862 concentration was incrementally increased, based on acceptance by the infant.

Age		Code	Aspartyl Phenylalanine	= L-Phenylalanine
Day 3-	Day 9	1/8th	.0029 g/cc	.0016 g/cc
10-	19	1/4	.0057 g/cc	.0031 g/cc
20-	29	3/8	.0086 g/cc	.0046 g/cc
30-	119	1/2	.0114 g/cc	.0063 g/cc
120-	179	5/8	.0143 g/cc	.007S g/cc
180	229	3/4	.0171 g/cc	.0094 g/cc
230	269	7/8	.02 g/cc	.011 g/cc
270	365	1	.022 g/cc	.012 g/cc

Milk intake was carefully recorded for each feeding, so that the amount of SC-18862 consumed per day per kg of body weight could be calculated, allowance being made for spillage. When the animals were 3 months old, a quarter of an apple and a quarter of an orange were placed in the cage once a day. The infant monkeys were fed SC-18862 with the milk formula. Water was available ad libitum. Animal quarters were air-conditioned with thermostats set to maintain a room temperature of 72^0F; artificial fluorescent lighting was provided on a 14 hour daily photoperiod.

Experimental design.

Seven newborn Rhesus monkeys, five males (M34, M38, M64, M79, P53) and two females (N14, P60), were randomly divided into three groups.

Treatment Group	Intended Dosage g/kg/day	Multiple of Estimated Daily Human Intake*	Animal No.	Sex	Date of Birth	Start Supplement Age (Days)
Low	1	33	P53	M	08-28-70	6
			P60	F	09-06-70	3
Medium	3	100	M64	M	03-19-70	3
			M79	M	04-05-70	3
			Nl4	F	04-26-70	2
High	4-6	133-200	M34	M	01-05-70	9
			M38	M	01-13-70	1

* Based on 30 mg/kg oral intake daily to a 27 kg child.

The treatment was arbitrarily terminated by the late Dr. Waisman's staff as indicated below.

Treatment Group	Animal No.	Treatment Initiated	Treatment Terminated	Total Days on Treatment
Low	P53	09-03-70	03-31-71	210
	P60	09-09-70	03-31-71	204
Medium	M64	03-23-70	03-18-71	363
	M79	04-08-70	04- 4-71	362
	N14	04-28-70	04-25-71	363
High	M34	01-14-70	01-05-71	357
	M38	01-14-70	10-20-70	279

Physical examinations and observations.

Animals were observed daily at the time of dosing and intermittently between dosing periods for survival arid behavioral changes. Body weights were recorded each day in the morning. Head circumference and body length (crown to heel length) were recorded at 4 week intervals. An evaluation of general motor and behavioral activity, locomotion, external appearance of teeth, nose, eyes, ears, perineum, hair coat and digital palpation for tissue masses was conducted immediately prior to the initiation of compound administration, and subsequently concurrent with each body weigh: measurement. Unusual signs, including indications of systemic pharmacologic or toxicologic effects, were routinely recorded at this time and whenever warranted.

Clinical laboratory procedures.

Hematologic and clinical chemical examinations which were performed on blood specimens of all animals, were collected via the saphenous vein at 3, 6, 9 and 12 months of compound administration.

Hematology.

The following hematologic parameters were measured:

Parameter	Method
Hematocrit (micro)	Micro method [2]
Hemoglobin	Cyanrnethemoglobin [3]
Total RBC count	Coulter Counter [4]
Total W3C count	Coulter Counter [4]
Diff. WBC count	Smear [5]

Clinical chemistry.

The following (plasma chemistry) parameters were measured for all groups:

Parameter	Method
Blood (plasma) urea nitrogen	Urograph method [6]
Uric acid	Brown [7]
Glutamic oxalacetic transaminase	Reitman & Frankel [8]
Alkaline phosphatase	Klein et al.[9]
Bilirubin	Malloy & Evelyn [10, 11]
Glucose	Nelson & Somogyi [12, l3]
Calcium	Barr [14]
Inorganic phosphate	Fiske & Subbahow [15]
Cholesterol	Abell et al. [16]
Total protein	TS Meter [17]
Phenylalanine	Undenfriend & Cooper [18]
Tyrosine	La Du & Michael [19]

Serum phenylalanine and tyrosine were monitored twice a week for the first 13 weeks; weekly for the next 17 weeks and once every two weeks thereafter.

Urinalysis.

Spontaneously voided urine specimens from individually housed monkeys were collected at 3, 6, 9 and 12 months of treatment. The following parameters were measured.

Parameter	Method
Specific gravity	Total solids meter
pH	Labstix (Ames)
Occult blood	Labstix (Ames)
Protein	Labstix (Ames)
Glucose	Labstix (Ames)
Ketones	Labstix (Ames)
Bilirubin	Labstix (Ames)
Phenylketones	Phenistix (Ames)

RESULTS

ANTEMORTEM OBSERVATIONS

The availability of acceptable historical and contemporary data on untreated control monkeys from the Waisman group reduced the necessity of a concurrent control group. The extremely limited availability of newborn Rhesus, as well as limitations in adequately skilled laboratory personnel, likewise contributed to our decision to eliminate the requirement of a concurrent control group in this study.

For comparative purposes the normal range of values from 14 historical control monkeys is superimposed on **Figures 1-9.**

Compound consumption.

The treatment of monkeys with SC-l8862 was initiated on the basis of availability of newborn monkeys as indicated on page 3. The sudden demise of Dr. Waisman necessitated termination of the study. At that point in time, the medium and high dose monkeys had completed 52 weeks of treatment, and the low dose monkeys had completed 29-30 weeks of treatment.

Mean values for SC-18862 ingestion by the low and medium dose group animals over the treatment period (**Table 1**) were within 5% of the proposed doses of 1.0 and 3.0 g/kg. The intended dosage of SC-l8862 for the high dose group was

4 to 6 g/kg; because of an unanticipated decrease in the intake of milk formula, presumably due to the intense sweetness of SC-18862, the realized mean intake of SC-l8862 over the entire study was 3.6 g/kg (range 1.21 to 5.33 g/kg). Hence, the SC-l8862 intake of high dose group animals was not notably different from the medium dose group animals. Irrespective of the actual intake of SC-l8862 levels, the results of this study are presented as data for the low dose group (0.97 g/kg intake), medium dose group (3.01 g/kg intake), and high dose group (3.62 g/kg intake), according to the original placement of animals within each group.

As pointed out in the methods section, the SC-18862 lots employed in this study contained 0.2 to 1% SC-19192, a conversion product of SC-1S362.

The actual group mean daily ingestion of SC-19l92 (**Table 2**) was computed from the actual intake of SC-18862 and from analytical data (Quality Control Department, Searle Laboratories) indicating the SC-19l92 content of each individual lot of SC-l8862 employed in this study. The group mean intake of SC-19192 over the entire study was 4.84, 15.07 and 18.12 mg/kg/day for the low, medium and high dose groups, respectively.

Growth and food consumption.

Absolute body weight and weight gain (g/kg/day) of individual monkeys in each group are presented in Figures **1**, **2** and **3**. Body weight gain per ml milk formula consumed and actual intake of liquid diet over the 52 week treatment period are depicted in Figures **4**, **5** and **6**.

The body weight in kilograms was within normal limits for P60, M64 and M34. One high dose monkey, M38, and two medium dose monkeys, N14 and M79, showed slightly lower body weight, but there seemed to be a leveling off in the weight as the animals approached one year on the diet.

Low dose monkey P53 exhibited evidence of physical deficiencies, apparently congenital in origin, shortly after birth. The animal was examined by selected consultants, and its suitability for inclusion in the study was questioned. A precise account of their findings is not available. The animal was continued on study irrespectively, however, since the supply of baby Rhesus was very limited. Subsequent poor growth of this animal (**Fig. 1**) was due to inappetance and may reflect the initial difficulties. Relative weight gain (g/kg/day) of all treated animals except monkey P53 was comparable to historical controls.

Rate of growth expressed per unit of diet intake (Figs**. 4**, **5**, **6)** was within normal limits despite the falling off of absolute body weight (Figs. **1**, **2**, **3**). This indicates that the dipeptide was utilized efficiently and did not effect the efficiency of food conversion.

There was a marked decrease in total intake of milk formula in all the treated animals (Figs. **4**, **5**, **6**). This could be attributed to the intense sweetness (200 x sucrose) of the dipeptide.

Individual daily body weight and milk formula intake of each experimental monkey may be found in Figures **1**, **2** and **3**

Body length of all treated animals is essentially within the historical control

range; head circumference is likewise within historical control range for 1/2 low level, 1/3 medium level and 2/2 high level monkeys, but is below control level in the remaining animals (Figs. **7**, **8**, **9**). The decrease in head circumference during treatment in low dose monkey P53 (**Fig. 7**) could be attributed to a proportional decrease in the relative weight gain (g/kg/day) of this monkey. Underdevelopment of this monkey is presumably related to the physical deficiencies observed at birth. An apparent decrease in the head circumference observed during treatment in two medium dose monkeys, M79 and N14 (**Fig. 8**), is attributed to a relatively lower head circumference at birth.

Observations, physical and behavioral signs.

All animals in the medium and high dosage groups exhibited seizure activity. Seizures were observed for the first time following 218 days of treatment. thereafter, sporadic convulsions occurred inconsistently at various times during the treatment period. Seizures occurred most frequently during physical handling of the animal for body weight measurements. The convulsions were of grand mal type similar to those induced by feeding L-phenylalanine to infant monkeys.

All animals in the medium and high dosage groups contracted a Shigella infection at various times during the treatment period. In an effort to treat the Shigella infection, these anima1s received appropriate antibiotic and intravenous fluid therapy.

One monkey, M38, of the high dose group, died after 300 days of treatment. The cause of death was not determined.. All other animals survived the treatment period.

General posture and locomotion, pelage, body orifices and excretions were otherwise unremarkable.

Clinical laboratory findings.

Hematology.

