

A THREE GENERATION STUDY OF EFFECTS OF INGESTION
OF THE PESTICIDES ABATE AND SENCOR
ON JAPANESE QUAIL

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by

Norman Craig English

Regina, Saskatchewan

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ABSTRACT

The study was initiated to evaluate whether Japanese quail could adapt to continuous ingestion of several concentrations of two pesticides; in both the short term (one generation) and the long term (three generations). Three generations of Japanese quail were used, each generation being 14 weeks. The two pesticides used in the study were Abate and Sencor. In all cases three concentrations of pesticide were fed to each generation of Japanese quail. In the short term study, comparisons were made between each level of pesticide fed and between the control. In the long term study, comparisons were made between each generation at each particular level of pesticide fed. In each case four parameters were looked at: body weight gain, egg production, organ weight and body weight.

Analysis of data showed that Japanese quail could tolerate high levels of Abate and Sencor for three generations. Chicks and adult birds did not differ significantly in tolerance.

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I. INTRODUCTION

Japanese quail (Coturnix Coturnix Japonica) are found all over the old world. They were originally imported into North America from Sicily during the period 1877 - 1882. However, due to the harsh conditions, they had died out by 1885 (Wetherbee, 1961). Japanese quail used in present day research are descended from 140 adult breeding birds imported from Japan in 1953 by J.W. Steinbeck of Concord, California (Wetherbee, 1961 and Padgett and Ivey, 1959).

By the late nineteen fifties and early sixties Japanese quail were gaining prominence as a valuable laboratory bird. A number of researchers including, Padgett and Ivey 1959, Wilson et al 1961, and Shellenberger 1965 and 1966 reported on the scientific potential of this avian species. About the same time other research workers in the field were actively exploring the biological parameters of this bird (Wetherbee, 1961, Abbott et al 1960, Howes 1964, Wilson et al 1961, Padgett and Ivey 1959, Vohra et al 1970, Yo and Gilbreath 1971 and Smith et al 1969). They ascertained that quail had a clutch size ranging from 6 to 12 eggs. Under optimum incubating conditions (99.5°F and 89% relative humidity) a 16 - 18 day incubation period was required. Average hatchability ranges between 60 to 70% of fertile eggs set, although for a period following the birds

seventh week of age it can approximate 90% (Shellenberger 1965, Padgett 1959 et al and Walker 1969 et al). According to Howes 1964, two frequent causes of low hatchability are microscopic cracks in the egg shells leading to dehydration and death of the embryo, and storing of eggs for periods of greater than 14 days prior to setting. Wilson et al 1961 cited the storage of eggs for more than 3 weeks before setting as a common cause of poor hatchability. He stated that hatchability drops at a fairly constant rate of 3% for each day of egg storage prior to setting.

Although, Wetherbee 1961, reported that females have produced eggs when only four and one half weeks old, peak egg production (0.8 eggs per hen per day) is not reached until the 13th week, (Wilson et al 1961 and Daniels 1968). This rate can be maintained until the 26th week of age and then production begins to decline (Woodard et al and Abplanalp 1971).

Egg fertility is affected by the ratio of males to females. The optimum ratio according to Woodard and Abplanalp 1967, was about one male to three or fewer females. Vogt 1971 reported that a 1:1 ratio gave him the best results. Vogt 1971 as well as Woodard and Abplanalp 1967, noted that both fertility and hatchability were optimum when eggs were produced by birds 10 to 12 weeks old.

A number of studies indicate that when purified diets are fed to quail their egg production and hatchability decline, but when turkey commercial breeder diets containing about 23% protein are fed, hatchability and fertility are normal (Smith et al 1969, Latshow et al, 1970 and Gough et al 1968).

Thus using Japanese quail the work was initiated with two main questions in mind: could Japanese quail adapt to moderate and high levels of continuous Abate and Sencor ingestion over a period of three generations, and could they tolerate a continuous pesticide intake at a level that might occur environmentally following a pesticide application.

Literature Cited

Organophosphates

Different organophosphate insecticides are usually metabolized by different degradative pathways. The same insecticide can be metabolized differently by each species of bird it is tested on. It was found by Tucker et al, 1971, that of six species of birds tested for sensitivity against sixteen pesticides (nine of them organophosphates) each of the six species was the least susceptible to one or more of the chemicals. Tucker et al 1971 assessed the degree to which the LD₅₀ of a given chemical for one of the six species would predict the LD₅₀ of the same chemical for another species. The results indicate that such extrapolations are not reliable. For example, the LD₅₀ value for Abate with the six bird species ranged from a low of 31.5 mg/kg for pheasant to a high of 84.1 mg/kg body weight for Japanese quail. In general, he found that for a given chemical more than 50% of the time the LD₅₀ of one species of bird differed from the LD₅₀ of another species by more than two fold, and that nearly 5% of the time the difference was more than ten fold.

Differences in tolerance can also occur between young and mature birds. Sherman and Herrick 1972 fed the chemical S4087 (O-p-cyanophenyl-o-ethylphenylphosphonothioate) to two week old cockerels at 200 ppm in the diet and found no adverse effects.

However, when twenty-seven week old adults were fed a diet containing 100 ppm of the chemical there was a 77% mortality rate.

One common trait that emerges from numerous studies with birds is the tolerance of their reproductive system to organophosphates until they are administered at very high rates. Sherman et al 1971, Gough et al 1967, Sherman and Herrick 1966 and Shellenberger 1965 and 1966 all reported that even when fed at levels high enough to suppress egg production and cause some mortality there appeared to be no adverse effect on fertility or hatchability. Even at these high levels, Sherman and Herrick 1966 found no decrease in eggshell thickness. This is in striking contrast to the organochlorine insecticides which cause eggshell thinning even at the low intake rates that occur in the environment (Peakall 1970, Porter and Stanley 1969, Hickey and Anderson 1968, Bitman et al 1969). Effects caused by organophosphates are probably minimal because of the high excretion rate of the chemicals, for instance, Blinn 1968, found that 90% of administered Abate breaks down and is excreted within a 48 hour period.

Triazines

As a class of herbicides Triazines do not seem to be as toxic as either the organochlorine or organophosphate insecticides. Very little avian research has been reported on triazines, with no data available on LD₅₀ or pathways of metabolic breakdown. Most of the available information comes from mammalian research.

Excretion of triazine metabolites in mammals occurs in both the urine and the feces with the major excretory route being via the urine. The majority of the dose is excreted within a 24 - 72 hour period following ingestion, (Bakke et al 1972, Bakke et al 1971, Bakke et al 1967, Larson and Bakke 1971, Angelucci et al 1965, Zins 1965, Zins 1965, Oliver et al 1969).

Leigh et al 1964 found only 1.85% of the dose excreted in the urine as intact atrazine with the remainder of the excreted products being metabolites. This indicates that the herbicide is extensively metabolized before it is excreted. The metabolism takes place by de-alkylation of the amino side chain leaving the triazine ring intact (Bakke et al 1971 and Oliver et al 1969, Bohme and Bar 1967, Larson and Bakke 1971, Bakke et al 1967, Bakke et al 1972).

A number of experiments have been performed using purified triazine herbicides but Bakke et al 1972 stated that direct feeding of pure atrazine to test cattle was not the best technique. He worked on the premise that the S-triazines when sprayed on plants are metabolized by the plants, and it is the metabolites produced by the plants that should be tested. They used both atrazine and the plant metabolite 2-hydroxyatrazine in their studies. When fed to the rat they found that 65.5% of the atrazine was excreted in the urine and 20.3% in the feces 72 hours after ingestion. For 2-hydroxyatrazine, 78% was excreted in the urine and 5.5% in the feces in 72 hours. Thus in comparing atrazine and its plant metabolite

there was a slight shift in excretion routes but the net result after 72 hours was similar.

Triazines seem to be relatively nontoxic. Johnson et al 1972, sprayed atrazine and prometone on pastures at the highest recommended levels and then fed the freshly cut forage to mature cattle and sheep. Average animal intakes of atrazine over the 27 day test period were 30 mg/kg/day for cattle and 40 mg/kg/day for sheep. The prometone intakes were 27 mg/kg/day for cattle and 47 mg/kg/day for sheep. These intake rates did not produce any measurable effects either physiologically or morphologically. Dunachie et al 1967 and Dunachie et al 1970 using an egg injection technique found the triazines to be relatively harmless. Dunachie and Fletcher 1967, injected atrazine and simazine into hens eggs at a concentration of 100 ppm. The hatchability was equal to that of control eggs which were injected with acetone alone. Whereas when 2, 4-D was injected at 100 ppm there was a 30% decrease in hatchability.

2. MATERIALS AND METHODS

Forty Japanese quail (Coturnix coturnix japonica) were obtained from the University of Saskatchewan in the late fall of 1971. These birds were used to establish a colony at the University of Manitoba for toxicological studies.

Incubators

Two-three hundred egg incubators; designed by the University of Manitoba, Animal Science Department, were used for the hatching of the Japanese quail eggs. Styrofoam sheets with holes one inch deep and one inch in diameter were placed on the egg trays. The quail eggs were placed in these holes. This allowed tilting of the trays without damaging the eggs. Trays containing the eggs were automatically tilted once every four hours. The incubators were maintained at $99^{\circ}\text{F} \pm \frac{1}{2}^{\circ}$ and at a relative humidity of 80% until the sixteenth day of incubation. On the sixteenth day of incubation the eggs were removed from the incubating trays and placed in hatching trays in the same incubator and the relative humidity raised to 95%. Hatching was completed by the eighteenth day and chicks were transferred to the brooder.

Chick Rearing

The brooder illustrated in Fig. 2:1 had two removable floors.

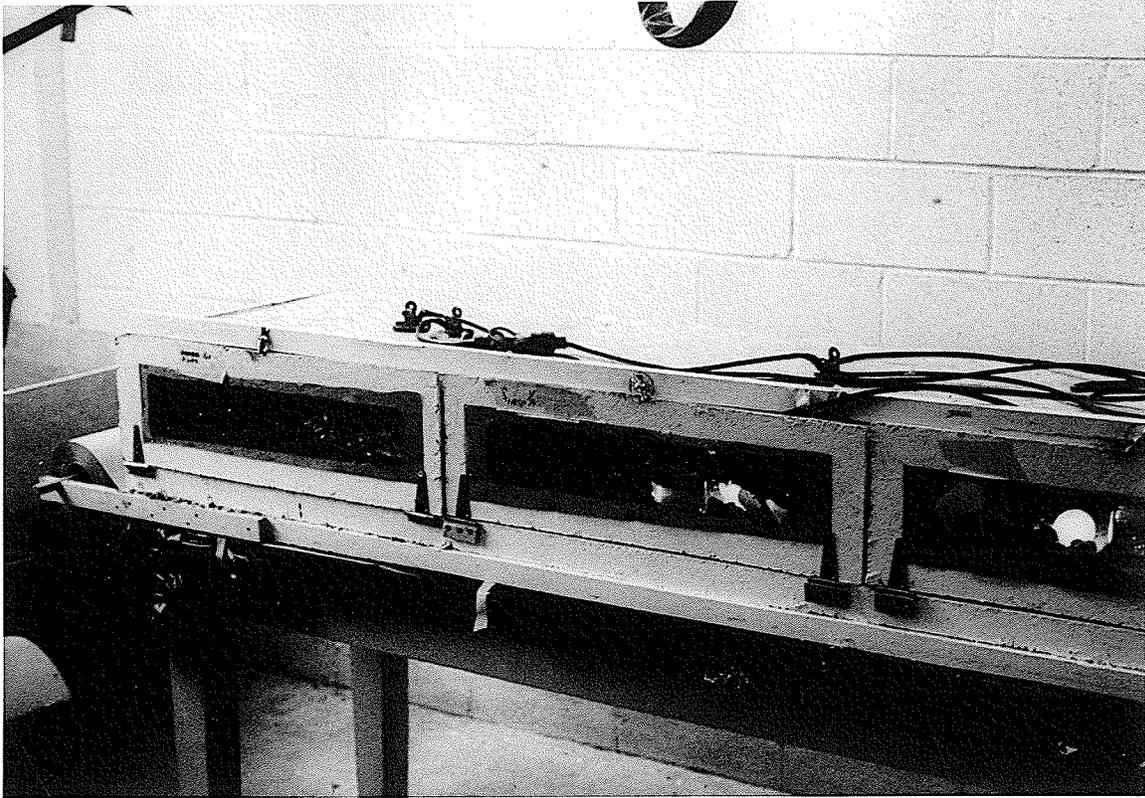


Fig. 2:1--Brooder

One floor being sixteen inch Twilweld wire mesh and used when the quail were two (after eighteenth day of incubation) to seven days of age. The other was eight inch Twilweld wire mesh and used for eight to fourteen day old chicks. Heat for the brooder was supplied by four 25 watt bulbs that could be raised or lowered in relation to the floor of the brooder to keep the temperature between 80° and 90°F.

At two weeks of age the quail were transferred to the top four decks of a five deck cage (Hawkins Million Dollar Hen Inc.) for rearing. Each deck had a separate heating element which was used until the quail were three weeks old. Then they were reared at room temperature. Each deck was equipped with a two foot long feeder, and two automatic waterers operated on a gravity flow system from a tank above the cage.

Lighting

Each deck of the battery was illuminated with identical flourescent lights. These lights were automatically set for a 16:8 hour day-night cycle.

Standard Diet

A 17% protein commercial cage layer all mash crumble and a 40% protein commercial laying supplement were mixed equally to yield a diet containing 28.5% protein and 4.5% calcium. This was the standard diet fed to the quail from week 1 to week 14.

Determination of Acute Oral LD₅₀ and
Environmental Treatment Level

Single oral doses of Abate were given to the quail in gelatin capsules at 7.5, 30, 75, 120, and 150 mg/kg. a.i.¹ Each dose was administered to five adult female birds to determine the acute oral LD₅₀. The percent mortality in each group was then plotted against the log₁₀ of dose (Fig. 2:2) and the LD₅₀ was calculated to be 69.1 mg/kg.

Similarly, the acute oral LD₅₀ for Sencor was determined with 9 dose levels of: 200, 250, 300, 350, 400, 450, 700, 1,000 and 2,000 mg/kg. a.i. Data plotted in Fig. 2:3 gave an acute oral LD₅₀ of 372 mg/kg.

The approximate plant biomass that would be accumulated by foraging Japanese quail over a summer was estimated from work done by Golly 1960, and Odum 1959. The recommended rates of spray for Abate and Sencor were then calculated and compared to the average amount of plant matter available on a particular day to administer a pesticide ration for that day. By this process the daily environmental intake for the birds, for one day, was calculated to be .025 of the acute oral LD₅₀ for both Abate and Sencor. Two other dose rates were then established; one at which a pesticide effect might be expected (0.25 of the LD₅₀ dose per day) and an intermediate dose (0.10 of the LD₅₀ dose per day).