Individual values of hematology parameters evaluated are presented in **Tables 3** and **4.** The Primate Research Center, Madison, Wisconsin, supplied mean hematologic values of 16 historical control monkeys of the same age group as the experimental animals; these values are presented in **Table 5.** In general, hematologic values for individual treated animals were unremarkable; no biologically significant deviation from control ranges was observed. Statistical analysis was not performed due to the lack of individual values for the historical controls.

Clinical chemistry.

Individual values of clinical chemistry parameters evaluated are presented in **Table 6.** The Primate Research Center, Madison, Wisconsin, supplied clinical chemistry values of 5 historical control monkeys of the same age group as the experimental animals; these values are presented in **Table 7.** Clinical chemistry values from SC-18862 fed animals, in general, were comparable with the historical

control values. No obvious compound related changes were evident.

Serum phenylalanine and tyrosine.

The serum phenylalanine and tyrosine values from SC-18862 fed animals were monitored at frequent intervals and are depicted in **Figures 10, 11 and 12.** For comparative purposes the range of serum phenylalanine and tyrosine values from 4 historical positive control monkeys fed 2 to 2.5 g/kg/day L-phenylalanine are superimposed in the Figures. In the low dose (1 g/kg/day SC-18862) animals there was no appreciable change in the serum phenylalanine and tyrosine levels (**Fig. 10**). There was a significant increase in serum phenylalanine and tyrosine values in the medium and high dose monkeys (**Figs. 11 and 12**). These increased serum phenylalanine and tyrosine values are comparable to positive control L-phenylalanine fed animals. It is interesting to note that in the medium and high dose groups very high levels of serum phenylalanine were achieved after 200 days of feeding SC-l8862 (**Figs. 11 and 12**). As mentioned earlier, the convulsions in the medium and high dose animals were observed initially at 218 and 219 days on the experiment. Hence, the convulsions In the monkeys are correlated with and can be attributed to high serum phenylalanine levels. In the low dose monkeys (1 g/kg/day) serum phenylalanine levels were at a basal level (**Fig.10**) and no convulsions had been observed when the study was terminated (30 weeks of treatment).

Following the termination of treatment, medium and high dose monkeys were kept under observation for 3 months on powdered Similac. No further convulsions were detected during this period.

Serum phenylalanine and tyrosine values of individual animals monitored at various times in the study may be found in the Appendix.

Urinalysis.

The results of urinalyses performed on individual monkeys are presented in **Table 8**. No meaningful variations were consistently present in the parameters measured: pH, Sp. Gr., blood, protein, glucose, ketones, bilirubin. There was a significant increase in the urinary excretion of phenylketones in the medium and high dose group monkeys. This was consistent with a concomitant increase in serum phenylalanine levels in these monkeys.

P0STM0RTEM OBSERVATIONS

Animals were not available for sacrifice and necropsy at the termination of compound administration, due to a shortage of personnel and supervision following Dr. Waisman's death. Likewise, necropsy data on the one non-survivor, high dose monkey M38 that died after 300 days of compound ingestion, was lost for similar reasons.

SUMMARY AND CONCLUSIONS

A 52 week oral toxicity study of SC-18562 was conducted, employing oral administration of the compound to newborn Rhesus monkeys. SC-18862 was mixed with Similac milk formula and fed four times daily. Mean daily dosage levels of

0.97, 3.01 and 3.62 g/kg were attained incrementally. These levels are multiples of 32, 100 and 120 times the estimated maximal human daily intake (30 mg/kg/day for 27 kg child). Physical examinations were performed regularly. Body weight and milk formula intake were recorded monthly. Hematology and clinical chemistry parameters were evaluated every three months. Serum phenylalanine and tyrosine levels were monitored at frequent intervals.

Survival was 100% in all treated groups except the high dose group; one monkey, M38 in the high dose group, died after 300 days of treatment. The cause of death is unknown. Animals in both the medium and high dose groups experienced grand mal convulsions after about 220 days of treatment. Similar convulsions may be induced in the monkey by feeding L-phenylalanine alone in equimolar quantities[1]. Occurrence of seizures coincided with the attainment of high serum phenylalanine levels. In the low dose group (1 g/kg/day) there was no appreciable increase in serum phenylalanine; thus, convulsions would not be expected irrespective of the duration of treatment. Physical examination findings were otherwise unremarkable.

Food intake and growth rate were mildly reduced by SC-18862 treatment. The head circumference of one low dose monkey (P53) and two medium dose monkeys (M79 and Nl4) was lower than the historical control range. This was attributed to physical deficiencies evident at birth and subsequent partial inanition in the former animal, and to unusually low head circumference measurements at birth in the latter two. The head circumference of all other monkeys was within normal range. The body length of all treated monkeys was within the historical control range.

Hematology and clinical chemistry parameters were generally unremarkable in treated animals, as compared with data from historical control animals of the same age from the same laboratory. No biologically significant alterations were observed except, as mentioned earlier, there was a significant increase in serum phenylalanine and tyrosine levels at the medium and high dose levels. Urinalysis parameters were generally unremarkable, except for a significant excretion of phenylketones in both medium and high dose groups after 6 months. This increase coincided with the increase of serum phenylalanine levels. Thus, the SC-18862 treated monkeys exhibited increased serum phenylalanine levels, increased urinary phenylketone levels, and episodes of grand mal seizures in relation to the phenylalanine moiety of the compound administered. At the low dose level (1 g/kg/day), none of the above alterations were observed through *30* weeks of treatment, at which point the study terminated.

It is concluded that dietary administration of SC-18862 *to* infant monkeys starting at birth and continuing for 30 consecutive weeks at approximately 1 g/kg/day, caused no biologically meaningful alterations in physical or behavioral findings or in clinical laboratory parameters. At higher dosages a significant increase in serum phenylalanine and tyrosine levels, an increase in urinary phenylketone excretion and episodes of grand mal type seizure activity were observed at this point, and continued through the 52 weeks of treatment. Both the nature and magnitude of the changes observed were comparable to historical positive control animals fed equivalent quantities of L-phenylalanine alone.

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TABLE 1

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Consumption of SC-18862 (mg per kg per day)
(Mean Values)

Treatment Intervals (days)													
Treatment Group	0-9	10-19	20-29	30-39	40-49	50-59	60-69	70-79	80-89	90-99	100-109	110-119	120-129
Low Dose	0.77	0.95	0.97	1.09	0.98	0.99	1.00	0.99	0.98	0.94	0.91	0.98	0.97
Medium Dose	0.94	1.76	2.90	3.55	3.51	3.54	3.20	3.25	3.42	3.23	3.35	3.24	3.85
High Dose	1.21	1.82	1.83	2.36	3.21	3.68	3.37	3.54	3.67	3.31	3.20	3.22	3.24
Treatment Intervals (days)													
Treatment Group	130-139	140-149	150-159	160-169	170-179	180-189	190-199	200-209	210-219	220-229	230-239	240-249	
Low Dose	1.06	1.29	1.36	1.13	1.09	0.99	0.93	0.62	--	--	--	--	
Medium Dose	3.69	3.73	3.44	3.49	2.78	2.80	2.75	2.54	2.36	2.38	2.66	2.48	
High Dose	3.88	4.38	4.38	4.38	4.33	5.33	4.96	3.83	3.99	4.21	4.84	4.24	
Treatment Intervals (days)													
Treatment Group	250-259	260-269	270-279	280-289	290-299	300-309	310-319	320-329	330-339	340-349	350-359	360-369	Mean 0-369
Low Dose	--	--	--	--	--	--	--	--	--	--	--	--	0.97*
Medium Dose	2.37	2.04	2.19	2.43	2.94	2.77	3.05	3.65	3.19	3.02	3.07	2.88	3.01
High Dose	3.70	3.58	3.75	4.32	4.83	4.33	3.30	4.04	3.37	3.11	3.31	2.50	3.62

* Mean for 0-209 days.

TABLE 2

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Consumption of SC-18862 (mg per kg per day)
(Mean Values)

	Treatment Intervals (days)												
Treatment Group	0-9	10-19	20-29	30-39	40-49	50-59	60-69	70-79	80-89	90-99	100-109	110-119	120-129
Low Dose	3.84	4.75	4.85	5.45	4.90	4.95	5.01	4.97	4.90	4.71	4.54	4.92	4.87
Medium Dose	4.71	8.80	14.48	17.73	17.56	17.71	16.02	16.25	17.11	16.17	16.74	16.22	19.26
High Dose	6.03	9.08	9.15	11.82	16.07	18.42	16.84	17.68	18.37	15.54	16.01	16.08	16.21
	Treatment Intervals (days)												
Treatment Group	130-139	140-149	150-159	160-169	170-179	180-189	190-199	200-209	210-219	220-229	230-239	240-249	
Low Dose	5.29	6.46	6.82	5.67	5.44	4.93	4.67	3.10	1.54	--	--	--	
Medium Dose	18.45	18.67	17.20	17.47	13.88	13.99	13.76	12.72	11.82	11.88	13.31	12.42	
High Dose	19.41	21.88	21.92	21.90	21.63	26.67	23.46	19.14	19.93	21.03	24.18	21.12	
	Treatment Intervals (days)												
Treatment Group	250-259	260-269	270-279	280-289	290-299	300-309	310-319	320-329	330-339	340-349	350-359	360-369	Mean 0-369
Low Dose	--	--	--	--	--	--	--	--	--	--	--	--	4.84
Medium Dose	11.87	10.18	10.94	12.17	14.72	13.87	15.27	18.27	15.95	15.12	15.33	14.42	15.07
High Dose	18.49	17.92	18.74	21.58	24.16	21.66	16.52	20.19	16.85	15.56	15.54	12.48	18.12

* Mean for 0-209 days.

TABLE 3

SC- 18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Hematology: Red Cell Data

Months of Treatment		3			6			9			12		
Treatment Group	Animal No	Hgb (g%)	Hct (%)	RBC ($x10^6$/cm)	Hgb (g%)	Hct (%)	RBC ($x10^6$/cm)	Hgb (g%)	Hct (%)	RBC ($x10^6$/cm)	Hgb (g%)	Hct (%)	RBC ($x10^6$/cm)
Low Dose	P3	11.8	39	6.14	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	P60	13.0	40	5.49	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Medium Dose	M64	11.8	37.5	4.93	11.2	35.0	5.38	13.5	41.0	6.18	N.D.	N.D.	N.D.
	M79	11.2	39.0	5.36	11.6	38.0	N.D.	11.5	35.5	5.27	N.D.	N.D.	N.D.
	N14	10.8	35.0	5.05	10.8	36.0	N.D.	11.8	38.0	5.78	N.D.	N.D.	N.D.
High Dose	M34	11.2	35.0	5.30	11.7	38.0	5.74	8.8	27.0	N.D.	11.5	37.0	5.40
	M38	12.8	39.0	4.88	11.0	36.0	4.65	10.1	30.0	5.20	N.D.	N.D.	N.D.

N.D. = No Data

TABLE 4

SC- 18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Hematology: White Cell Data

Months of Treatment		**3**						**6**					
Treatment Group	Animal No	Total WRC (10^3/cm)	PMN (%)	Lym (%)	Mon (%)	Eos (%)	Ret (%)	Total WRC (10^3/cm)	PMN (%)	Lym (%)	Mon (%)	Eos (%)	Ret (%)
Low Dose	P3	10.8	43	50	6	1	1.2	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	P60	8.6	5	94	1	0	0.4	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Medium Dose	M64	5.0	11	85	0	4	0.6	12.0	56	44	0	0	0.2
	M79	8.8	37	63	0	0	1.2	12.4	5	94	0	1	0.2
	N14	14.0	68	31	0	0	0.2	10.5	27	72	0	1	0.6
High Dose	M34	5.74	9	90	0	1	1.0	84	14	85	1	0	2.0
	M38	9.7	11	87	1	0	0.8	12.7	27	73	0	0	0.6
Months of Treatment		**9**						**12**					
Low Dose	P3	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	P60	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Medium Dose	M64	5.8	49	49	1	1	0.2	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	M79	13.9	74	24	0	2	0.4	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	N14	10.6	50	49	1	0	0.8	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
High Dose	M34	7.7	32	63	4	1	3.0	13.8	72	28	0	0	N.D.
	M38	9.7	18	81	0	1	0.8	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.

Press BACK to Return to Text - Return to Index

TABLE 5

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Hematology Data
Historical Control Values of Infant Monkeys
(Primate Center, Madison, Wisconsin)

Age Months		Hgb (g%)	Hct (%)	RBC ($x10^6$/cm)	WBC ($x10^3$/cm)	PMN (%)	Lym (%)	Mon (%)	Eos (%)	Ret (%)
3	Mean	13.3	39.5	5.4	9.7	17.6	78.7	2.5	.9	1.4
	S.D.	.8	1.8	.4	2.9	8.4	8.6	2.1	1.2	.7
	+	14.1	41.3	5.8	12.6	26.0	87.3	4.6	2.1	2.1
	-	12.5	37.7	5.0	6.8	9.2	70.1	.4	.3	.7
6	Mean	13.7	40.9	5.1	9.6	29.6	66.6	2.0	1.3	1.0
	S.D.	1.1	3.6	.5	1.9	11.3	11.6	1.2	1.0	.2
	+	14.8	44.5	5.6	11.5	40.9	78.2	3.2	2.3	1.2
	-	12.6	37.3	4.6	7.7	18.3	55.0	.8	.3	.8
9	Mean	13.4	41.1	5.2	9.9	35.6	59.8	1.9	2.0	1.0
	S.D.	1.0	3.4	.6	3.5	18.6	17.2	1.8	2.0	.4
	+	14.4	44.5	5.8	13.4	54.2	77.0	3.7	4.0	1.4
	-	12.4	37.7	4.6	6.4	17.0	42.6	.1	.0	.6
12	Mean	13.4	41.1	5.5	10.8	34.8	61.7	1.3	1.6	.7
	S.D.	.7	2.6	.6	3.3	18.1	18.3	2.0	1.3	.4
	+	14.1	43.7	6.1	14.1	52.9	80.0	3.3	2.9	1.1
	-	12.7	38.5	4.9	7.5	16.7	43.4	.7	.3	.3

Press BACK to Return to Text - Return to Index

TABLE 6

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Clinical Chemistry

Treatment Group	Animal No.	++ Ca (mg%)	Inor. Phos. (mg%P)	Glu (mg%)	Bun (mg%)	Uric Acid (mg%)	Chol (mg%)	T.P. (GM%)	Alb (gm%)	Bili (mg%)	A.P. (I.U./ml)	SGOT (I.U./ml)
3 Months of Treatment												
Low Dose	P53	9.9	5.29	75.5	9.0	0.05	147	6.7	2.28	0.13	350	32
	P60	12.0	7.75	86.0	14.5	0.12	162	6.4	4.08	0.30	350	58
Medium Dose	M64	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	M79	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	N14	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
High Dose	M34	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	M38	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
6 Months of Treatment												
Low Dose	P53	9.8	5.8	68.0	9.0	1.10	187	6.9	2.5	0.1	350	48
	P60	11.2	6.2	74.0	18.4	0.50	202	7.2	4.2	0.2	350	114
Medium Dose	M64	10.7	6.2	82.0	8.3	0.90	175	7.2	3.8	0.4	350	38
	M79	10.3	6.6	84.0	11.0	0.10	158	6.3	3.4	0.4	350	38
	N14	10.4	5.7	97.0	12.2	0.01	155	6.3	4.0	0.3	350	114
High Dose	M34	10.6	5.1	90.0	16.0	0.00	169	6.5	4.4	0.1	570	54
	M38	10.7	6.3	80.0	13.00	0.40	161	6.7	3.8	0.3	350	55

N.D. = No Data

Table 6 (Continued on next page)

TABLE 6 (Continued)

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Clinical Chemistry

9 Months of Treatment												
Low Dose	P53	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	P60	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Medium Dose	M64	11.1	6.8	74	11.2	0.2	212	7.2	4.6	0.2	350	58
	M79	12.1	6.8	115	13.7	0.6	165	7.2	4.3	0.7	350	69
	N14	11.5	6.9	88	16.5	1.2	217	7.3	4.7	0.1	350	68
High Dose	M34	10.7	7.0	80	13.2	0.7	145	6.90	4.1	0.5	350	96
	M38	10.3	6.4	100	13.2	0.2	177	6.90	3.7	0.2	350	33
12 Months of Treatment												
Low Dose	P53	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	P60	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Medium Dose	M64	10.3	6.2	75.2	12.5	1.3	225	6.9	4.3	0.1	350	96
	M79	12.1	6.6	115	14.5	1.4	170	7.4	4.6	0.00	350	39
	N14	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
High Dose	M34	10.8	7.1	82.0	11.9	0.3	225	7.4	4.2	0.1	350	45
	M38	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.

N.D. = No Data

TABLE 7

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Clinical Chemistry
Historical Control Values of Infant Monkeys
(Primate Center, Madison, Wisconsin)

Control No	++ Ca (mg%)	Inor. Phos. (mg%)	Glu (mg%)	Bun (mg%)	Uric Acid (mg%)	Chol (mg%)	T.P. (gm%)	Alb (gm%)	T. Bili (mg%)	A.P. (I.U./ml)	SGOT (I.U./ml)
Age of Monkey - 3 Months											
1	11.1	6.28	87	10.0	0.85	152	5.15	3.0	0.28	350	72.0
2	11.1	6.89	80	12.0	0.49	160	6.34	3.5	0.30	350	47.0
3	10.7	7.65	67	13.8	1.05	178	6.28	3.5	0.46	350	48.0
4	10.9	7.25	76	12.4	1.10	175	6.48	4.0	0.28	350	70.0
5	10.6	7.34	70	12.5	0.48	182	6.10	4.1	0.20	350	58.0
X	10.8	7.08	76	12.1	0.79	169	6.07	3.6	0.30	350	59.0
Age of Monkey - 6 Months											
1	11.2	6.36	85	11.8	1.45	165	5.56	3.4	0.28	350	40.0
2	9.8	5.60	75	16.4	1.20	192	6.10	3.9	0.22	-	47.0
3	13.1	7.50	135	12.3	1.60	145	7.90	3.9	0.40	-	157.0
4	11.2	6.80	70	12.5	0.00	215	6.70	4.3	0.28	-	67.0
5	10.8	7.10	108	16.0	0.25	233	6.50	4.1	0.32	-	43.0
X	11.2	6.67	95	13.8	0.90	190	6.55	3.9	0.30	-	70.8
Age of Monkey - 9 Months											
1	11.2	6.08	76	14.0	0.32	265	6.90	4.3	0.28	350	112.0
2	10.9	5.92	82	15.9	0.85	254	6.80	4.5	0.40	350	64.0
3	12.4	6.50	81	11.8	0.59	269	7.50	4.8	0.20	350	53.0
4	10.1	6.28	65	17.8	0.00	156	6.10	3.5	0.04	350	45.0
5	9.8	5.44	89	16.7	1.35	215	6.50	3.6	0.20	350	85.0
X	10.9	6.04	79	15.2	0.62	232	6.80	4.1	0.22	350	71.8
Age of Monkey - 12 Months											
1	11.8	5.05	80	16.5	1.10	230	6.60	3.8	0.00	350	110.0
2	10.9	5.80	70	14.8	0.66	210	7.40	4.0	0.00	350	60.5
3	9.0	5.00	76	11.5	0.60	136	5.75	4.0	0.10	350	51.0
4	-	-	-	-	-	-	-	-	-	-	-
5	-	-	-	-	-	-	-	-	-	-	-
X	10.6	5.28	75	14.2	0.79	192	6.58	3.9	-	350	73.8