¹ Active ingredient

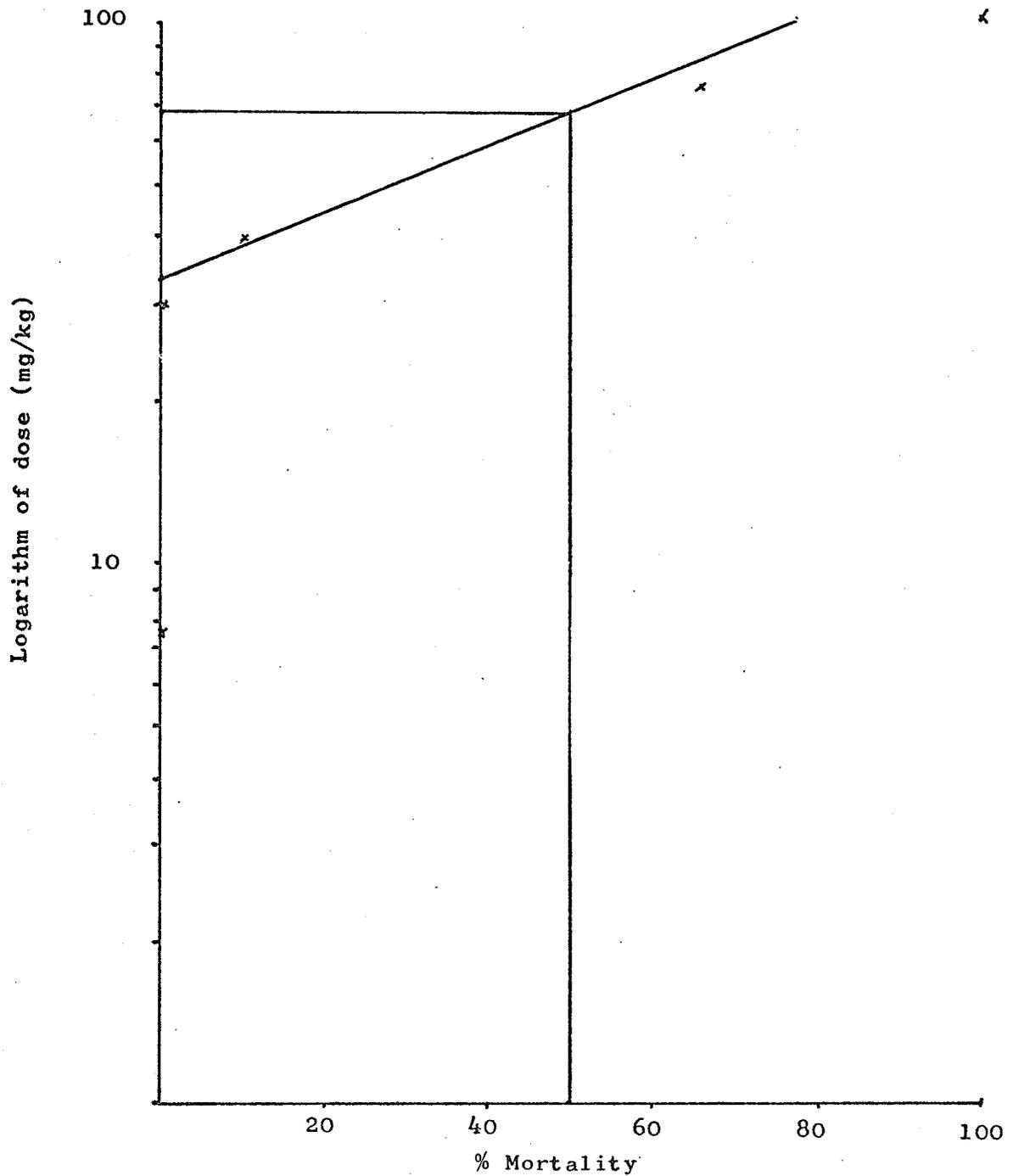


Fig. 2:2.--Acute oral LD₅₀ for Japanese quail fed Abate

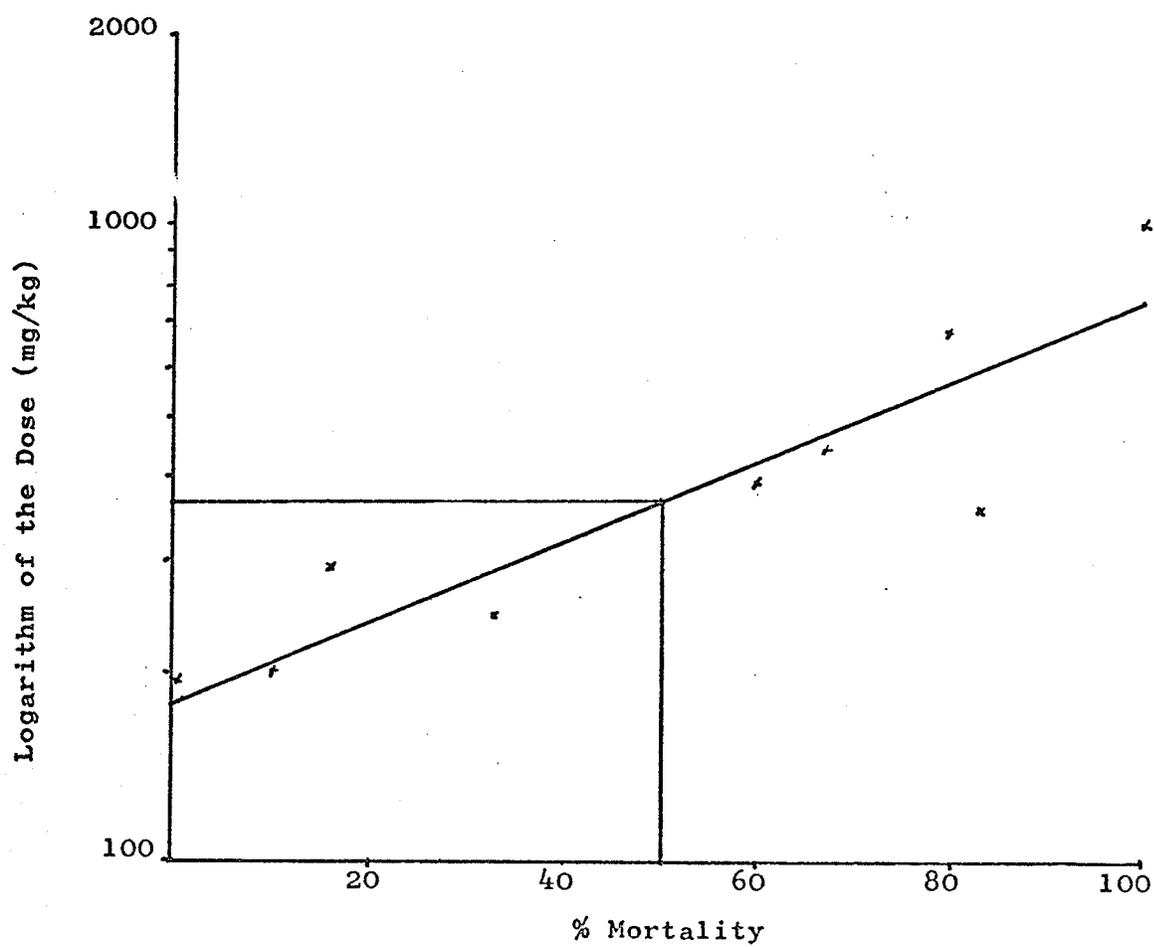


Fig. 2:3.--Acute oral LD₅₀ for Japanese quail fed Sencor

Treatments

Commencing at four weeks of age in generation I Abate (O, O, O', O' - Tetramethyl, O, O'-thiodi-p-phenylene phosphorothioate) and Sencor (4-amino-6-tert-butyl-3-(methylthio)-s-triazin-5(44) one) were mixed with the diet and continuously fed to the quail for the next three consecutive generations. Each generation lasted for 14 weeks.

The insecticide Abate was mixed with the standard diet at a concentration of 8, 32, and 80 ppm. These diet concentrations approximate daily intakes of 0.025, 0.1 and 0.25 of the acute oral LD₅₀ dose, respectively.

A stock mixture was made by dissolving 3 mls. of Abate 4-E (43% a.i. Cyanamid) in 200 mls. acetone then pouring this over 500 gms. of standard diet. The mixture was blended in a Hobart mixer for four minutes, poured into trays and the acetone allowed to evaporate. The resultant dry cake was reblended and stored as stock mixture for up to one month. This stock mixture was used in preparation of experimental diets.

The stock mixture was diluted with standard diet 1:320 for a diet of 8 ppm, 1:80 for a diet of 32 ppm, and 1:32 for a diet of 80 ppm Abate. These experimental diets were fed within one month of date of preparation.

Sencor, a herbicide, was a powder. A Hobart mixer was used to dilute this powder with the standard diet. Dilutions were 1:160 for a diet of 37.2 ppm, 1:40 for a diet of 149 ppm and 1:16 for a diet of 372 ppm Sencor. These diets approximate daily intakes of 0.025, 0.10 and 0.25 of the acute oral LD₅₀ dose for this chemical,

respectively.

The standard diet and experimental diets were fed to groups of 35 birds (25 female and 10 male) from the day of hatching until the end of the fourteenth week in each generation. During the fourteen week period, weight gain and egg production were recorded as parameters that could indicate a pesticide effect upon the birds. At the end of fourteen weeks the birds were killed by ether inhalation, body weight recorded and then heart, liver, lung, kidney and gonads were removed and weighed. Any significant change in organ or body weight was interpreted as a pesticide effect. This was repeated for three generations. Eggs for each subsequent generation were collected when the female birds were 9 - 10½ weeks of age and stored for a period of up to ten days at 55°F. On the tenth day the eggs were placed in incubators and these eggs produced the next generation of quail.

Statistical Analysis

Two methods were used for statistical analysis. Weight gain and egg production curves were analyzed by a Wilcoxon's two sample test for unpaired observations as described by Steel and Torrie, 1960. The test for significance was conducted by comparing the lowest and highest curve of any figure. If significance did occur then tests were run with other curves. Thus in some (but not all) cases, although there are six possible comparisons between the control

and the three Abate or Sencor treatments only one comparison and one Wilcoxon value were calculated at the 5% level of significance (Steel and Torrie 1960).

When comparing between generations three comparisons are possible between the three generations. As was stated earlier only one value is presented if no significance was found when the two extreme curves (highest and lowest) were compared.

In the case of body and organ weights a F test was first performed, followed by a Duncan's test (Steel and Torrie 1960).

3. RESULTS

The results are presented separately for the two pesticides, Abate and Sencor. Each pesticide section consists of a comparison of weight gain, egg production, body weight, and organ weight, between treatments within a generation, followed by a comparison of weight gain, egg production, body weight, and organ weight, between generations within a treatment.

A. Abate

I. General Comments

Japanese quail displayed organophosphate poisoning only on the 80 ppm treatment. On this treatment the birds developed tremors, listlessness and loss of balance. Breeding behavior was greatly reduced or almost stopped. Egg fertility dropped to near zero by the second generation making it impossible to produce a third generation.

II. (a) Body weight gain between Abate Treatments within the Three Generations of Japanese Quail

In generation I, significant differences were found in body weight gain for the birds between the control and 80 ppm, between 8 and 80 ppm and between 32 and 80 ppm treatments, with the birds fed the lower Abate dose in each case having the greater weight gain (Fig. 3:1, Table 3:1). For generation II, no significant differences

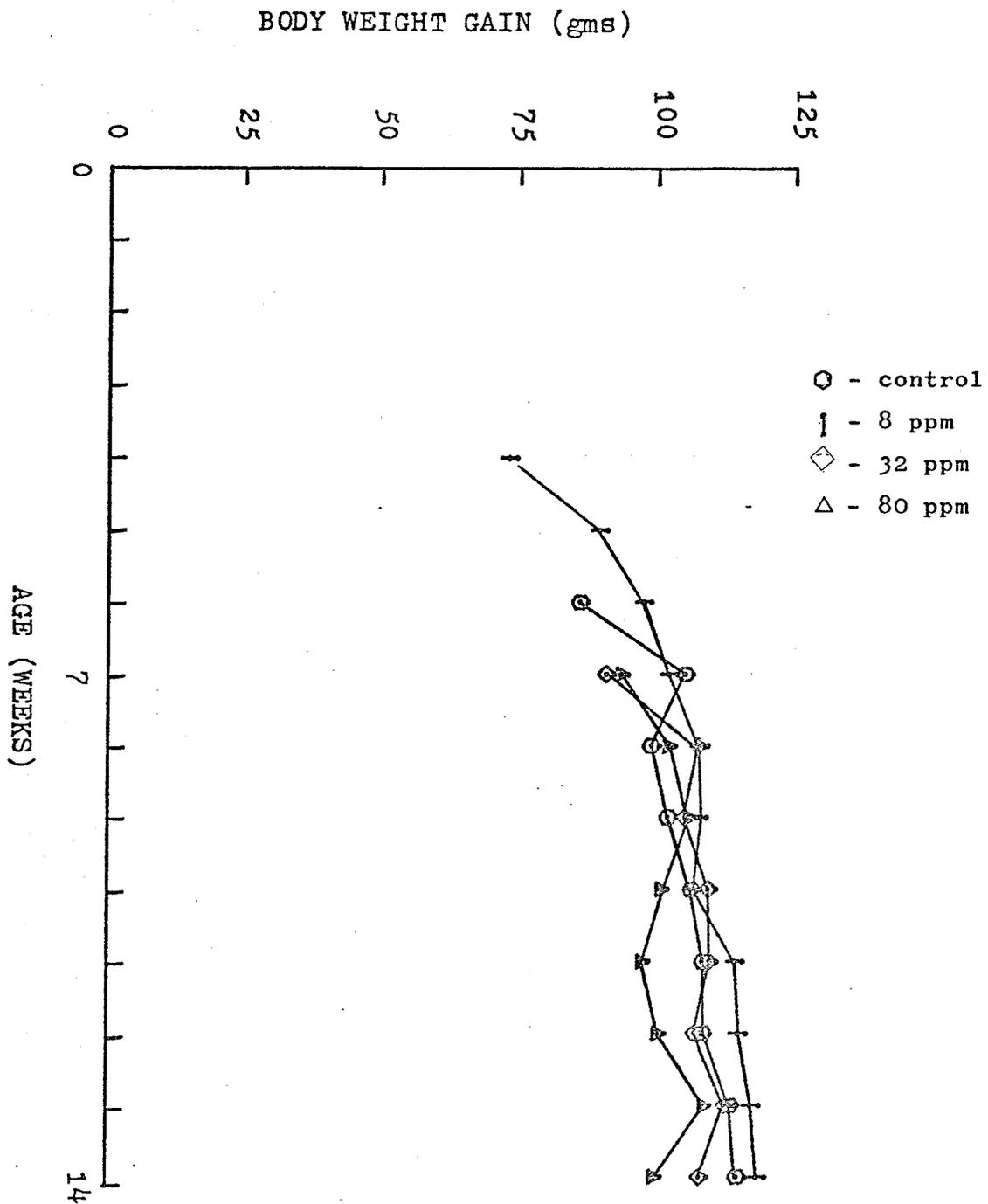


Fig. 3:1.--Fourteen weeks body weight gain of Japanese quail fed four Abate treatments in generation I

TABLE 3:1

WILCOXON TWO SAMPLE TEST VALUES FOR BODY WEIGHT GAIN
BETWEEN ABATE TREATMENTS WITHIN GENERATIONS

Gener- ation	Treatments ^{1,2}					
	0 vs 8	0 vs 32	0 vs 80	8 vs 32	8 vs 80	32 vs 80
I			48*(80)		41*(80)	49*(80)
II	132(8)					
III		104(0)				

¹Wilcoxon values with an asterisk indicate the 2 treatments being compared are significantly different at the 5% level.

²Figures in brackets represent the treatment (ppm) in the comparison with the lowest mean weight. If no significance occurred only one value per generation is presented and this value is the closest value to significance that occurred in that generation.

in weight gain were found (Fig. 3:2, Table 3:1). Similarly in generation III, no significant differences in body weight gain were found between the different treatments (Fig. 3:3, Table 3:1) during the fourteen week experimental period.

(b) Comparison of body weight gain between the three generations of Japanese quail within treatments of Abate

There was no difference between the growth rate of the controls in the three generations (Fig. 3:4, Table 3:2). Of the three Abate treatments, only the quail on the 32 and 80 ppm treatments showed a significant difference in weight gain between generations. On the 80 ppm treatment a significant difference occurred between generation I and II (Fig. 3:7, Table 3:2), and on the 32 ppm treatment a significant difference occurred between generation I and III (Fig. 3:6, Table 3:2), with generation I having the smaller weight gain in each case.

III. (a) Egg production between Abate treatments within the three generations of Japanese Quail

Egg production of the Japanese quail fed the three Abate treatments 8, 32, and 80 ppm was measured for three generations. Fig. 3:8 (generation I), Fig. 3:9 (generation II), and Fig. 3:10 (generation III) provide the data for a Wilcoxon test of significance on egg production which is found in Table 3:3. Egg production for

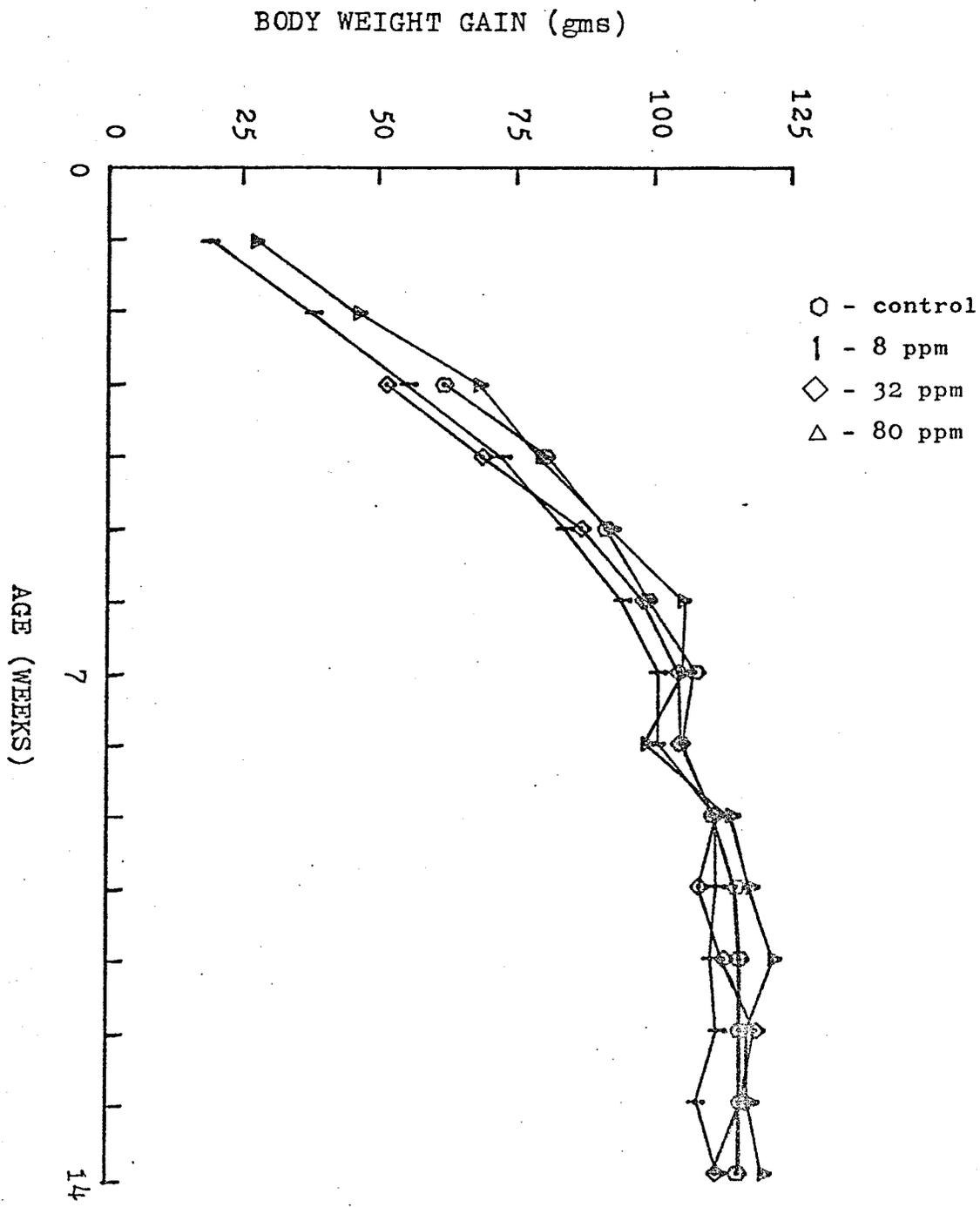


Fig. 3:2.--Fourteen weeks body weight gain of Japanese quail fed four Abate treatments in generation II