TABLE 8

SC18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Urinalysis Data

		3 Months of Treatment								6 Months of Treatment							
Treatment Group	Animal No.	SpGr	pH	Bili	Prot	Glucose	Ketone	Blood	Pkt* (mg%)	SpGr	pH	Bili	Prot	Glucose	Ketone	Blood	Pkt* (mg%)
Low Dose	P53	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	1.008	8.0	Neg	Neg	Neg	Neg	Neg	Trace
	P60	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	1.001	8.0	Neg	Neg	Neg	Neg	Neg	Trace
Medium Dose	M64	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	1.005	6.0	Neg	Neg	Neg	Neg	Neg	100
	M79	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	1.011	8.0	Neg	Neg	Neg	1+	Neg	100
	N14	1.008	8.0	Neg	Neg	Neg	Neg	Neg	Neg	1.010	6.5	Neg	Neg	Neg	Neg	Neg	100
High Dose	M34	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	100
	M38	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	100
		9 Months of Treatment								12 Months of Treatment							
Low Dose	P53	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	P60	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Medium Dose	M64	1.010	7.0	N.D.	Trace	Neg	Neg	Neg	100	1.010	8.0	Neg	Trace	Neg	Neg	Neg	100
	M79	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	1.007	8.0	Neg	Neg	Neg	Neg	Neg	100
	N14	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
High Dose	M34	1.015	8.0	Neg	Neg	Neg	1+	Neg	100	1.001	7.0	Neg	Neg	Neg	Neg	Neg	100
	M38	1.010	6.0	Neg	Trace	Neg	1+	Neg	100	1.004	6.5	Neg	Neg	Neg	Neg	Neg	100

N.D. No Data * Pkt = Phenylketones

FIG 1

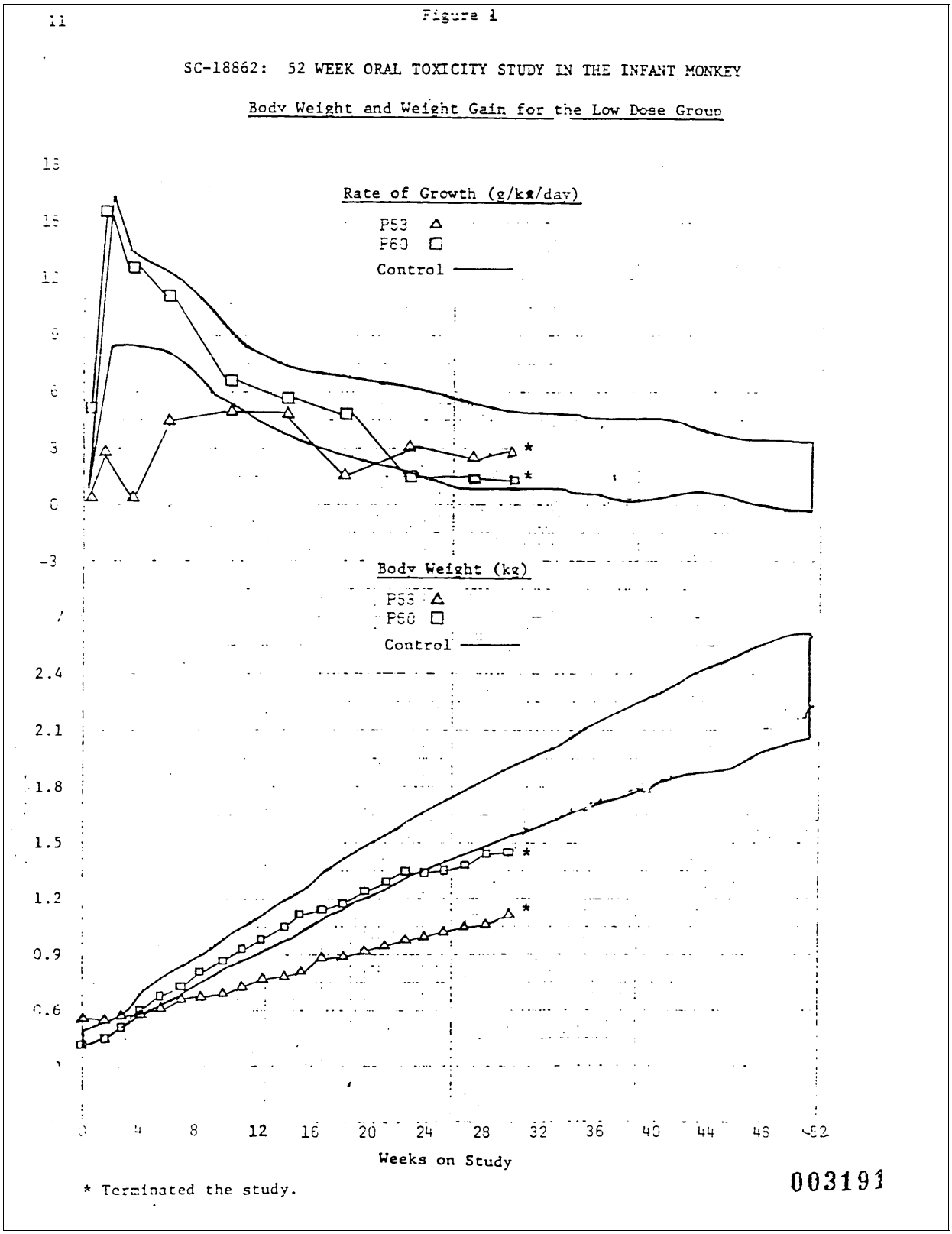


Figure 1
SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY
Body Weight and Weight Gain for the Low Dose Group
Rate of Growth (g/kg/day)
P53
P60
Control
Body Weight (kg)
P53
P60
Control
2.4
2.1
1.8
1.5
1.2
0.9
0.6
0
4
8
12
16
20
24
28
32
36
40
44
48
52
Weeks on Study
* Terminated the study.
003191

Fig 2

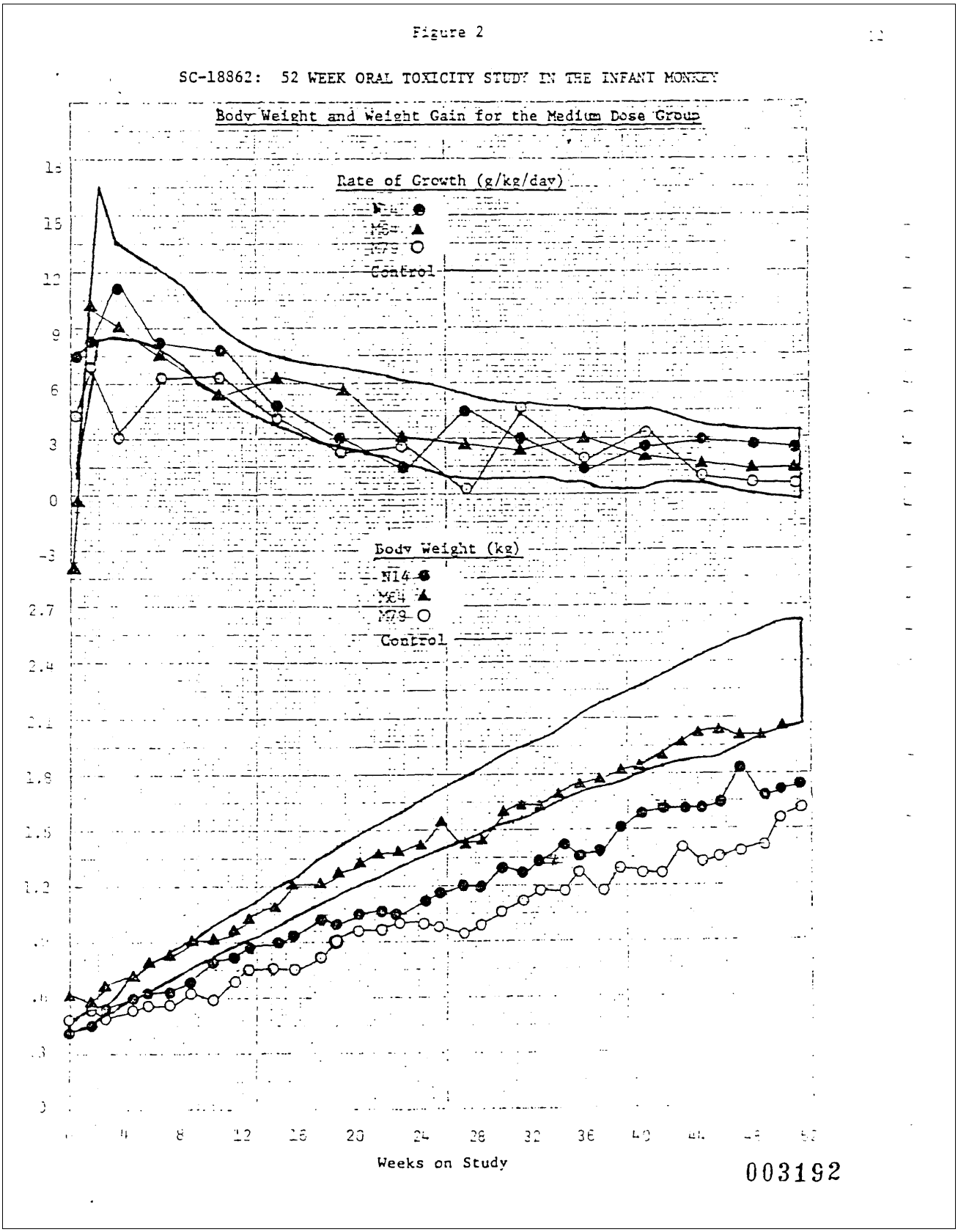


Figure 2
SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY
Body Weight and Weight Gain for the Medium Dose Group
Rate of Growth (g/kg/day)
Control
Body Weight (kg)
N14
M24
M29
Control
Weeks on Study
003192

Fig 3

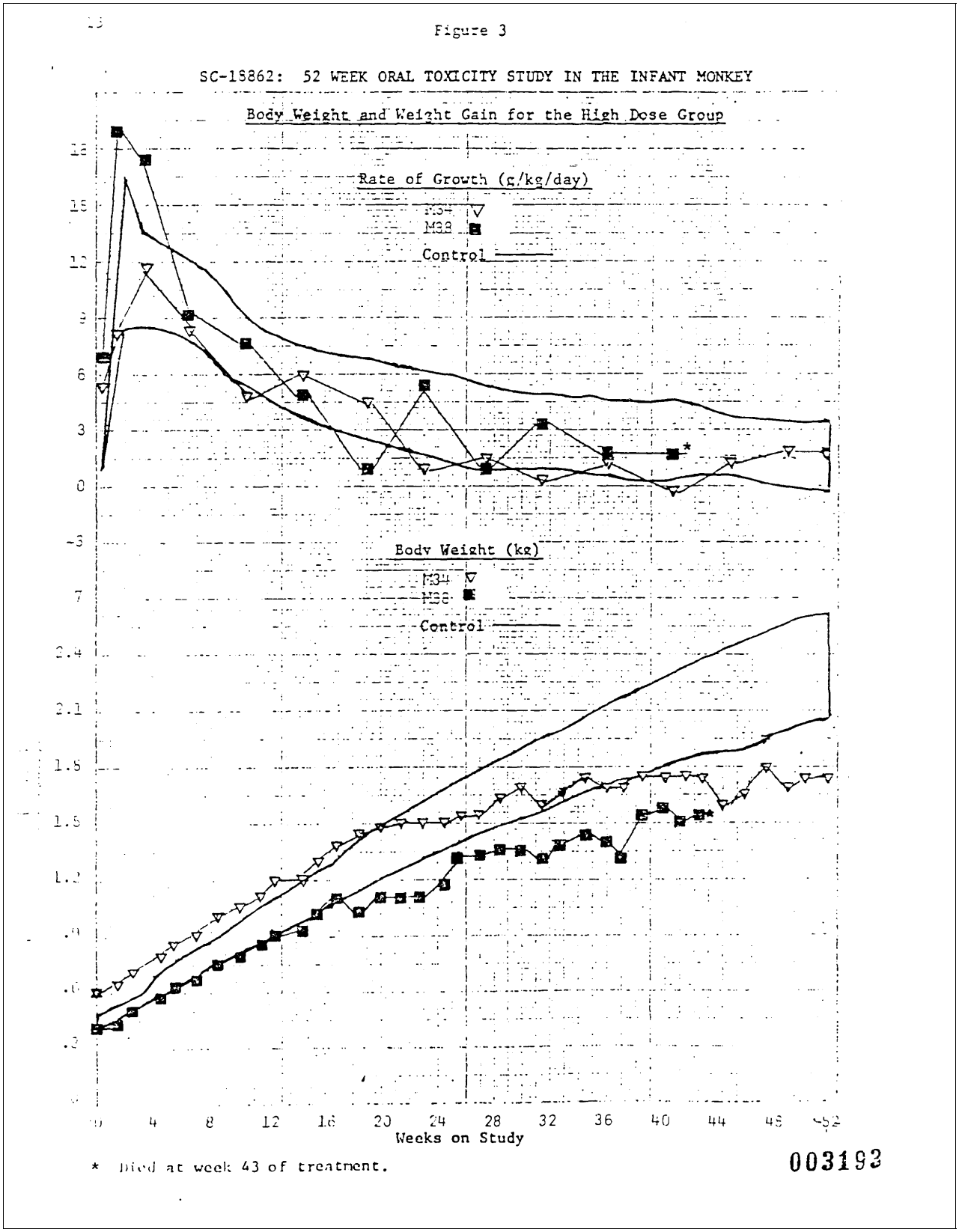
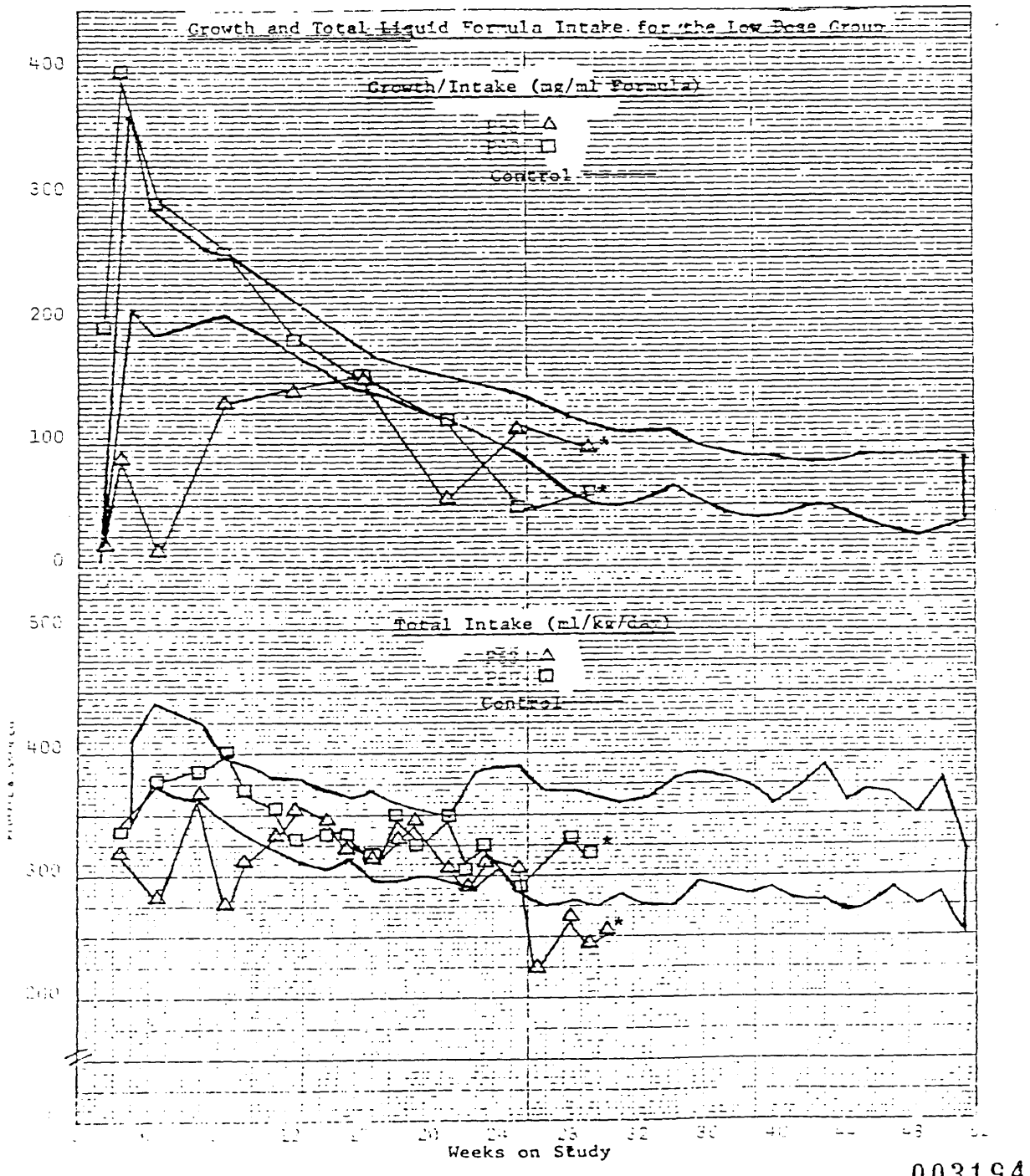


Figure 3
SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY
Body Weight and Weight Gain for the High Dose Group
Rate of Growth (g/kg/day)
M34
M38
Control
Body Weight (kg)
M34
M38
Control
0
4
8
12
16
20
24
28
32
36
40
44
48
52
Weeks on Study
* Died at week 43 of treatment.
003193

Fig 4

Figure 4

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY



003194

* Terminated the study.