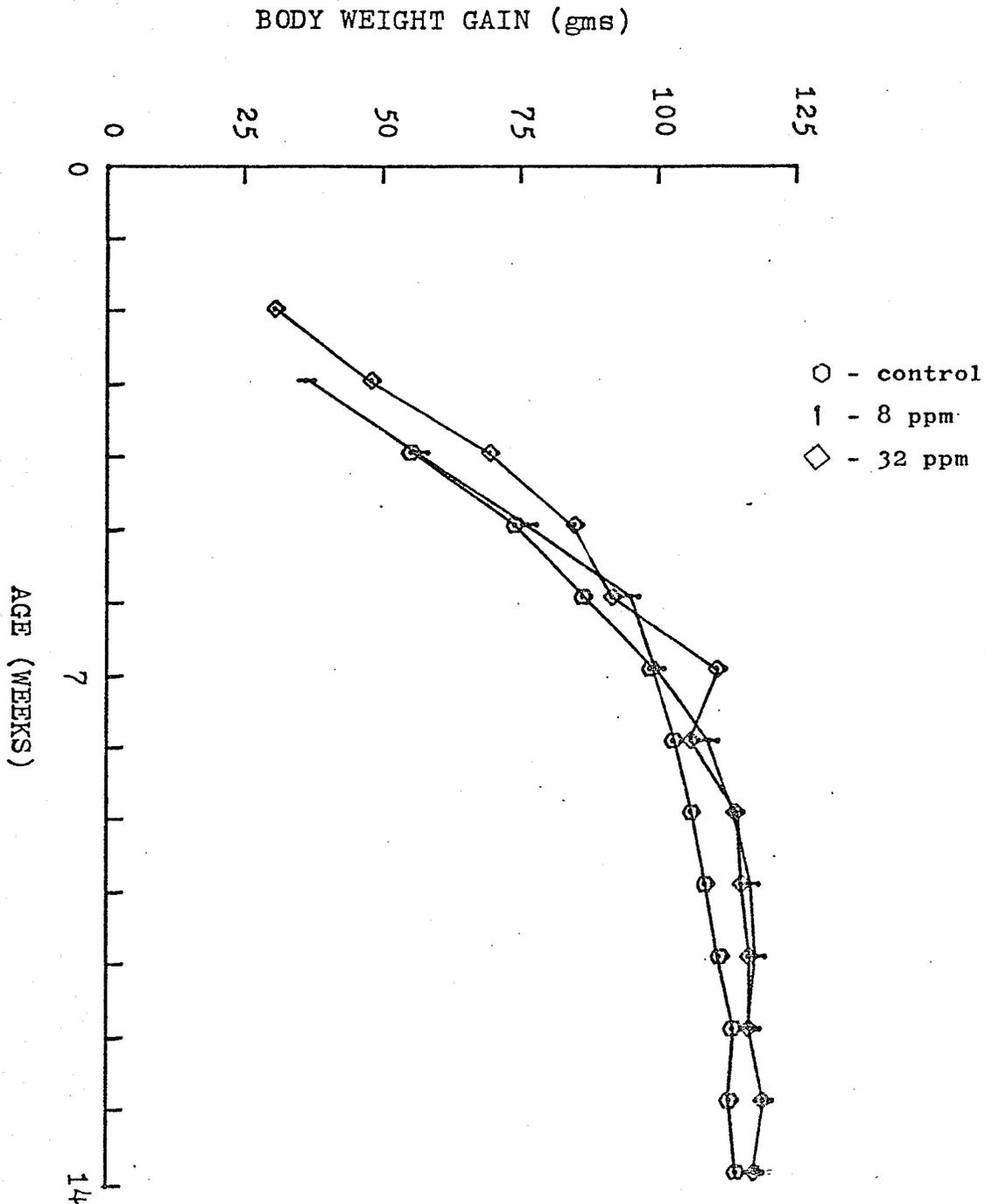


Fig. 3:3.--Fourteen weeks body weight gain of Japanese quail fed three Abate treatments in generation III

TABLE 3:2

WILCOXON TWO SAMPLE TEST VALUES FOR BODY WEIGHT GAIN
 BETWEEN GENERATIONS WITHIN ABATE TREATMENTS

Treatment (ppm)	Generations ^{1,2}		
	I vs II	I vs III	II vs III
0	65 (I)		
8	118 (II)		
32	55 (I)	43* (I)	
80	44* (I)		

¹Wilcoxon value with an asterisk indicates the 2 generations being compared are significantly different at the 5% level.

²The generation in brackets behind each Wilcoxon value had the lowest mean in the comparison. If no significance occurred, only one value per treatment is presented and this value is the closest value to significance that occurred in that treatment.

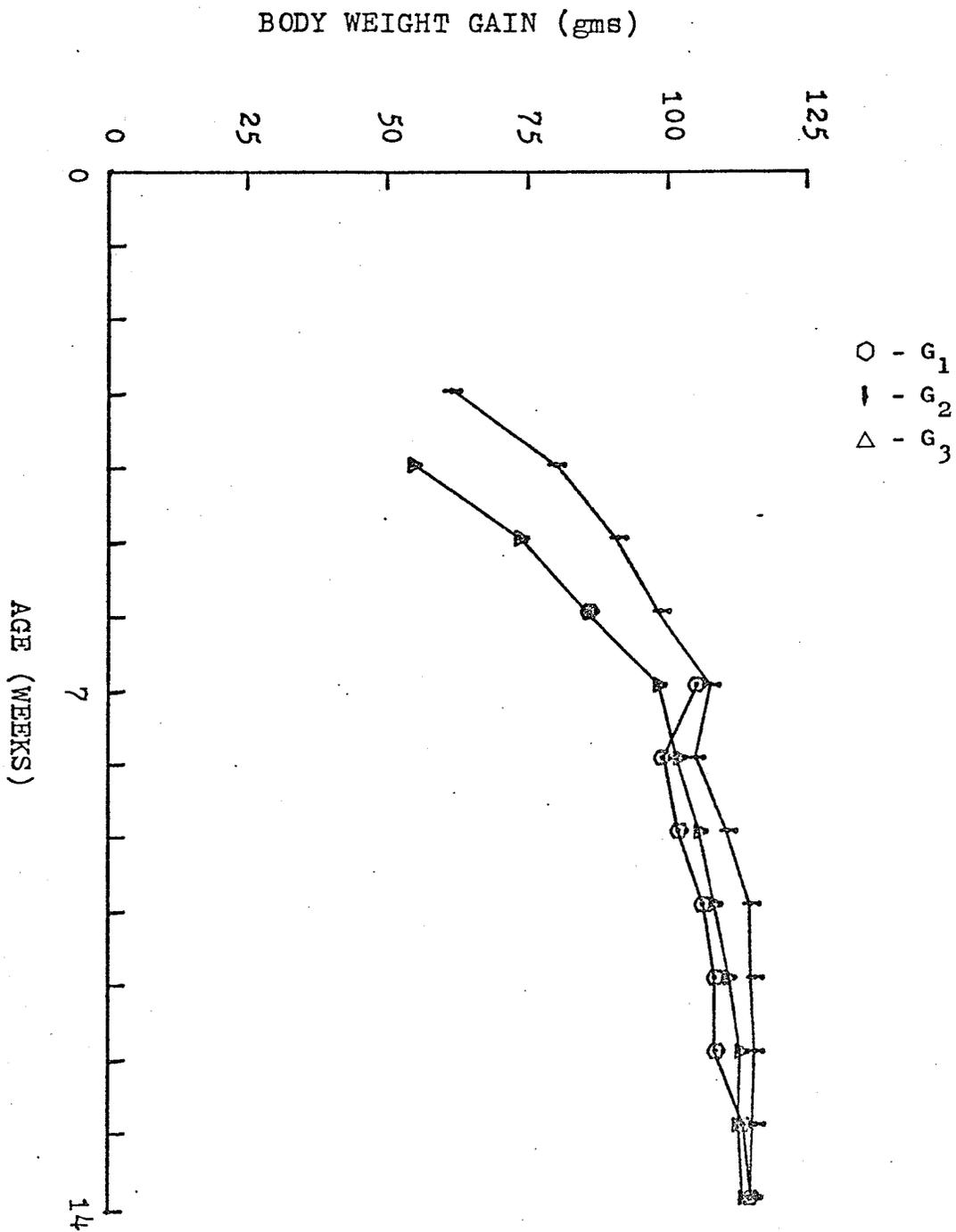


Fig. 3:4.--Fourteen weeks body weight gain for 3 generations of Japanese quail fed Abate at 0 ppm

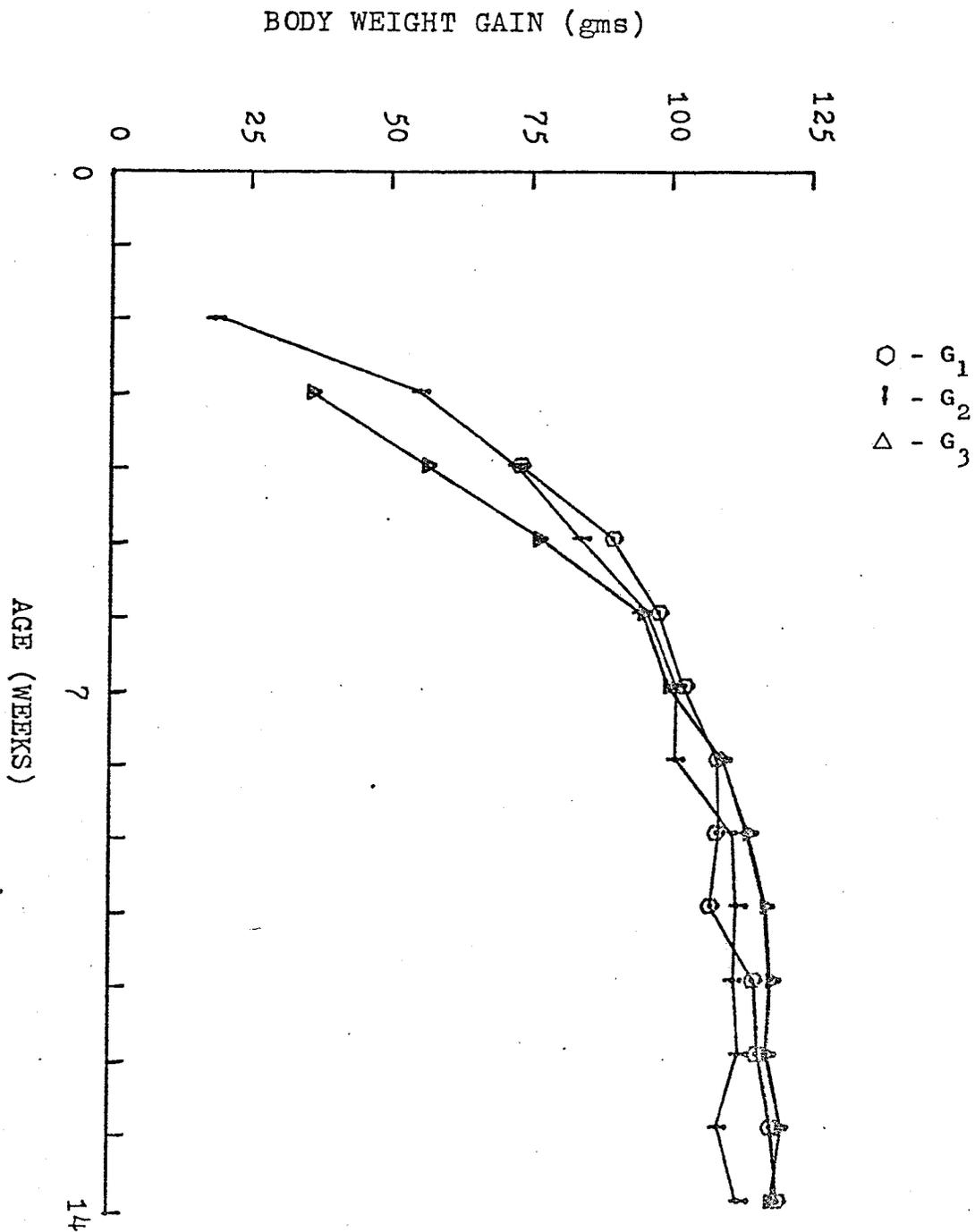


Fig. 3:5.--Fourteen weeks body weight gain for 3 generations of Japanese quail fed Abate at 8 ppm

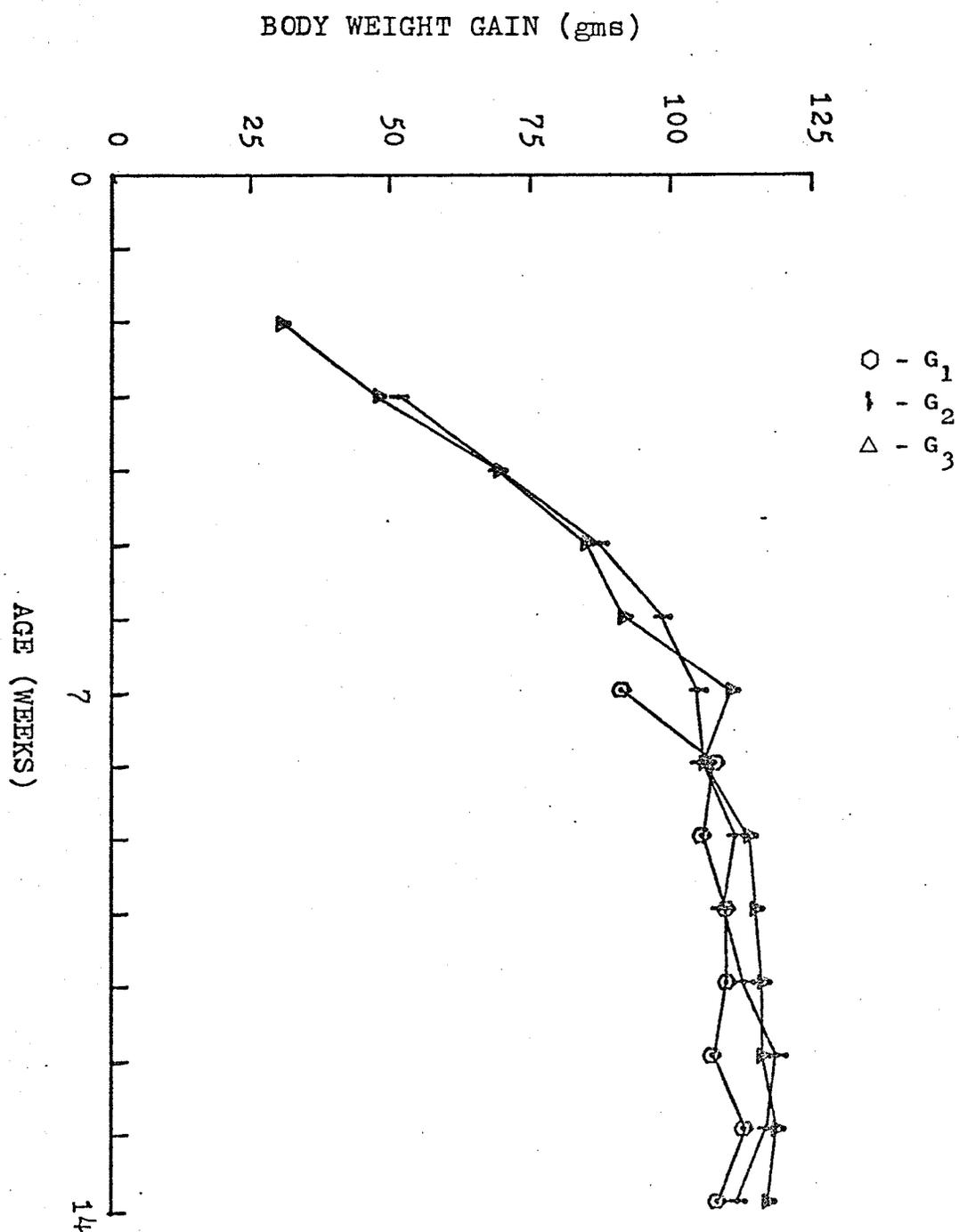


Fig. 3:6.--Fourteen weeks body weight gain for 3 generations of Japanese quail fed Abate at 32 ppm