Fig 5

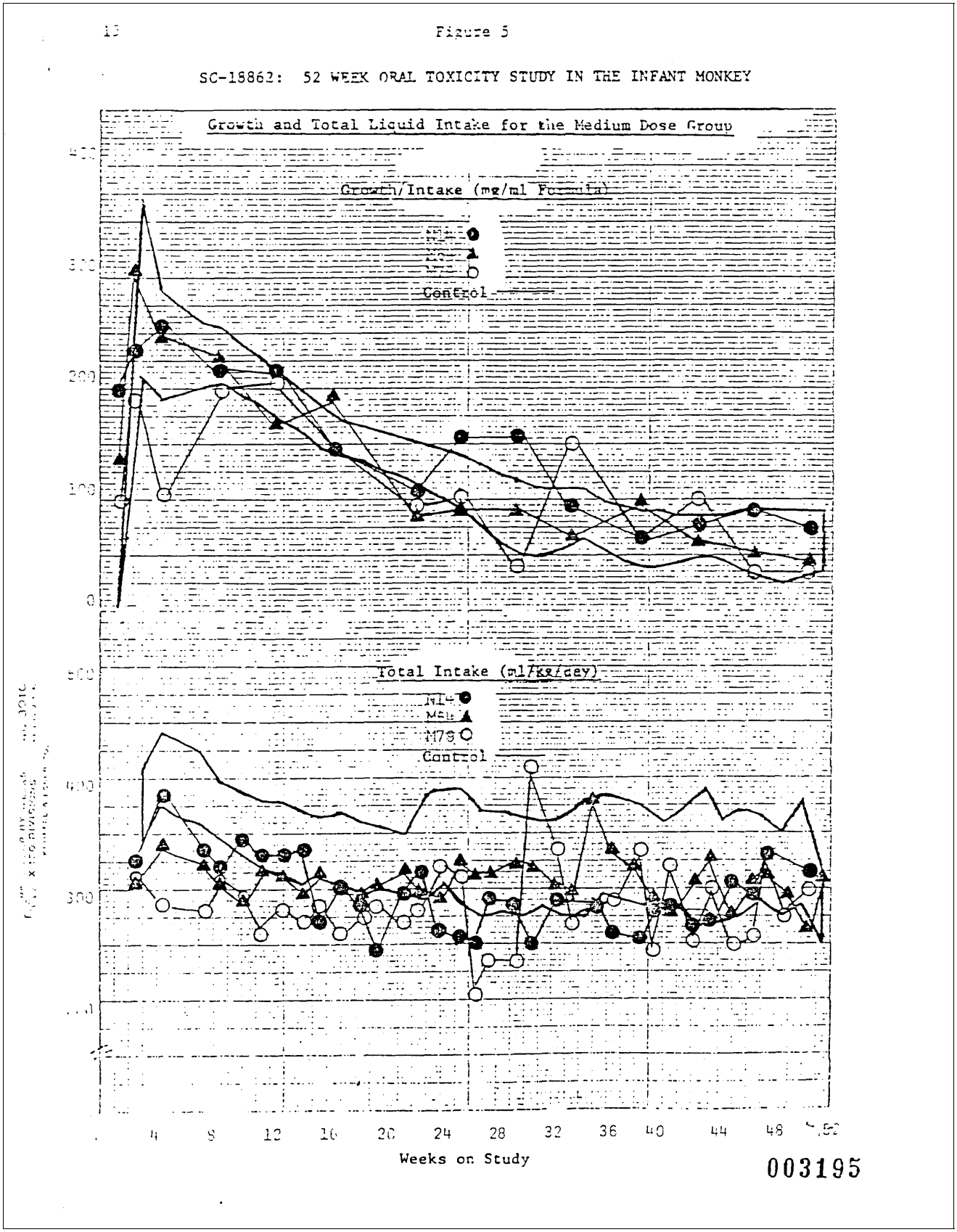


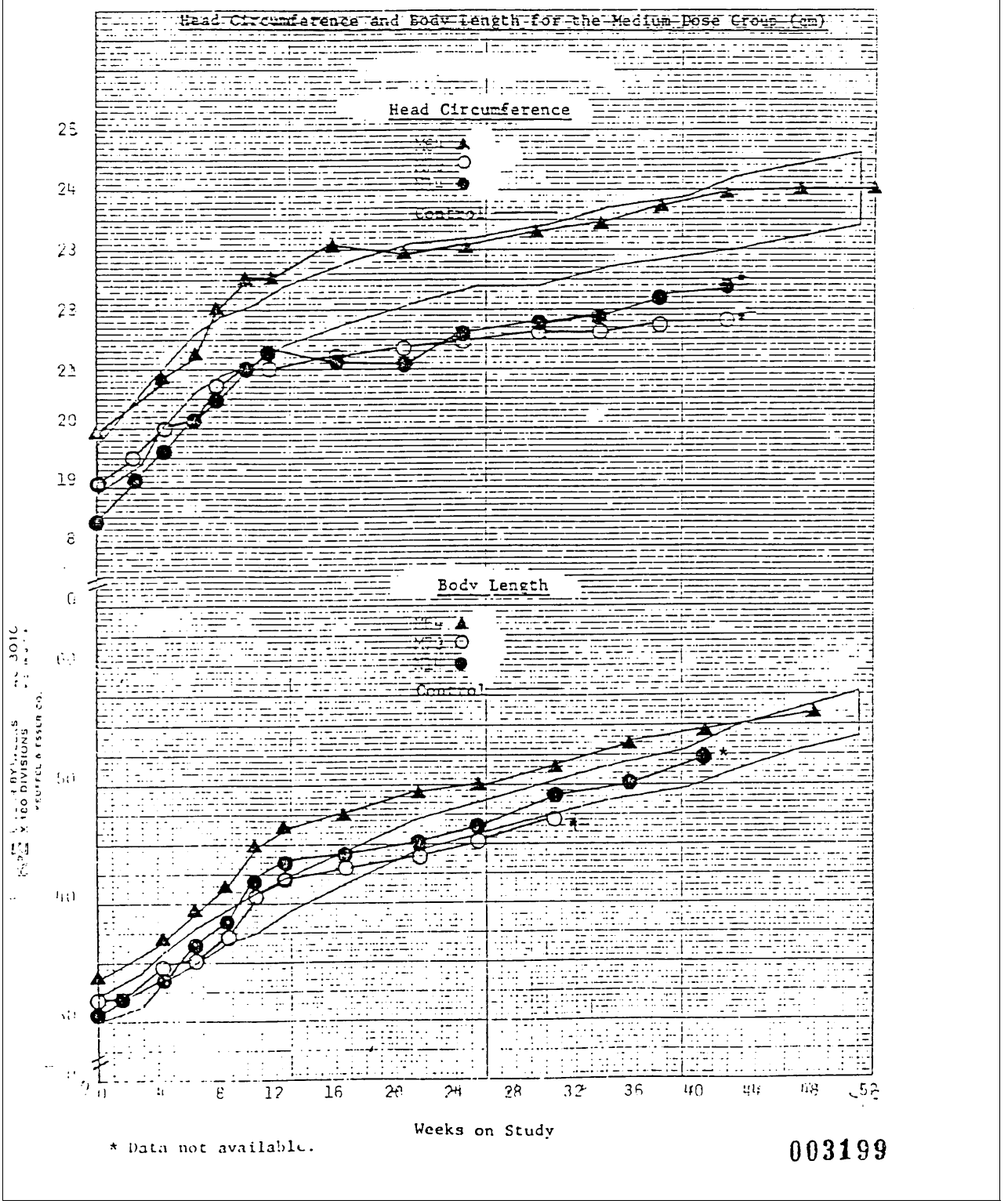
Figure 5
SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY
Growth and Total Liquid Intake for the Medium Dose Group
Growth/Intake (mg/ml Formula)
Control
Total Intake (ml/kg/day)
Control
4
8
12
16
20
24
28
32
36
40
44
48
52
Weeks on Study
003195

Fig 6

19

Figure 8

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY



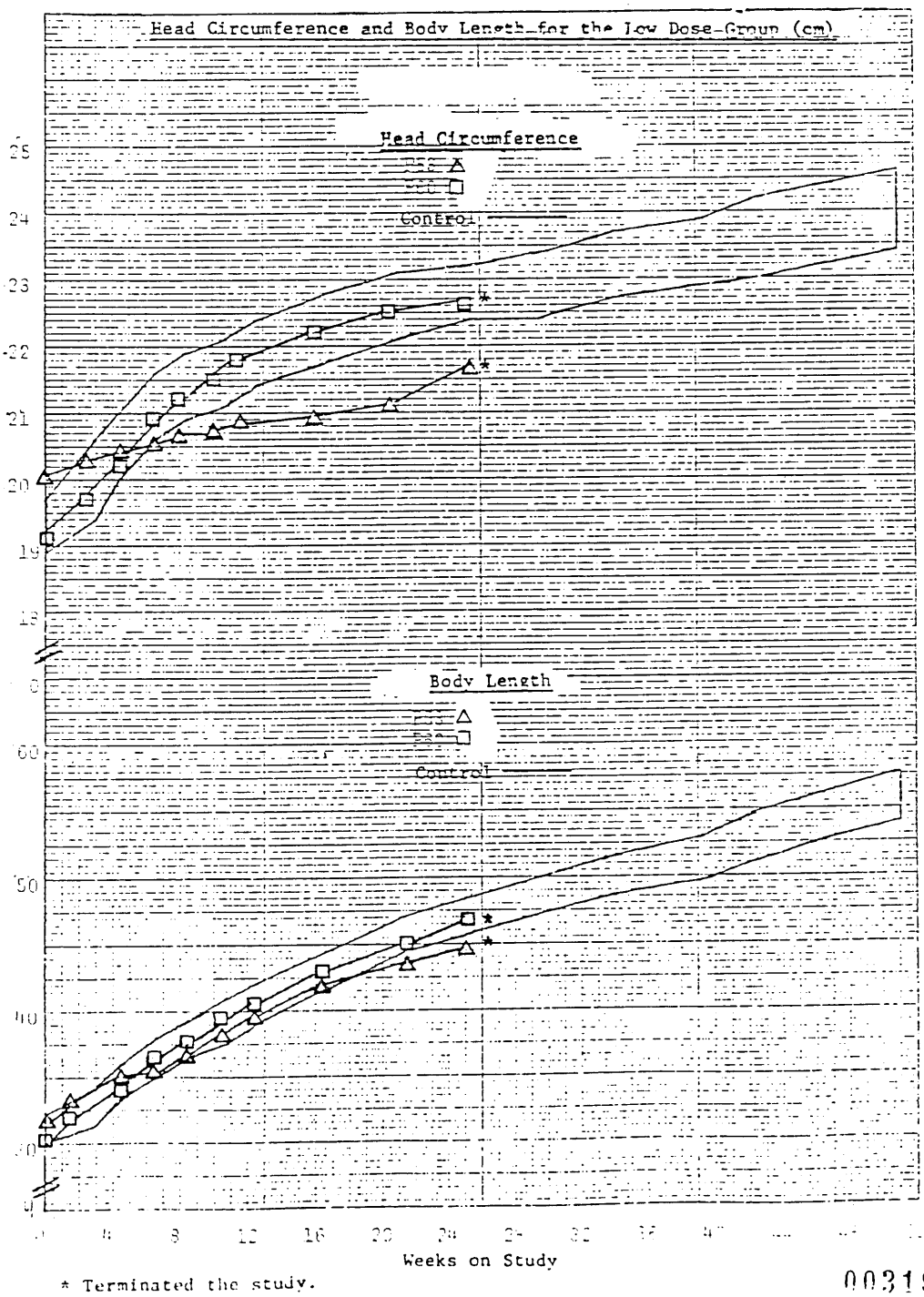
* Data not available.

003199

Fig 7

Figure 7

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

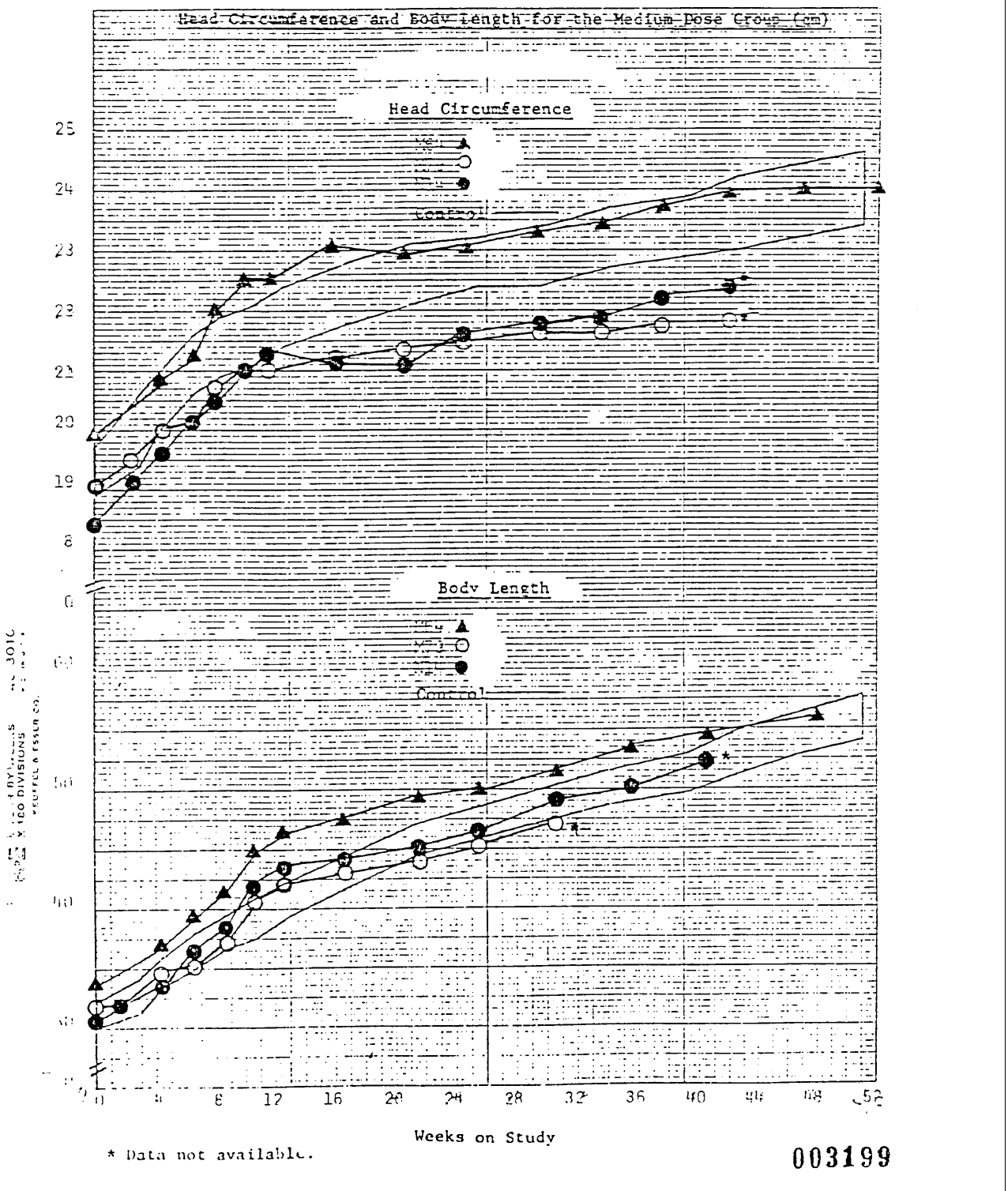


003198

Fig 8

19 Figure 8

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY



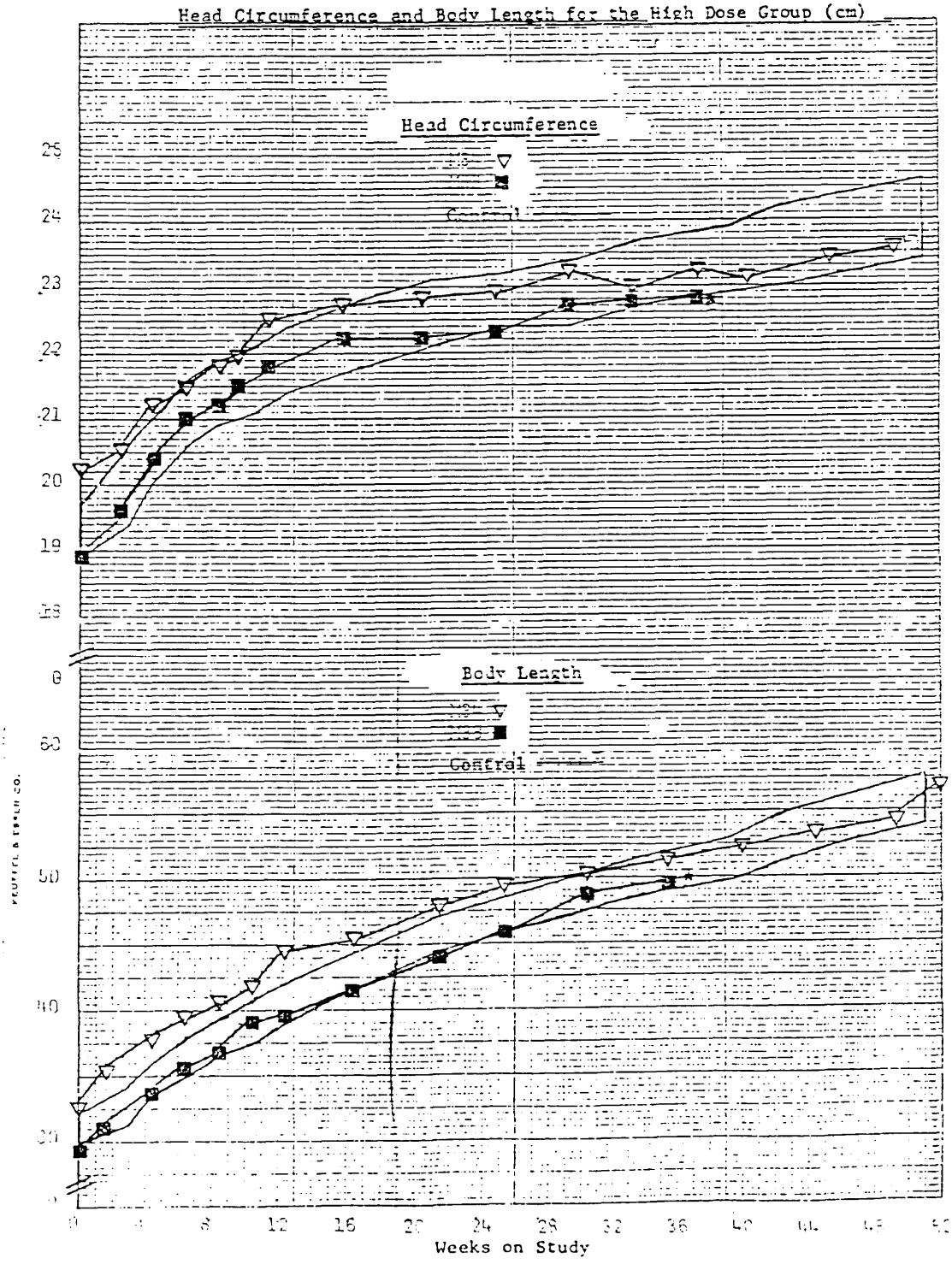
003199

Fig 9

Figure 9

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Head Circumference and Body Length for the High Dose Group (cm)



* Died at week 43 of treatment.