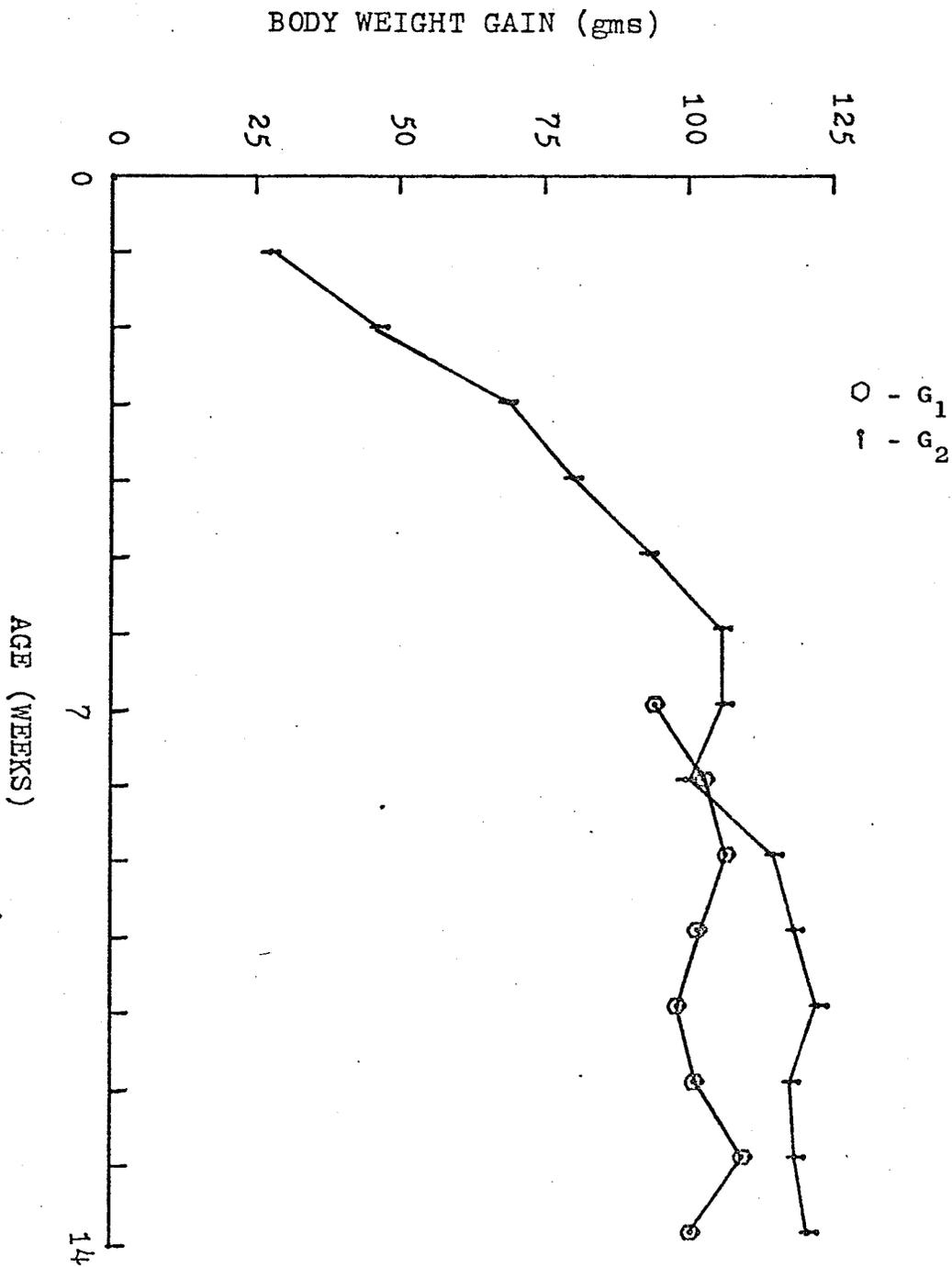


Fig. 3:7.--Fourteen weeks body weight gain for 2 generations of Japanese quail fed Abate at 80 ppm

PERCENT HEN-DAY PRODUCTION

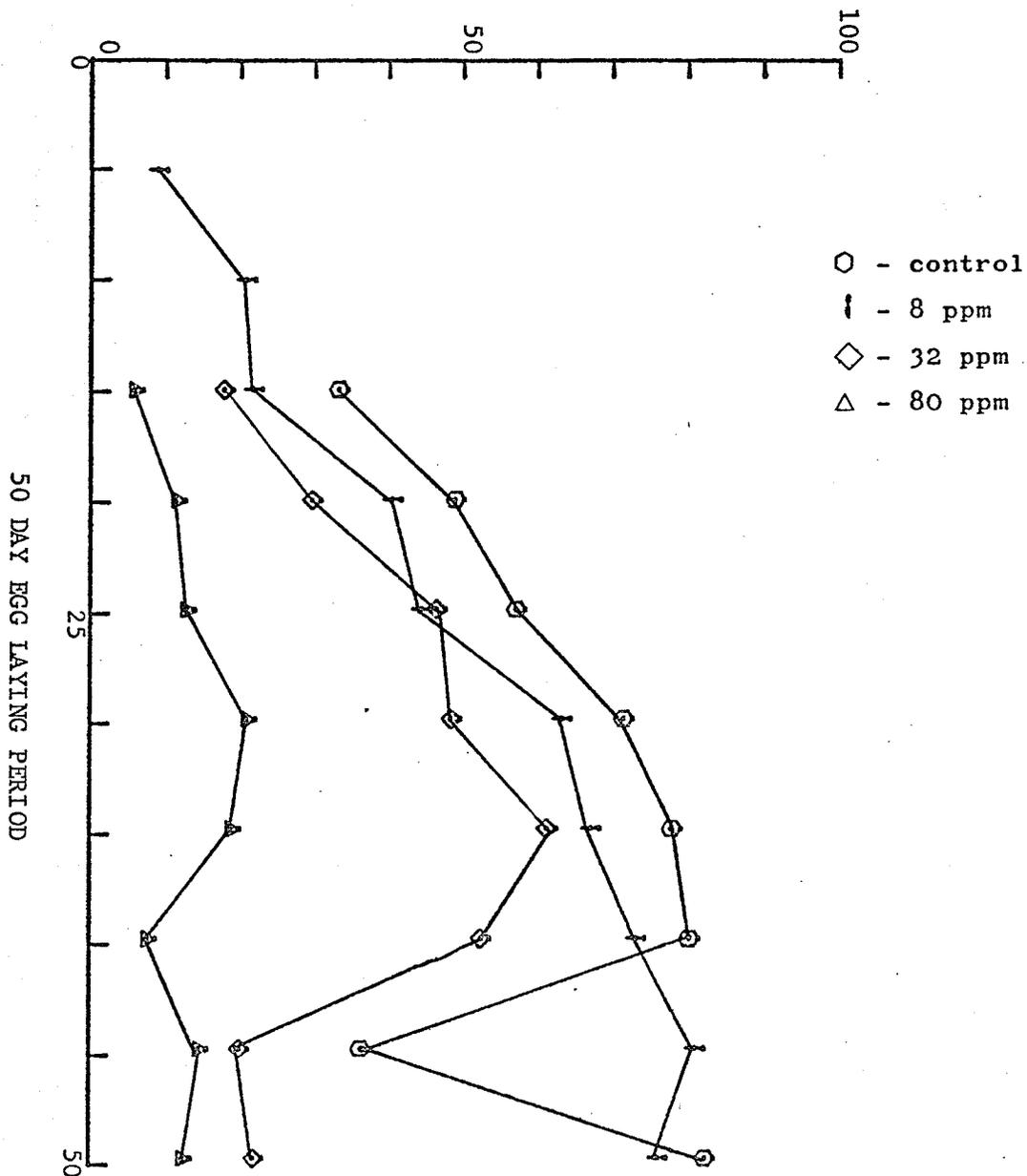


Fig. 3:8.--Egg production of Japanese quail on four Abate treatments in generation I

PERCENT HEN-DAY PRODUCTION

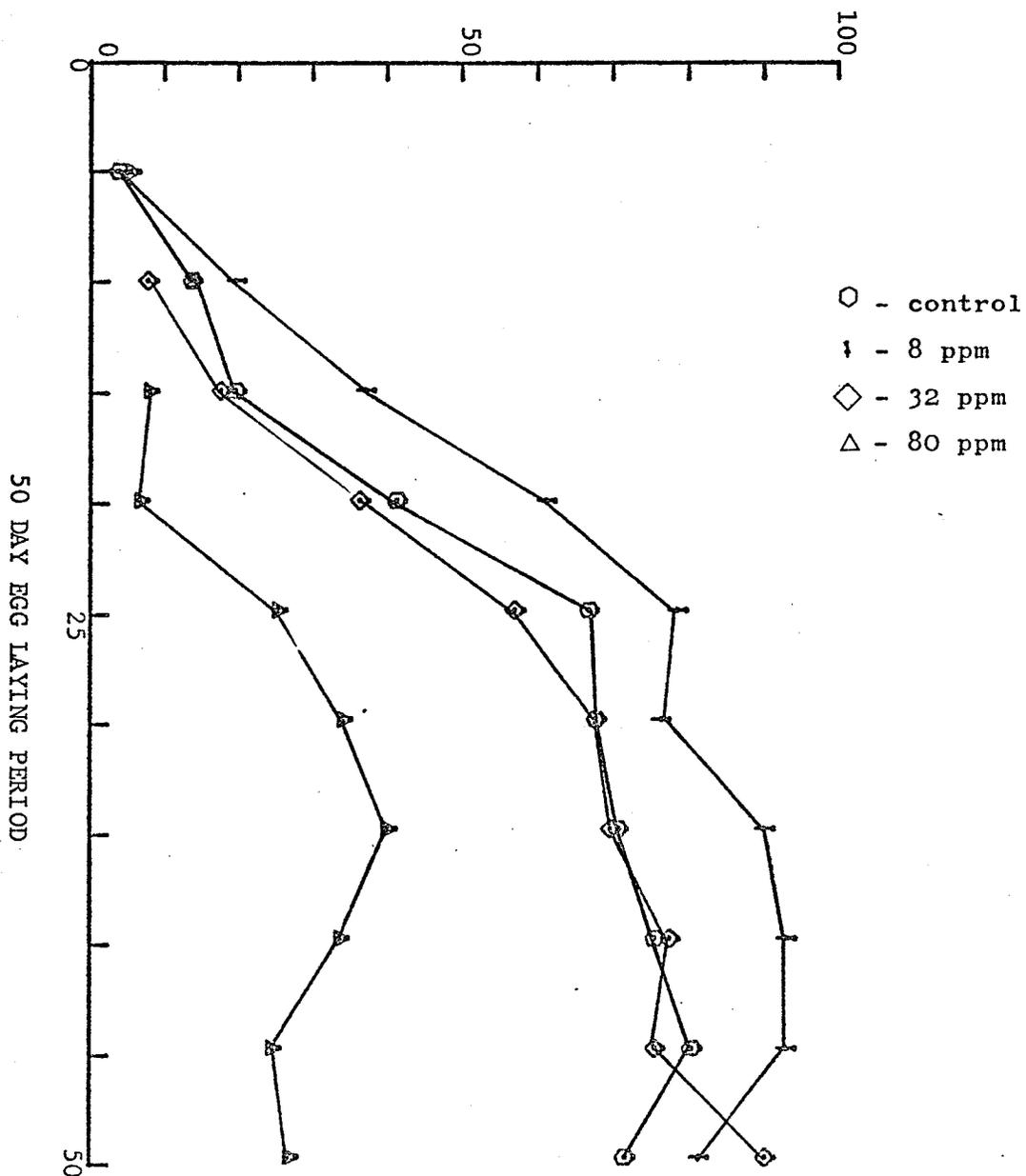


Fig. 3:9.--Egg production of Japanese quail on four Abate treatments in generation II

PERCENT HEN-DAY PRODUCTION

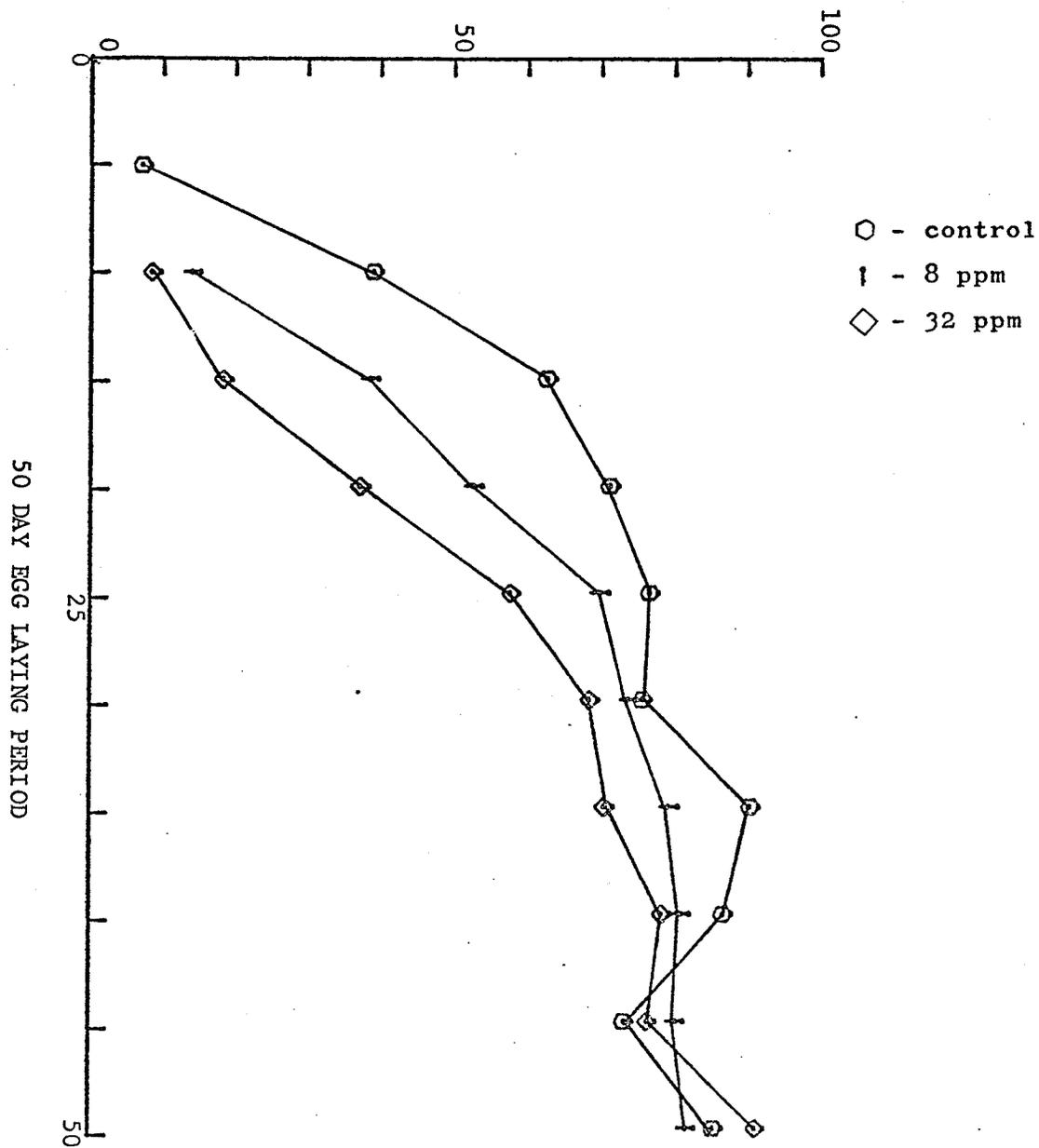


Fig. 3:10--Egg production of Japanese quail on three Abate treatments in generation III

TABLE 3:3

WILCOXON TWO SAMPLE TEST VALUES FOR EGG PRODUCTION
BETWEEN ABATE TREATMENTS WITHIN GENERATIONS

Gener- ation	Treatments ^{1,2}					
	0 vs 8	0 vs 32	0 vs 80	8 vs 32	8 vs 80	32 vs 80
I		47*(32)	36*(80)		52.5*(80)	39*(80)
II			77.5*(80)		73*(80)	
III			90(32)			

¹Wilcoxon values with an asterisk indicates the 2 treatments being compared are significantly different at the 5% level.

²Figures in brackets represent the treatment (ppm) in the comparison with the lowest mean weight. If no significance occurred, only one value per generation is presented and this value is the closest value to significance that occurred in that generation.

Fig. 3:8 to 3:10 was calculated for a 50 day laying period. The thirty-sixth day of age was used as day one and production was calculated as % hen-day production for the subsequent 50 days (see Appendix I for calculations of hen-day production).

One factor that can be noted in Fig. 3:8 is the large significant decrease in egg production for the control, 32 and 80 ppm treatment birds from day 35 to day 45. There was a subsequent recovery by day 50 for the control and partial recovery for the 32 and 80 ppm treatment birds. Birds on the 8 ppm level were not reared under the same stress. Commencing on day 35 it was extremely noisy and warm in the rearing room due to the installation of an air conditioning system. Work was completed by day 45. Thus this stress factor seriously hindered egg production during this period. Generation II and III of Abate and all three generations of Sencor were reared subsequent to this event with no such depressions in egg production.

When comparing egg production results for generation I, no significant differences occurred between birds on the 8 ppm treatment and the control (Fig. 3:8, Table 3:3). Significant differences did occur between the control and 32 ppm, control and 80 ppm, 8 and 80 ppm and between 32 and 80 ppm treatments (Fig. 3:8, Table 3:3). In all cases significance means that birds fed the lower dose of Abate had higher egg production per bird.

In generation II significant differences were found between

the control and 80 ppm and between the 8 and 80 ppm treatments (Fig. 3:9, Table 3:3). Generation II birds on the highest treatment (80 ppm) produced very few eggs and none were fertile. As a result a third generation was not produced on this treatment. In generation III, with elimination of the 80 ppm treatment group no significant differences occurred (Fig. 3:10, Table 3:3).

(b) Comparison of egg production between the three generations of Japanese quail within treatments of Abate

Comparisons were made within the controls and within the three treatments for between the three generations of birds fed Abate. No significant differences in egg production were found between the control groups of each generation (Fig. 3:11, Table 3:4). The decline in egg production occurring in generation I of the controls between day 35 and 45 was explained previously. No significant differences were found between the generations of Japanese quail on the 8 ppm treatment (Fig. 3:12, Table 3:4). On the 32 ppm treatment, generation I produced significantly less eggs than generation III (Fig. 3:13, Table 3:4). Yet there was no difference between generation I and II. At the 80 ppm treatment two generations were produced with no significant differences between them (Fig. 3:14, Table 3:4). The third generation was not produced as indicated earlier.

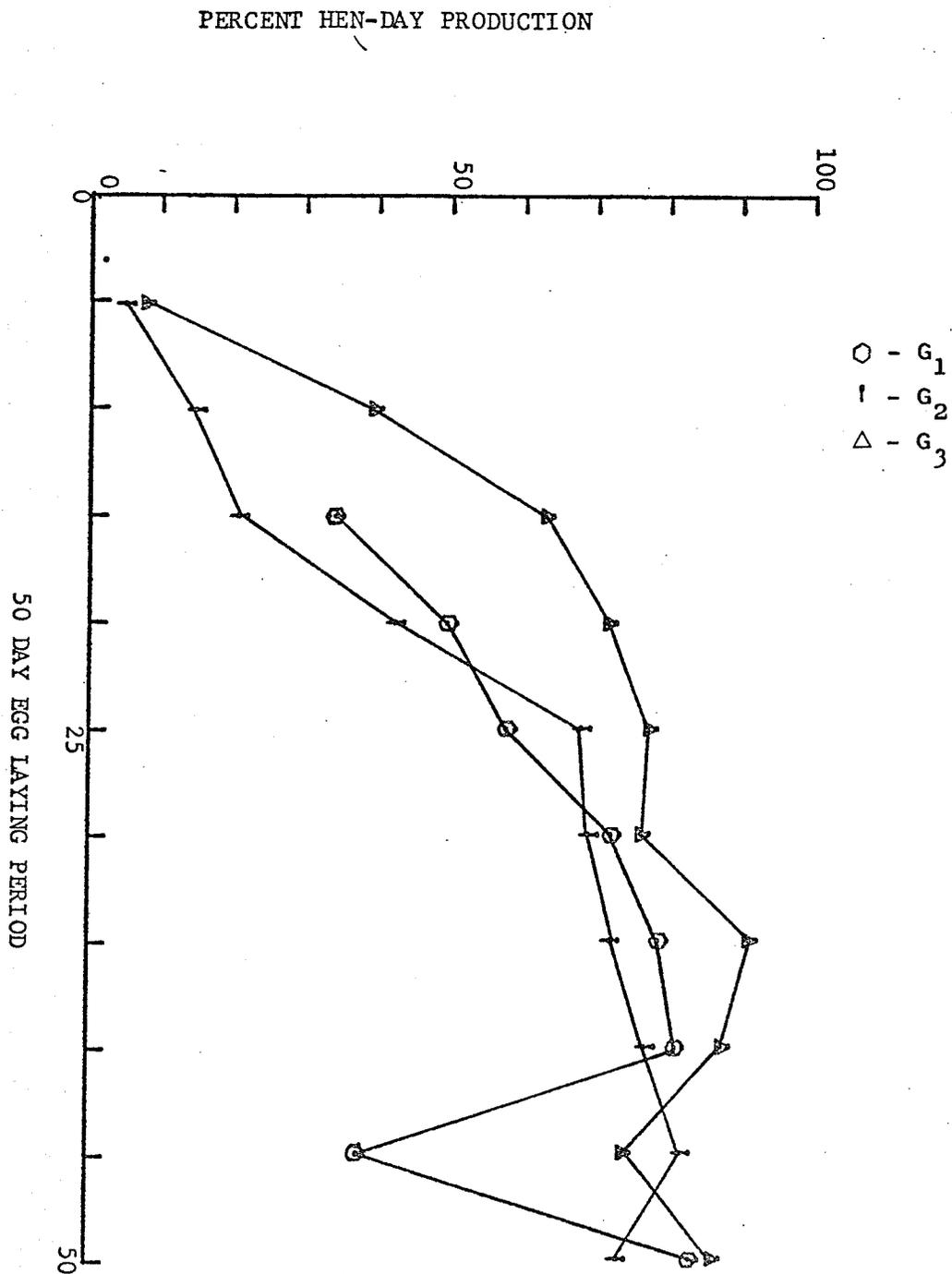


Fig. 3:11.--Egg production of Japanese quail fed Abate at 0 ppm for three generations

PERCENT HEN-DAY PRODUCTION

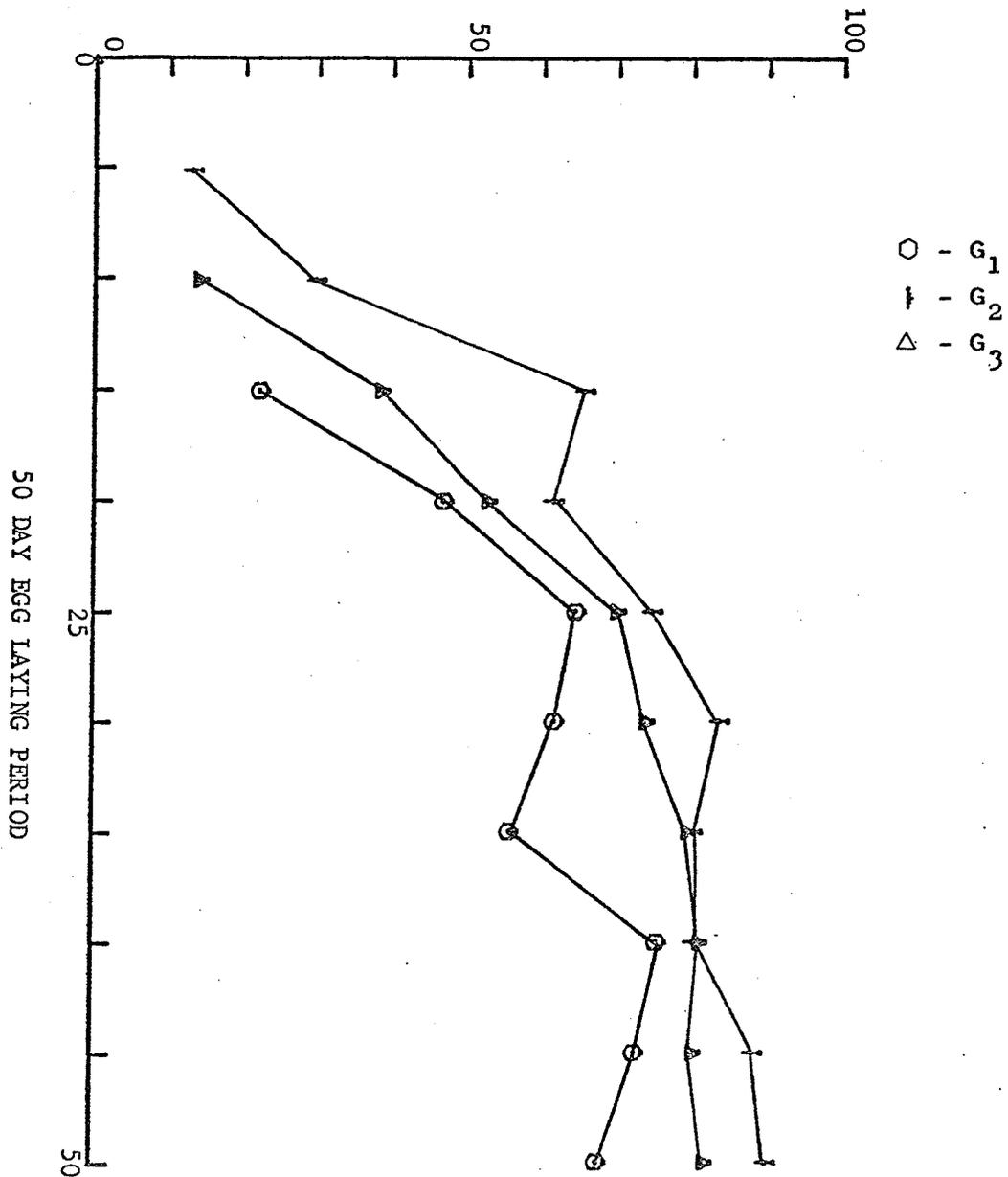


Fig. 3:12.--Egg production of Japanese quail fed Abate at 8 ppm for three generations

PERCENT HEN-DAY PRODUCTION

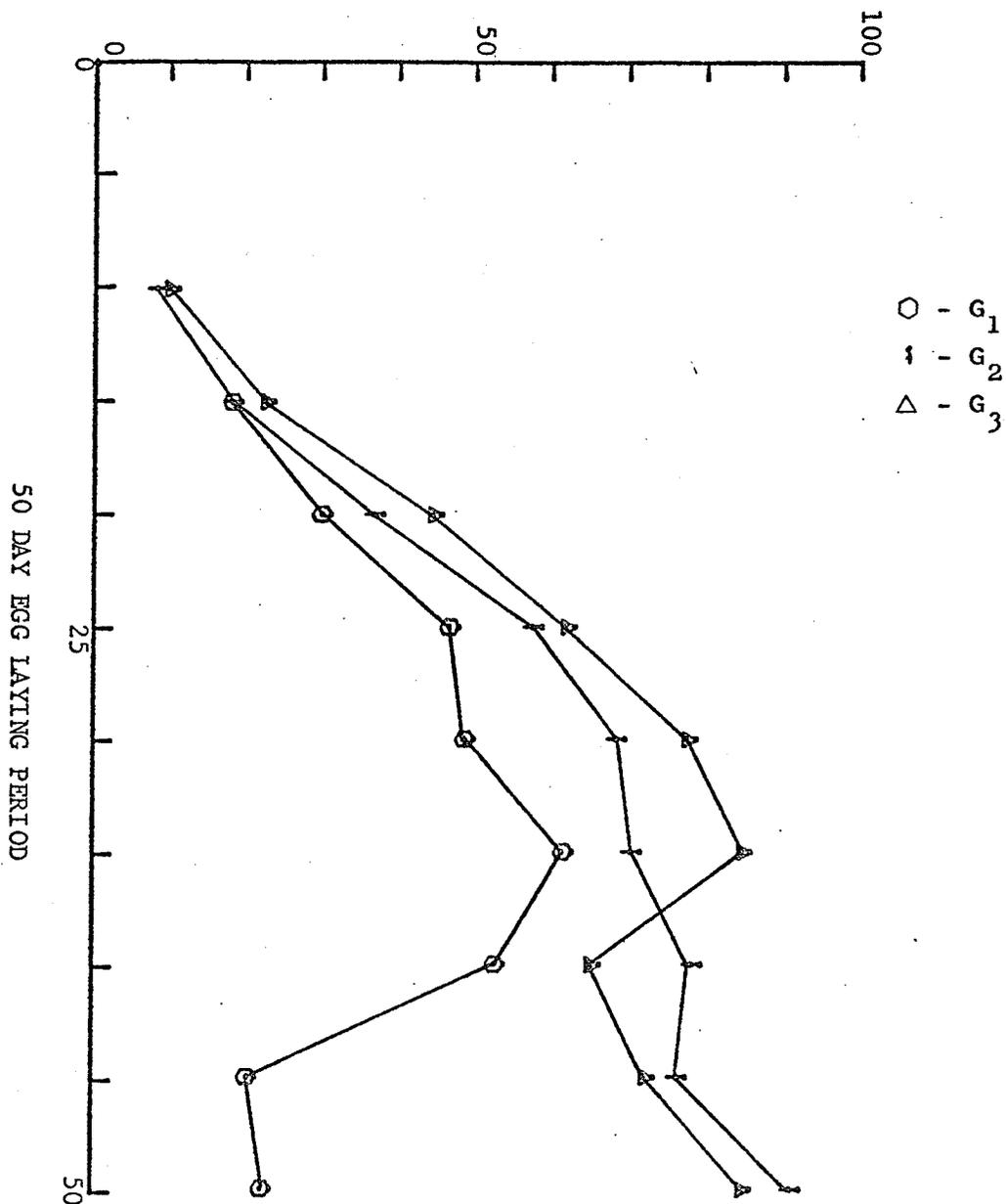


Fig. 3:13.--Egg production of Japanese quail fed Abate at 32 ppm for three generations

PERCENT HEN-DAY PRODUCTION

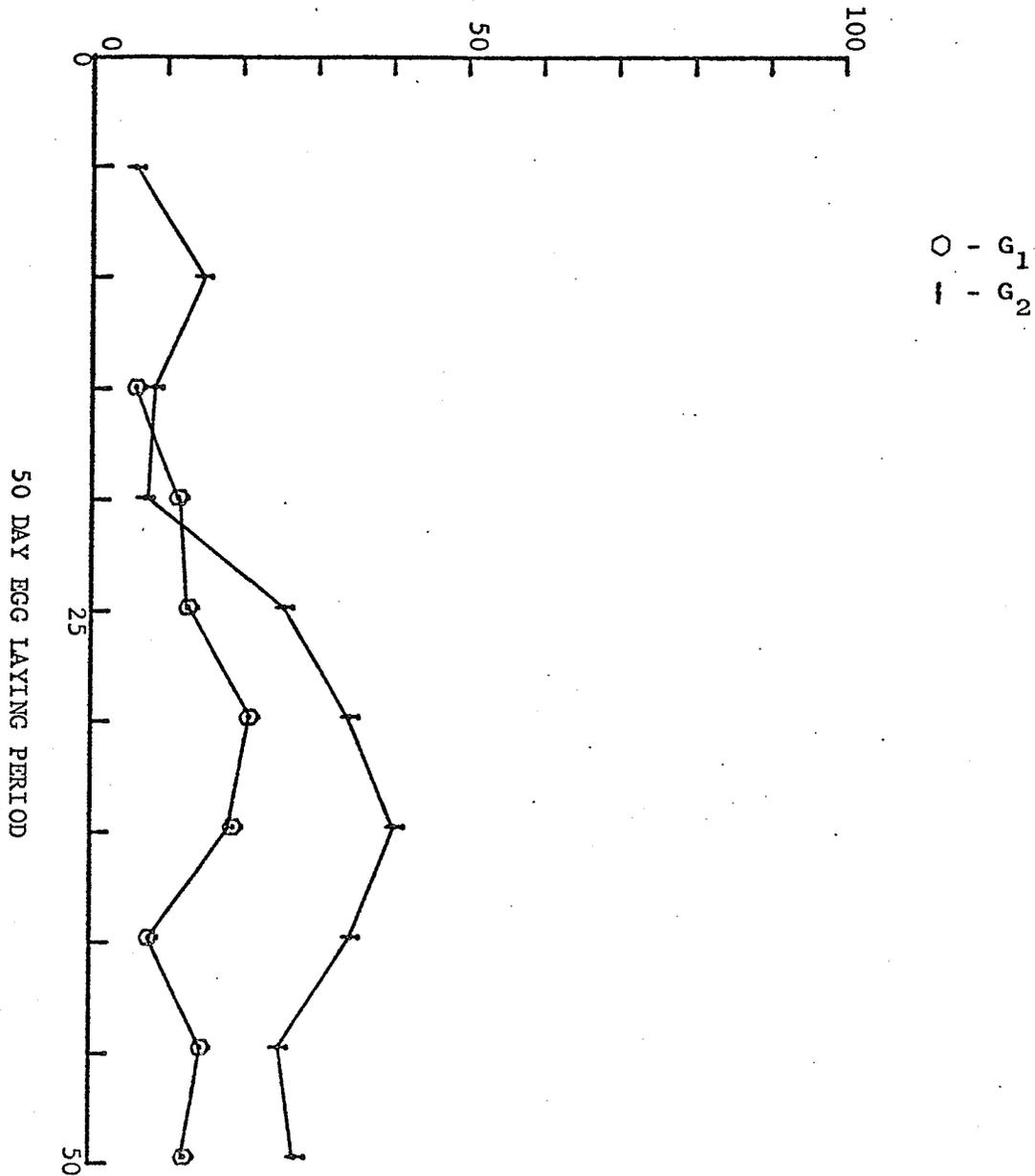


Fig. 3:14.--Egg production of Japanese quail fed Abate at 80 ppm for two generations

TABLE 3:4

WILCOXON TWO SAMPLE TEST VALUES FOR EGG PRODUCTION
BETWEEN GENERATIONS WITHIN ABATE TREATMENTS

Treatment (ppm)	Generations ^{1,2}		
	I vs II	II vs III	II vs III
0		86 (I)	
8	79 (I)		
32	65.5 (I)	62* (I)	
80	79 (I)		

¹ Wilcoxon value with an asterisk indicates the 2 generations being compared are significantly different at the 5% level.