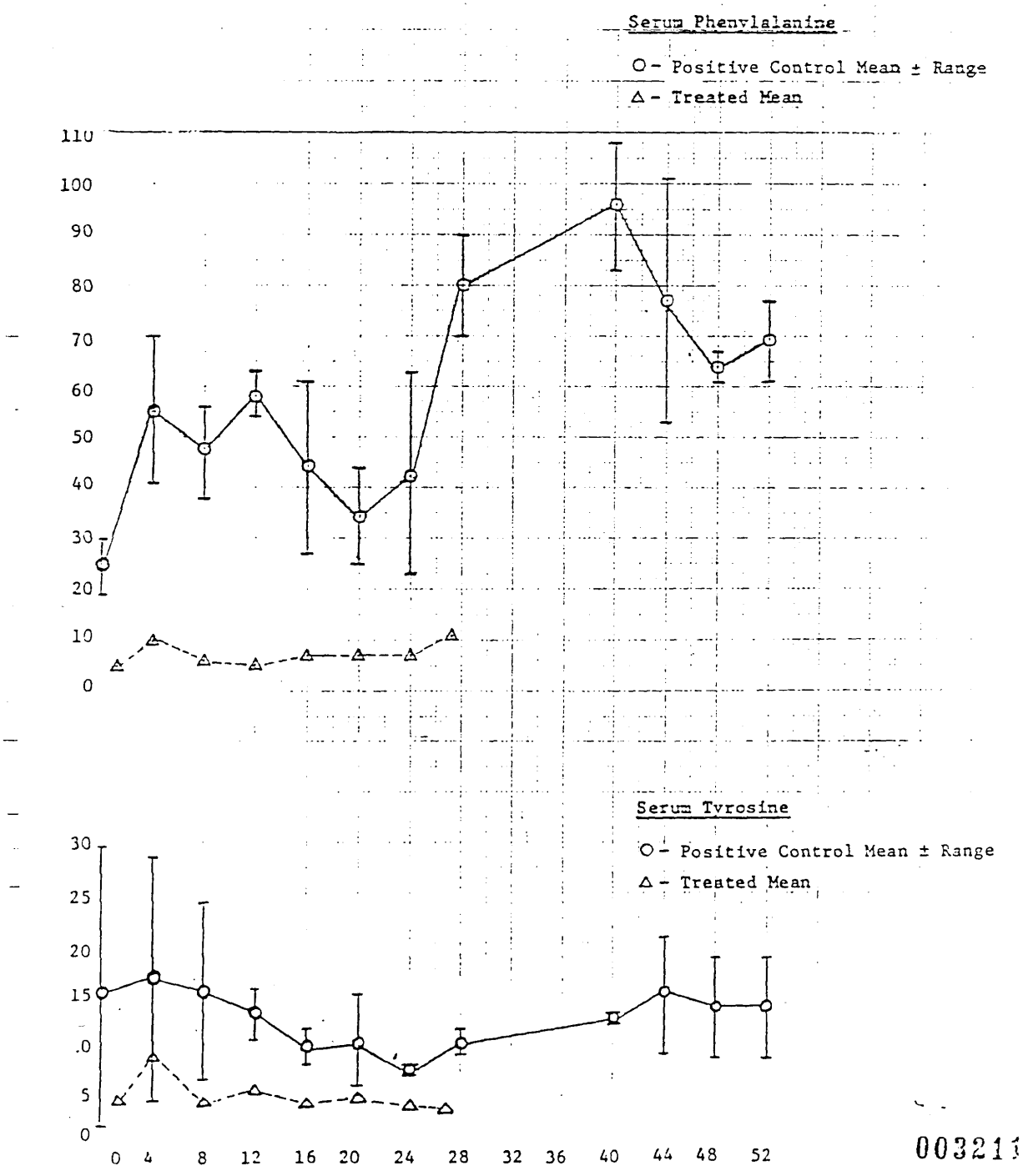
003200

Fig 10

Figure 10

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Serum Phenylalanine and Tyrosine Values for the Low Dose Group (mg%)



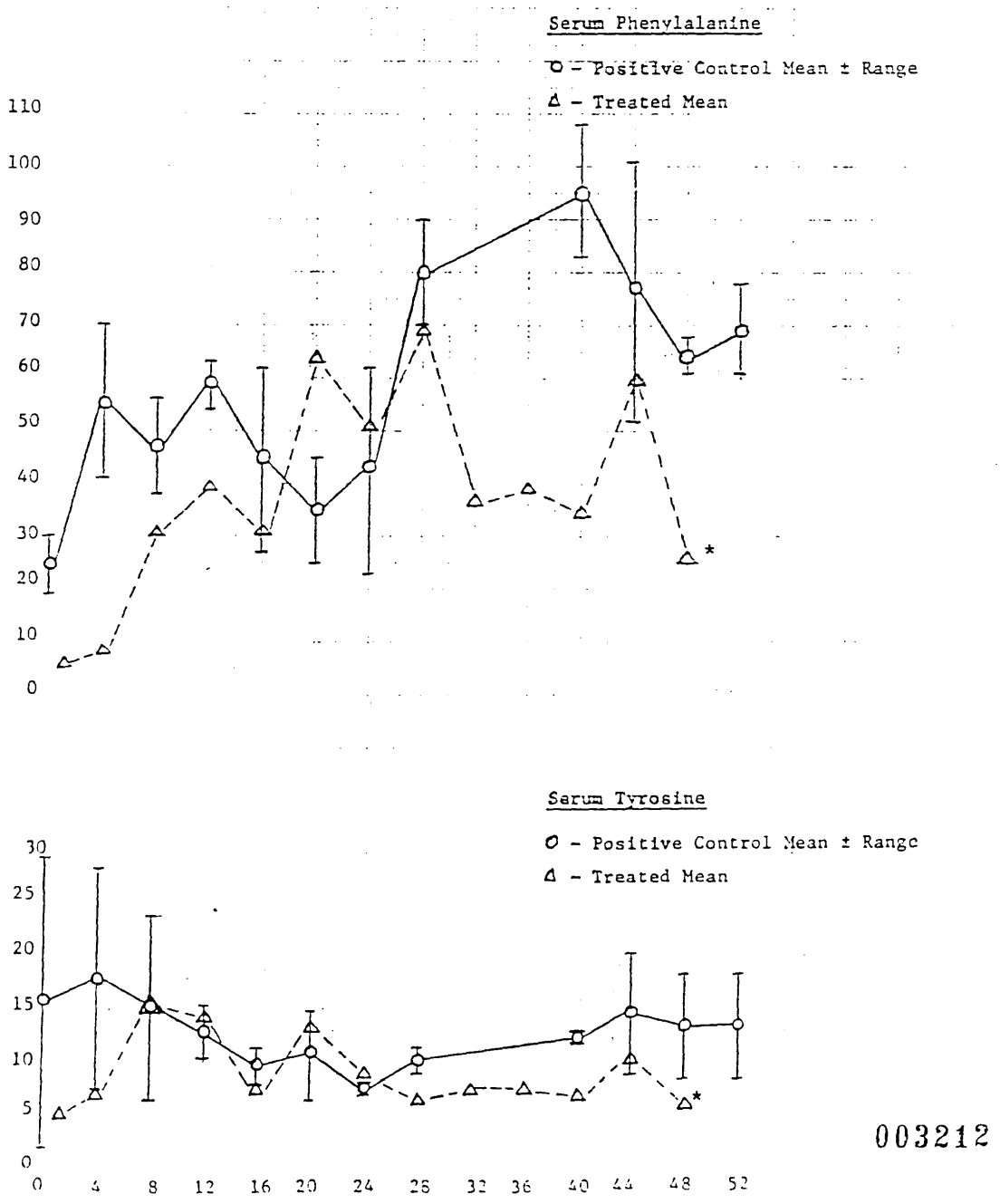
003211

Fig 11

Figure 11

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Serum Phenylalanine and Tyrosine Values for the Medium Dose Group (mg%)



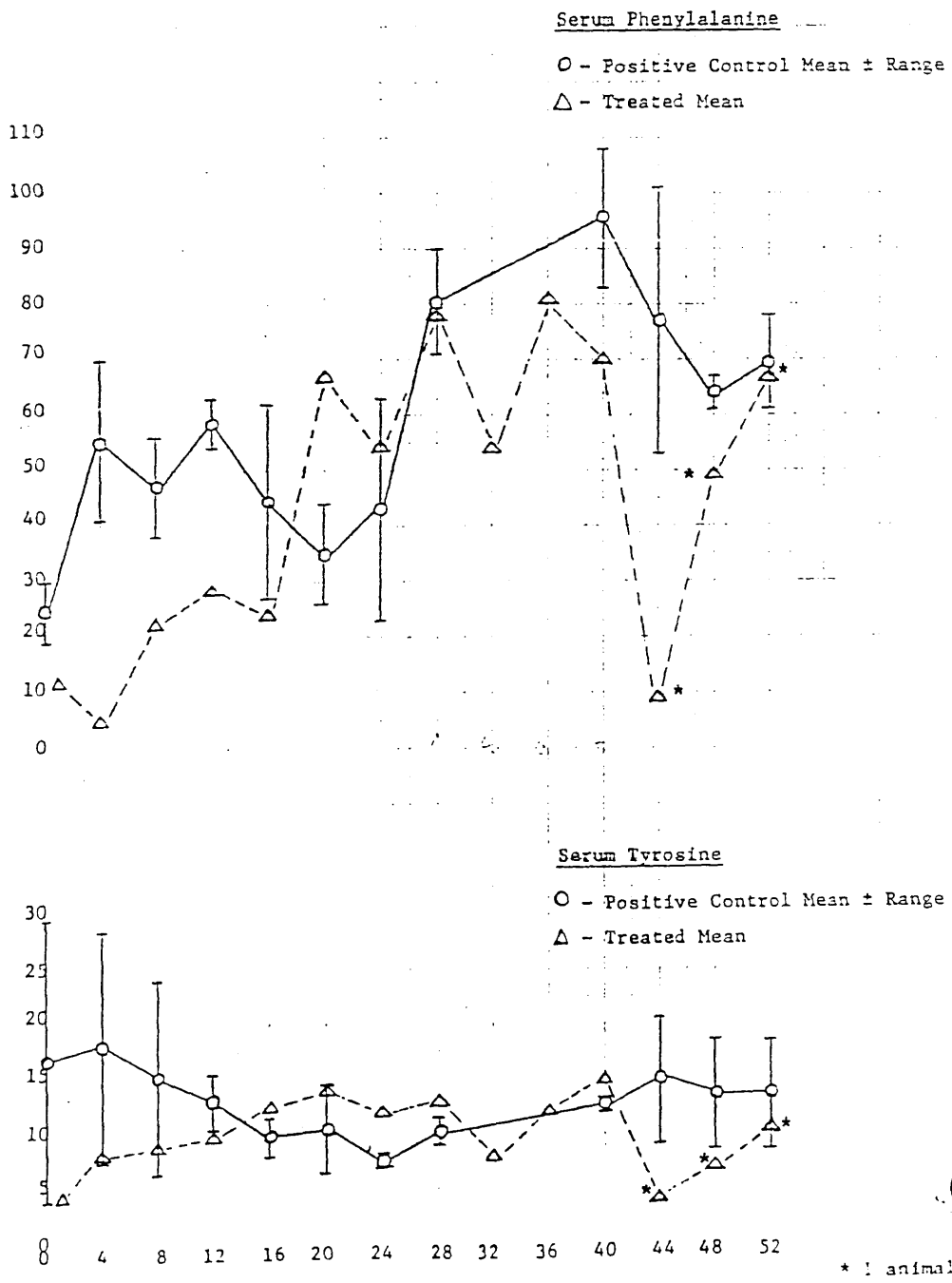
003212

Fig 12

Figure 12

SC-18862: 52 WEEK ORAL TOXICITY STUDY IN THE INFANT MONKEY

Serum Phenylalanine and Tyrosine Values for the High Dose Group (mg%)



003213