² The generation in brackets behind each Wilcoxon value had the lowest mean in the comparison. If no significance occurred, only one value per treatment is presented and this value is the closest value to significance that occurred in that treatment.

IV. (a) Organ and body weight comparisons
between Abate treatments within the
three generations of Japanese quail

Table 3.5 presents the F values for the analysis of variance between the generations I, II and III for organ and body weights measured at 14 weeks of age. Tables 3:6 to 3:8 are the mean weights for each organ and body weight of each treatment in each generation. Tables 3:6 to 3:8 also contain the results of a Duncan's test on the means between the different treatments.

Liver

In generation I the birds fed 80 ppm Abate had significantly lower liver weights than the birds fed 0, or 8 ppm Abate (Table 3:6). Generation II showed some liver weight differentiation between the treatment groups (Table 3:7). For instance there is a significant rise in liver weight from the control to the 8 ppm treatment, while mean liver weight of the 80 ppm treated birds is significantly less than those on 8 or 32 ppm Abate, but the same as the control birds. Generation III showed a similar rise and fall in liver weight means although no significant differences occurred (Table 3:8).

Heart

In generation I the birds fed 0 to 8 ppm Abate had significantly heavier hearts than those on the 32 or 80 ppm treatments (Table 3:6). In both generation II and III, no significance was found between heart weights (Tables 3:5, 3:7, 3:8).

TABLE 3.5

ANALYSIS OF VARIANCE, F VALUES, FOR ORGAN AND BODY WEIGHTS AT 14 WEEKS OF AGE FOR THREE GENERATIONS OF ABATE FED JAPANESE QUAIL¹

Gener- ation	ORGAN					
	Liver	Heart	Lung	Kidney	Ovary	Body wt.
I	13.10*	4.56*	2.50	9.61*	19.19*	16.68*
II	6.80*	1.80	1.12	1.42	7.51*	5.99*
III	.13	2.90	2.45	2.13	0.50	4.22*

¹F values with asterisk are significant at the 5% level

TABLE 3:6

DUNCAN'S COMPARISON FOR ORGAN AND BODY WEIGHT MEANS (GMS) FOR BIRDS AT 14 WEEKS OF AGE BETWEEN ABATE TREATMENTS WITHIN GENERATION I^{1,2}

Treatment (ppm)	ORGAN					
	Liver	Heart	Lung	Kidney	Ovary	Body Wt.
0	5.57a	0.92abc	1.19a	1.77a	4.71bc	123.42a
8	5.28a	0.97a	1.18a	1.79a	5.78c	123.36a
32	4.46ab	0.85b	1.05a	1.55b	3.74b	114.56b
80	4.08b	0.86bc	1.02a	1.31c	1.80a	103.70c

¹Means with the same subscript letters are not significantly different at the 5% level of significance.

²Weights (gms) are the mean of 25 birds.

TABLE 3:7

DUNCAN'S COMPARISON FOR ORGAN AND BODY WEIGHT MEANS (GMS)
FOR BIRDS AT 14 WEEKS OF AGE BETWEEN ABATE
TREATMENTS WITHIN GENERATION II^{1,2}

Treatment ppm	Liver	Heart	Lung	Kidney	Ovary	Body wt.
0	5.11b	1.08a	1.21a	1.81a	5.64a	123.51b
8	5.98a	1.09a	1.22a	1.89a	6.03a	127.80b
32	5.77a	1.05a	1.26a	1.83a	5.85a	134.44a
80	5.10b	0.98a	1.34a	1.65a	4.01b	125.77b

¹ Means with the same subscript letters are not significantly different at the 5% level of significance.

² Weights (gms) are the mean of 25 birds

TABLE 3:8

DUNCAN'S COMPARISON FOR ORGAN AND BODY WEIGHT MEANS (GMS)
FOR BIRDS AT 14 WEEKS OF AGE BETWEEN ABATE
TREATMENTS WITHIN GENERATION III^{1,2}

Treatment ppm	Liver	Heart	Lung	Kidney	Ovary	Body wt.
0	5.60a	1.12a	1.17a	1.85a	4.95a	125.74ac
8	5.76a	1.12a	1.15a	1.70a	5.39a	121.08bc
32	5.59a	1.03a	1.26a	1.84a	5.40a	129.38a

¹ Means with the same subscript letters are not significantly different at the 5% level of significance.

² Weights (gms) are the mean of 25 birds.

Lung

Table 3:5 shows no significant differences in the lung weight between the various treatment levels in all three generations.

Kidney

In generation I birds on the 0, 8 and 32 ppm treatments had significantly heavier kidney mean weights than on the 80 ppm treatment (Table 3:6). In addition the control and the 8 ppm treatment, birds were significantly heavier than on the 32 ppm treatment (Table 3:6). As can be seen from Tables 3:5, 3:7, 3:8, tests of significance for generations II and III showed no significant weight differences for the kidney at the end of the fourteen week experimental period.

Ovary

In generation I ovary weights for 0, and 8 ppm fed birds were the same. As Abate intake increased to 32 ppm there was a significant decrease in ovary weight and a further significant decrease as the intake increased to 80 ppm for a fourteen week period. In generation II ovary weights were lower when birds were fed 80 ppm Abate than when fed 0, 8 or 32 ppm (Table 3:7). Generation III had no significant differences in the ovary mean weights, (Table 3:8).

Body Weight

Body weight differences did occur in all three generations. In generation I a pattern did seem to occur (Table 3:6). That is,

weights declined significantly on the 32 and 80 ppm treatments. Generation II and III did not show this pattern although significances in mean weights did occur (Table 3:7 and 3:8). In fact, in generation II the 32 ppm treated birds had a significantly greater mean body weight than the controls.

(b) Comparison of internal organ and body weight between the three generations of Japanese quail within treatments of Abate

Table 3:9 is the analysis of variance between the three generations within treatments for each organ and body weight. To determine the generation between which the group means are significantly different, Duncan's tests were done and are presented on Table 3:10.

As Table 3:10 points out, any significance that occurred in organ or body weights between the generations on any one treatment occurred only with the first generation, which on the whole was lighter. One exception occurred. In this case the lung in generation I of the control birds was significantly larger than generation II and III (Table 3:10). No differences occurred between generation II and III, except at the 8 ppm treatment for body and heart weight (Table 3:10). In the case of body weight, generation I and III were significantly smaller than generation II, and in the case of heart weight, generation II was significantly smaller than generation III.

TABLE 3:9

ANALYSIS OF VARIANCE, F-VALUES, FOR ORGAN AND BODY WEIGHTS
 AT 14 WEEKS OF AGE BETWEEN GENERATIONS WITHIN
 TREATMENTS FOR ABATE FED JAPANESE QUAIL¹

Treatment (ppm)	ORGAN					
	Liver	Heart	Lung	Kidney	Ovary	Body wt.
0	2.27	10.32*	4.13*	0.24	2.28	0.39
8	3.05	6.51*	0.34	1.49	0.90	3.30*
32	2.00	17.21*	7.65*	3.42*	12.04*	22.88*
80	17.28*	8.18*	9.72*	35.59*	15.08*	49.51*

¹F values with asterisk are significant at the 5% level.

TABLE 3:10

DUNCAN'S COMPARISON FOR ORGAN AND BODY WEIGHT MEANS (GMS)
 BETWEEN GENERATIONS WITHIN ABATE TREATMENTS¹

Organ	Gener- ation	Mean Weight of Organs (gms)			
		Control	8 ppm	32 ppm	80 ppm
Liver	I	5.57a	5.30a	5.44a	4.07b
	II	5.11a	5.98a	5.83a	5.10a
	III	5.60a	5.77a	5.61a	
Heart	I	.92b	0.95b	0.86b	.82b
	II	1.08a	1.09b	1.05a	.95a
	III	1.11a	1.13a	1.03a	
Lung	I	1.45b	1.16a	1.06b	1.01b
	II	1.21a	1.19a	1.26a	1.84a
	III	1.17a	1.15a	1.25a	
Kidney	I	1.77a	1.78a	1.57b	1.31b
	II	1.01a	1.89a	1.82a	1.67a
	III	1.85a	1.70a	1.84a	
Ovary	I	4.71a	5.75a	3.58b	1.81b
	II	5.68a	6.03a	5.85a	4.01a
	III	4.95a	5.39a	5.40a	
Body Weight	I	123.21a	122.95a	115.65b	102.57b
	II	123.51a	127.80b	134.44a	125.74a
	III	125.76a	121.08a	129.38a	

¹Means with the same subscript letter are not significantly different at the 5% level of significance.

B. Sencor

I. General Comments

Japanese quail fed the herbicide Sencor at 37.2, 149 and 372 ppm in their diet displayed no physical symptoms of poisoning and in all three generations appeared healthy. Data for body and organ weights, at treatment level 37.2 ppm generation III was not collected due to a critical time factor at the end of the experiment which forced these observations to be omitted. This should not be critical to the experiment since it was the lowest treatment level and few differences showed up even in the higher dose levels.

II. (a) Body weight gain between Sencor treatments within the three generations of Japanese quail

The mean data taken from the body weight gain curves for different treatments (0, 37.2, 149, and 372 ppm) was compared using the Wilcoxon test for unpaired data and no differences in weight gain were found between treatments within generation I, II, or III (Fig. 3:15, 3:16, 3:17 and Table 3:11). No explanation can be given for the dip occurring in treatment 372 ppm, generation III other than random chance (Fig. 3:17).

(b) Comparison of body weight gain between the three generations of Japanese quail within treatments of Sencor

Results from within treatments showed only three differences in weight gain occurred between the generations. On the 37.2 and 149

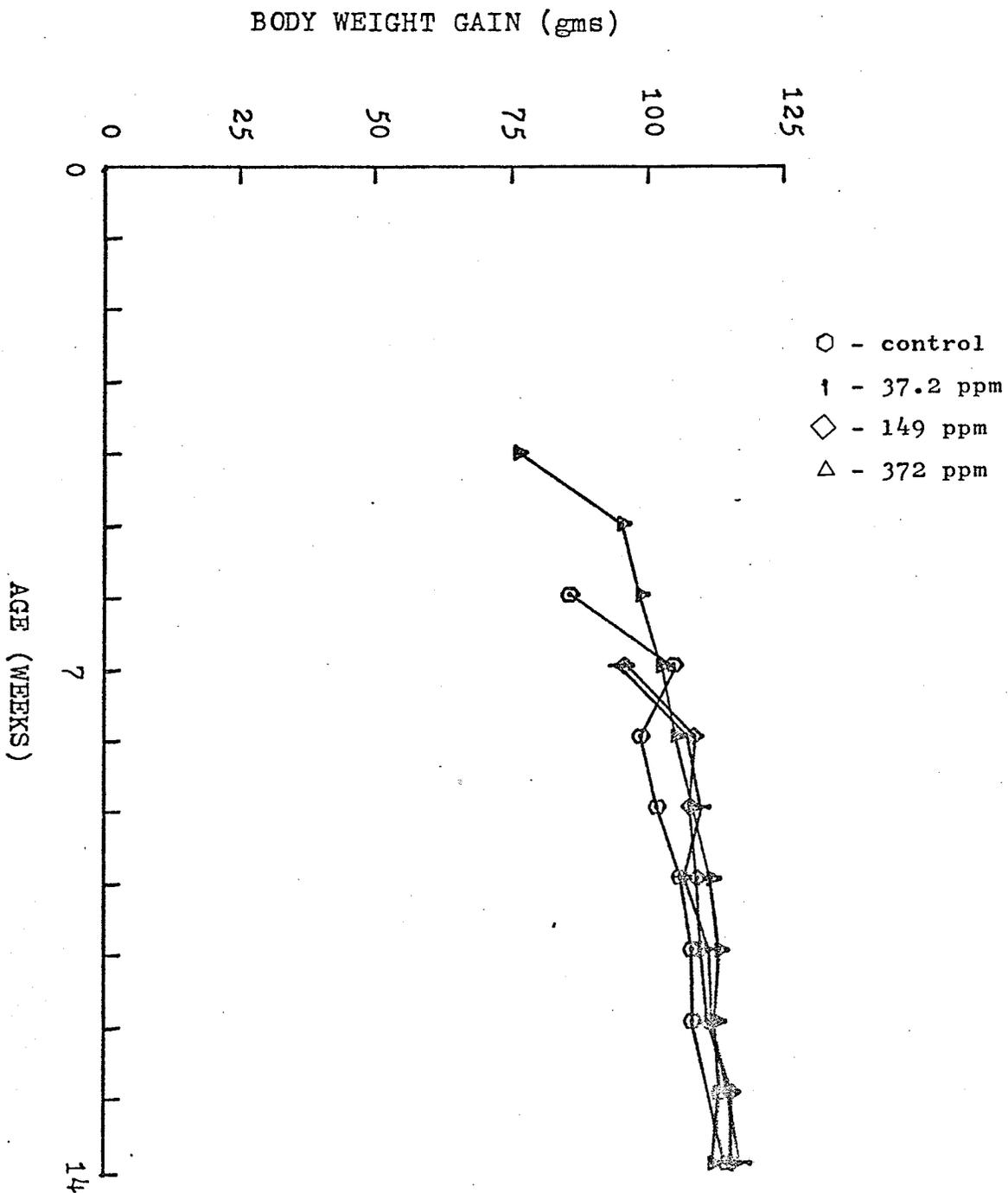


Fig. 3:15.--Fourteen weeks body weight gain of Japanese quail fed four Sencor treatments in generation I

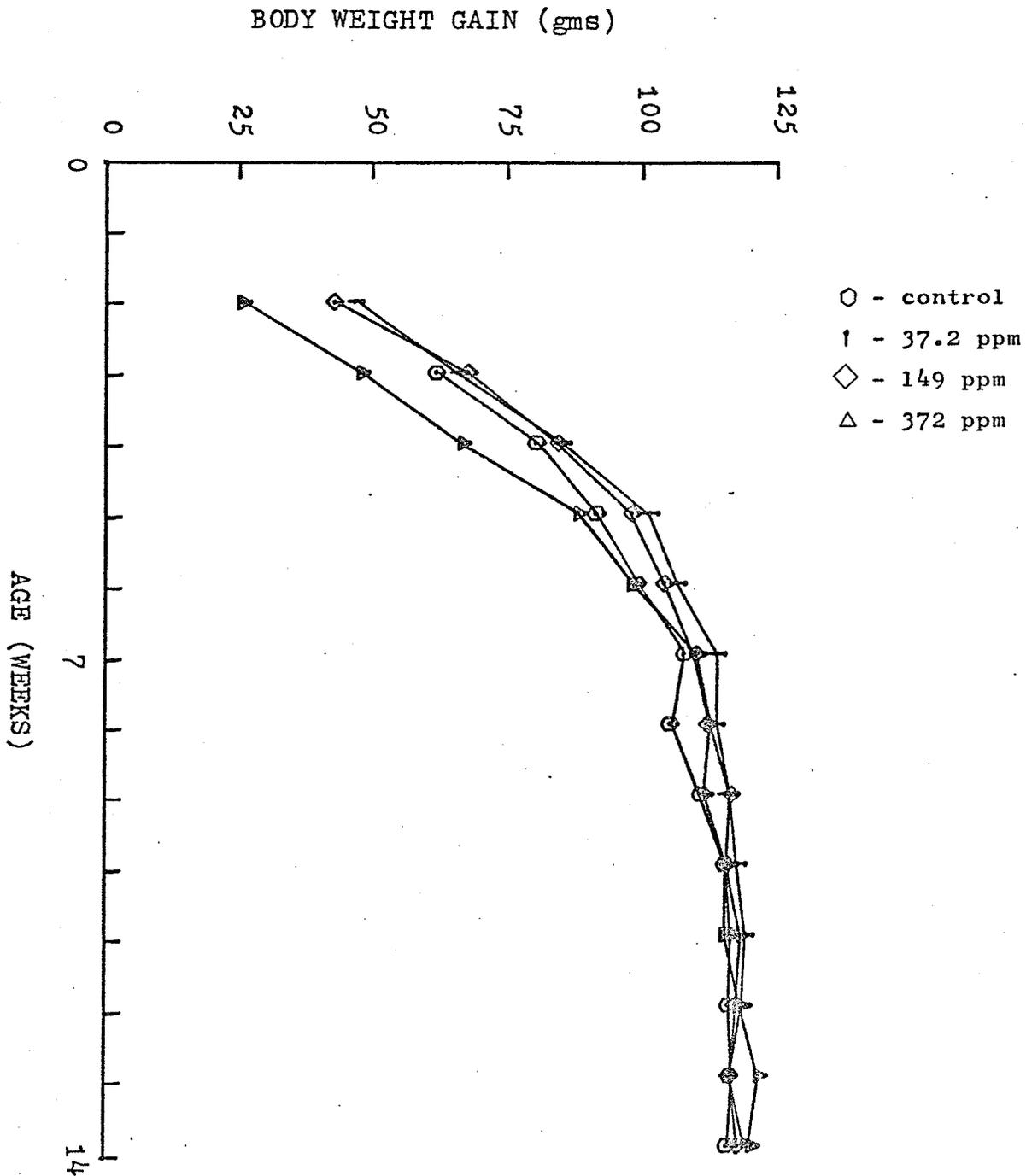


Fig. 3:16.--Fourteen weeks body weight gain of Japanese quail fed four Sencor treatments in generation II

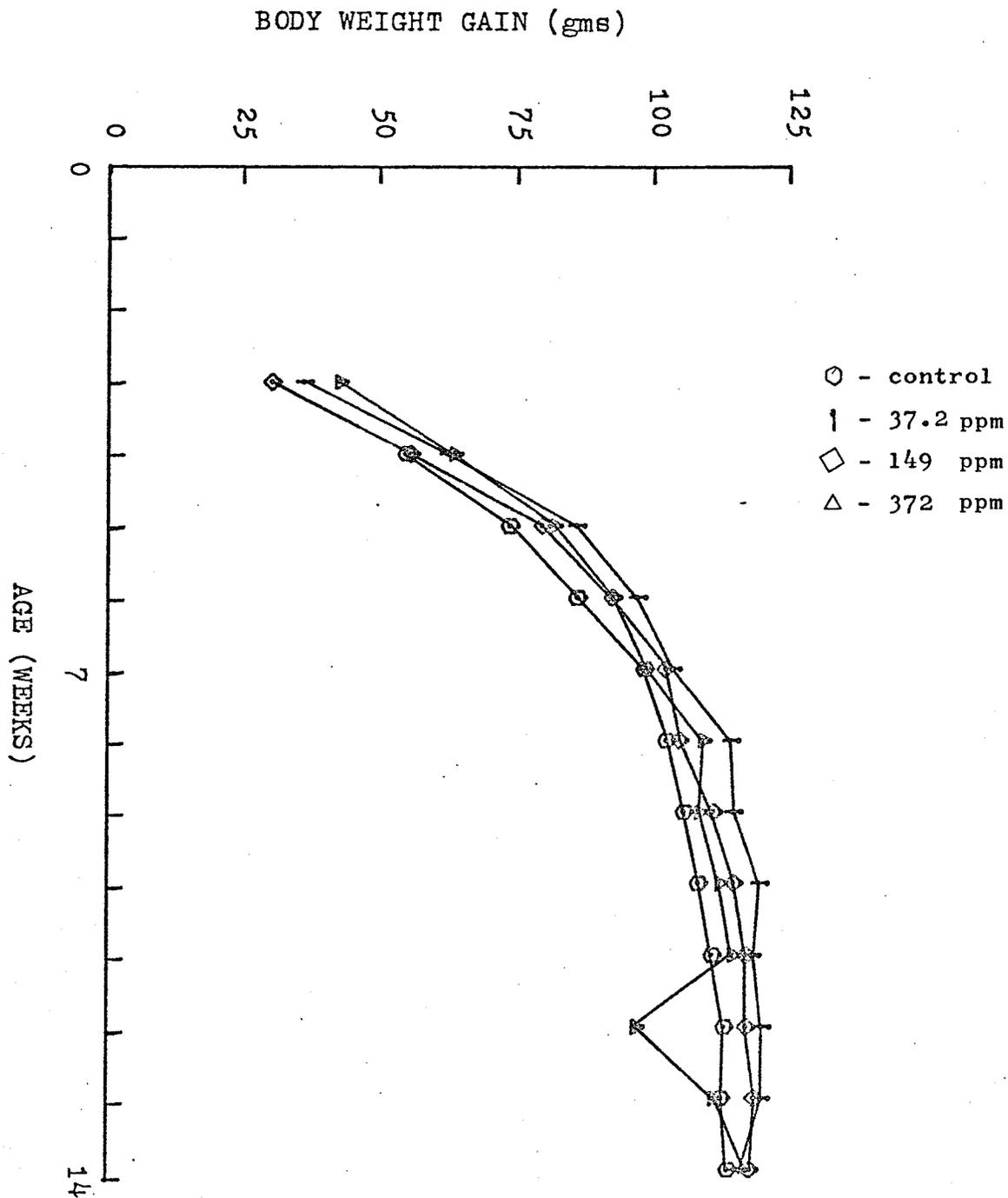


Fig. 3:17.--Fourteen weeks body weight gain of Japanese quail fed four Sencor treatments in generation III

TABLE 3:11

WILCOXON TWO SAMPLE TEST VALUES FOR BODY WEIGHT GAIN
BETWEEN SENCOR TREATMENTS WITHIN GENERATIONS

Gener- ation	Treatments ^{1,2}					
	0 vs 37.2	0 vs 149	0 vs 372	37.2 vs 149	37.2 vs 372	149 vs 372
I		56 (0)				
II		130 (0)				
III	99(0)					

¹Wilcoxon values with an asterisk indicates the two treatments being compared are significantly different at the 5% level.

²Figures in brackets represent the treatment (ppm) in the comparison with the lowest mean weight. If no significance occurred, only one value per generation is presented and this value is the closest value to significance that occurred in that generation.

ppm treatments, birds in generation I were significantly smaller than those in generation II, and the 37.2 ppm treatment birds in generation I were significantly smaller than those in generation III (Fig. 3:18, 3:19, Table 3:12). No significance occurred between birds on the 0 ppm treatment or the 372 ppm treatment (Fig. 3:4, 3:20, Table 3:12).

III. (a) Egg production between Sencor treatments within the three generations of Japanese quail

When Sencor was fed in the diet at 0, 37.2, 149 and 372 ppm for three generations, egg production was used as a parameter to determine if Sencor was affecting the birds' biological functions. Table 3:13 is the Wilcoxon tests of significance for data derived from Figures 3:21 to 3:23 representing generations I, II and III, respectively. Data for egg production was calculated on a 50 day laying period as described in the Abate section. In all cases egg production stabilized between day 25 to 50.

There were no significant differences for comparisons between treatments of generations I, II and III (Fig. 3:21, 3:22, 3:23, Table 3:13). The dip in Fig. 3:21 for the control was explained earlier in the Abate section.

(b) Comparison of egg production between the three generations of Japanese quail within treatments of Sencor

When data for the particular treatments were compared (0,

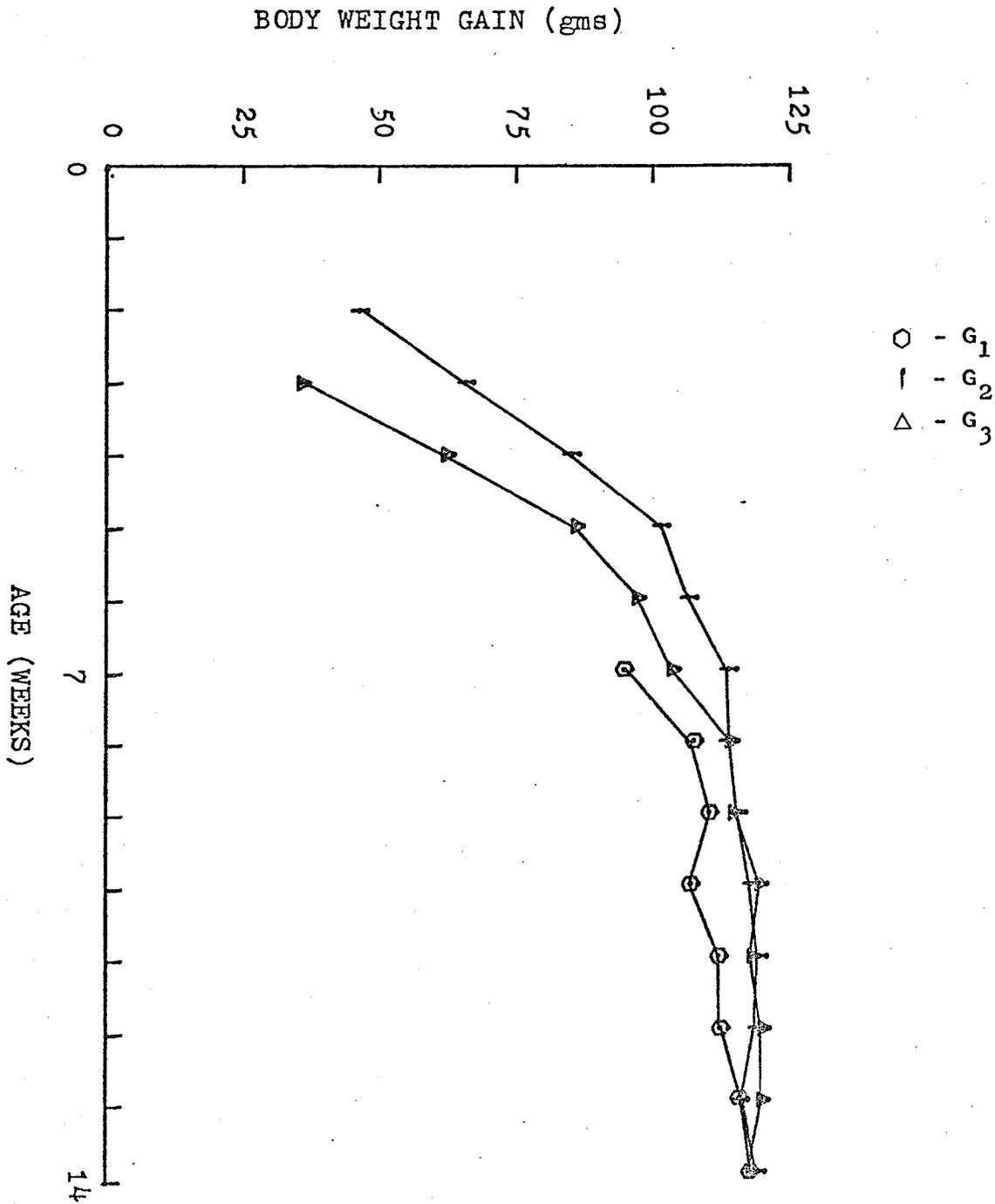


Fig. 3:18.--Fourteen weeks body weight gain for three generations of Japanese quail fed Sencor at 37.2 ppm

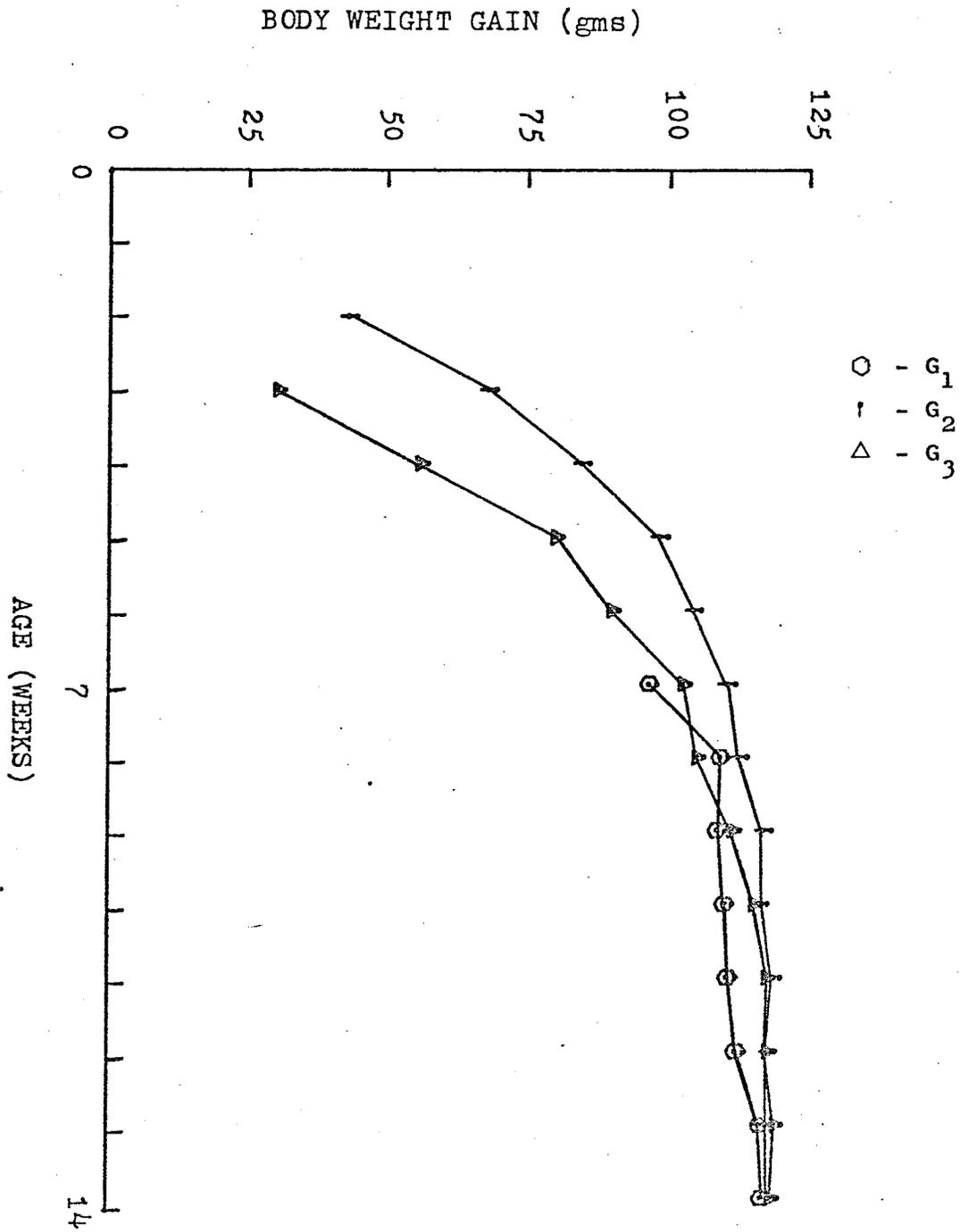


Fig. 3:19.--Fourteen weeks body weight gain for three generations of Japanese quail fed Sencor at 149 ppm

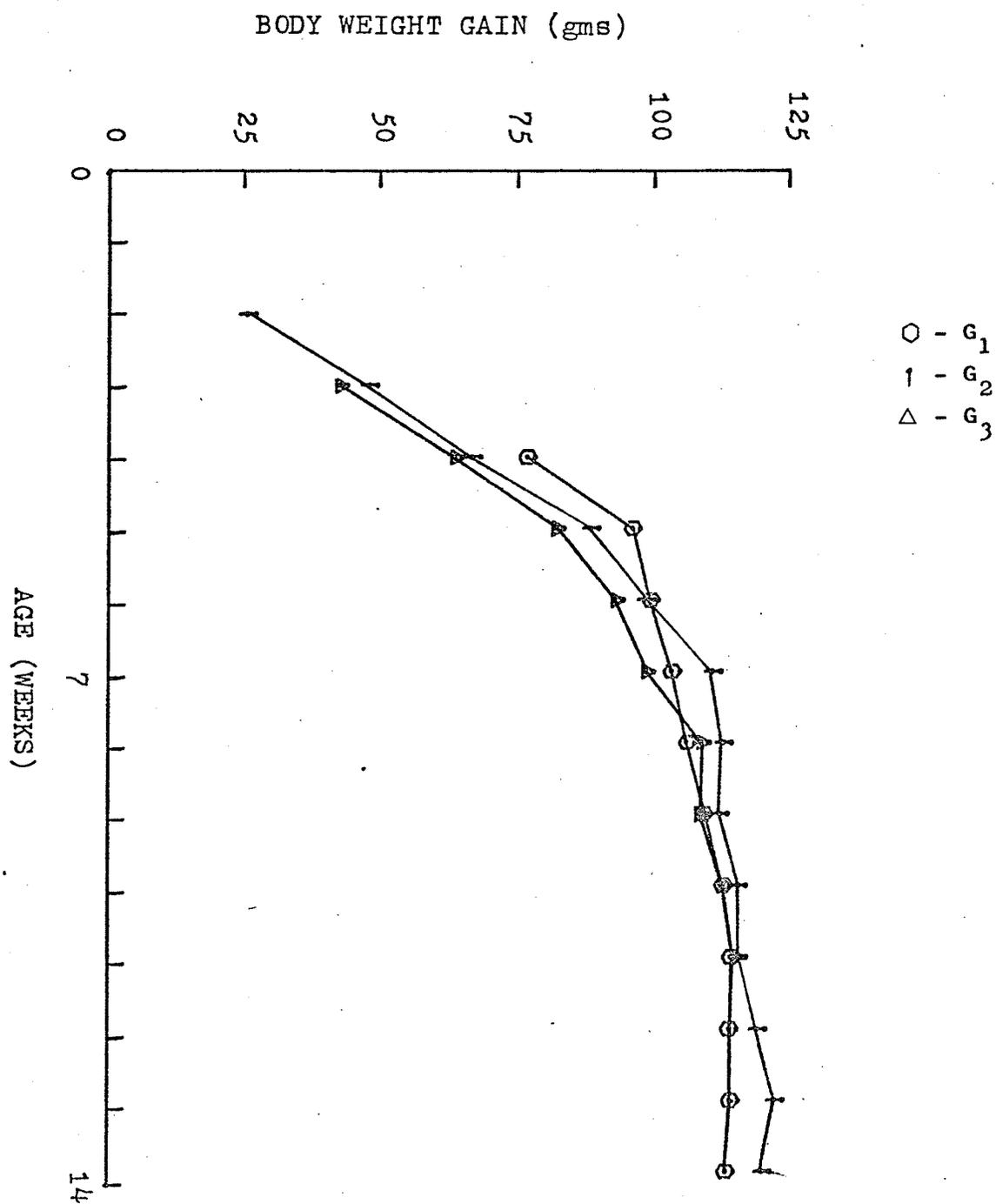


Fig. 3:20.--Fourteen weeks body weight gain for three generations of Japanese quail fed Sencor at 372 ppm

TABLE 3:12

WILCOXON TWO SAMPLE TEST VALUES FOR BODY WEIGHT GAIN
BETWEEN GENERATIONS WITHIN SENCOR TREATMENTS

Treatment (ppm)	Generations ^{1,2}		
	I vs II	I vs III	II vs III
0	65 (I)		
37.2	42* (I)	47* (I)	
149	43* (I)		
372			76 (II)

¹Wilcoxon value with an asterisk indicates the 2 generations being compared are significantly different at the 5% level.

²The generation in brackets behind each Wilcoxon value had the lowest mean in the comparison. If no significance occurred only one value per treatment is presented and this value is the closest value to significance that occurred in that treatment.

PERCENT HEN-DAY PRODUCTION

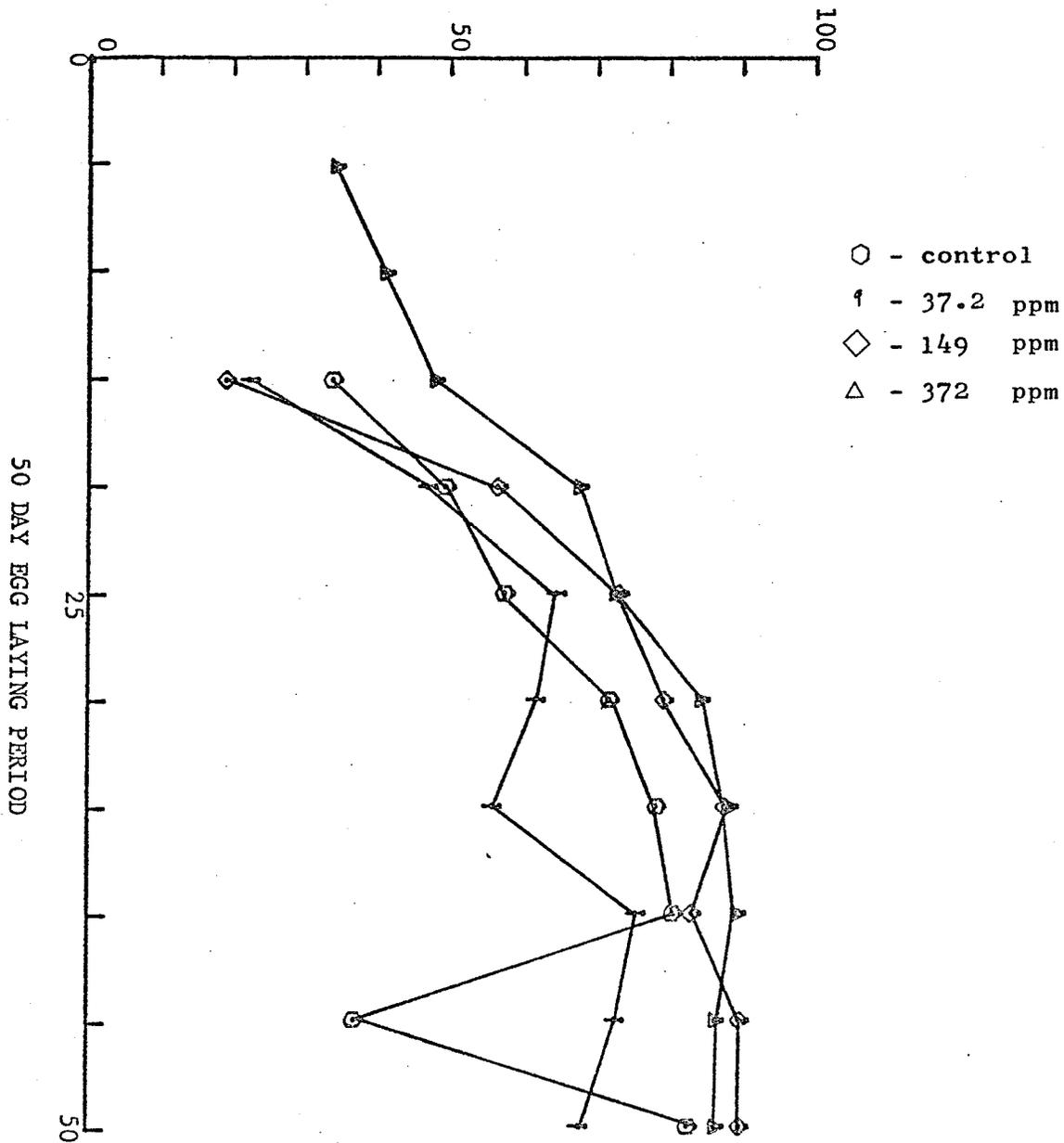


Fig. 3:21.--Egg production of Japanese quail on four Sencor treatments in generation I

PERCENT HEN-DAY PRODUCTION

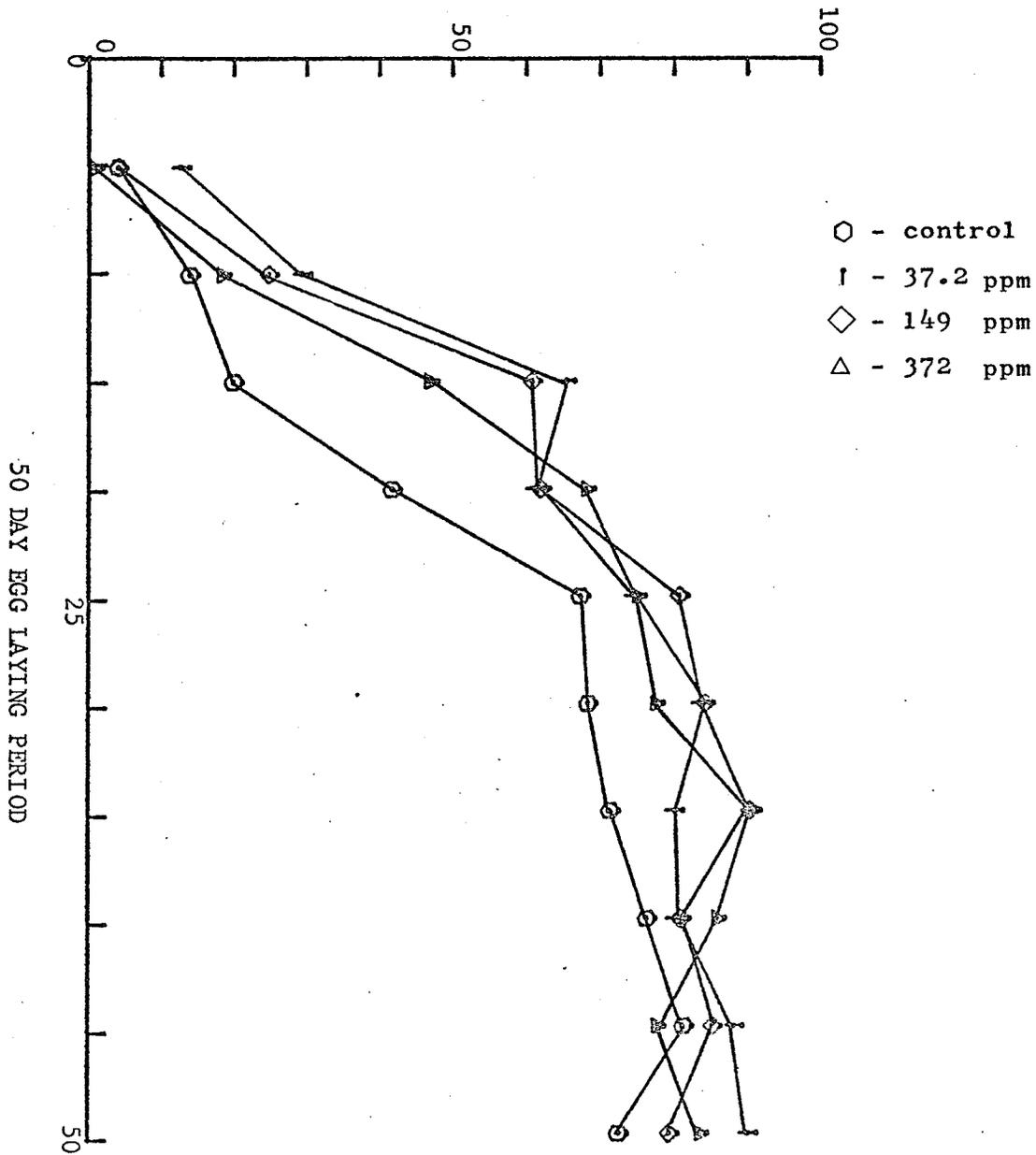


Fig. 3:22.--Egg production of Japanese quail on four Sencor treatments in generation II

PERCENT HEN-DAY PRODUCTION

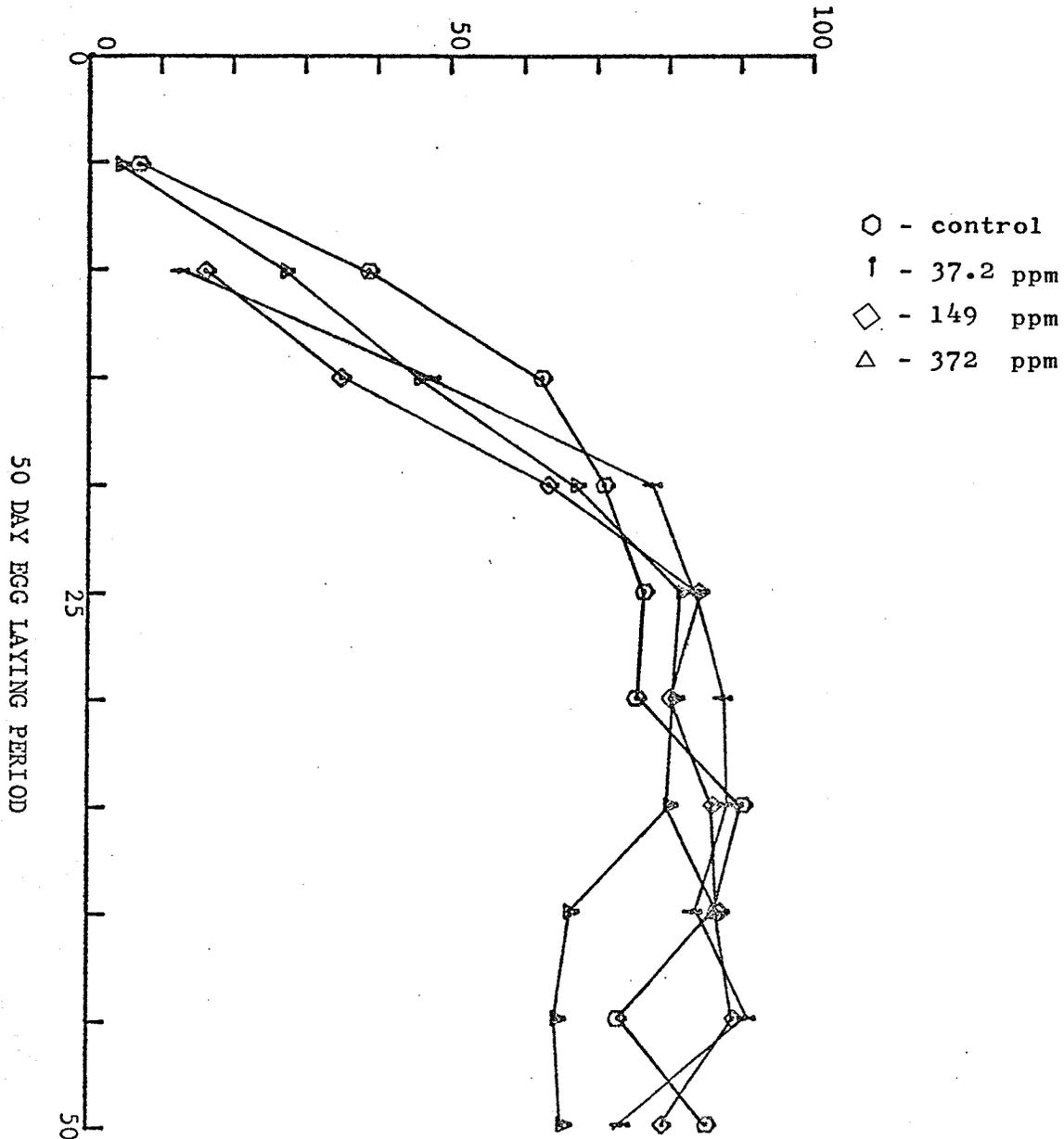


Fig. 3:23.--Egg production of Japanese quail on four Sencor treatments in generation III

TABLE 3:13

WILCOXON TWO SAMPLE TEST VALUES FOR EGG PRODUCTION
BETWEEN SENCOR TREATMENTS WITHIN GENERATIONS

Gener- ation	Treatments ^{1,2}					
	0 vs 37.2	0 vs 149	0 vs 372	37.2 vs 149	37.2 vs 372	149 vs 372
I					79 (37.2)	
II	88 (0)					
III					86 (37.2)	

¹Wilcoxon values with an asterisk indicates the two treatments being compared are significantly different at the 5% level.

²Figures in brackets represent the treatments (ppm) in the comparison with the lowest mean weight. If no significance occurred, only one value per generation is presented and this value is the closest value to significance that occurred in that generation.

TABLE 3:14

WILCOXON TWO SAMPLE TEST VALUES FOR EGG PRODUCTION
BETWEEN GENERATIONS WITHIN SENCOR TREATMENTS

Treatment (ppm)	Generations ^{1,2}		
	I vs II	I vs III	II vs III
0		86 (I)	
37.2	79 (I)		
149	100.5 (I)		
372		84 (III)	

¹Wilcoxon value with an asterisk indicates the 2 generations being compared are significantly different at the 5% level.

²The generation in brackets behind each Wilcoxon value had the lowest mean in the comparison. If no significance occurred, only one value per treatment is presented and this value is the closest value to significance that occurred in that treatment.

TABLE 3:15

ANALYSIS OF VARIANCE, F VALUES, FOR ORGAN AND BODY WEIGHT AT 14 WEEKS OF AGE FOR THE THREE GENERATIONS OF SENCOR FED JAPANESE QUAIL¹

Gener- ation	Organ					
	Liver	Heart	Lung	Kidney	Ovary	Body Wt.
I	15.43*	5.33*	.95	4.05*	43.15*	19.70*
II	4.90*	2.82*	1.58	1.89	1.25	2.25
III	24.57*	1.24	1.00	.40	2.17	.31

¹F values with asterisk are significant at the 5% level

TABLE 3:16

DUNCAN'S COMPARISON FOR ORGAN AND BODY WEIGHT MEANS (GMS) FOR BIRDS AT 14 WEEKS OF AGE BETWEEN SENCOR TREATMENTS WITHIN GENERATION I^{1,2}

Treatment (ppm)	Organ					
	Liver	Heart	Lung	Kidney	Ovary	Body wt.
0	5.57b	0.93b	1.19a	1.77ab	4.71b	123.21a
37.2	4.75c	0.93b	1.15a	1.62b	5.92a	120.96a
149	6.60a	0.90b	1.09a	1.74ab	1.82c	105.64b
372	5.85b	1.10a	1.20a	1.94a	5.67a	123.28a

¹Means with the same subscript letters are not significantly different at the 5% level of significance.

²Weights (gms) are the mean of 25 birds.

TABLE 3:17

DUNCAN'S COMPARISON FOR ORGAN AND BODY WEIGHT MEANS (GMS)
FOR BIRDS AT 14 WEEKS OF AGE BETWEEN SENCOR
TREATMENTS WITHIN GENERATION II^{1,2}

Treatment (ppm)	Organ					Body wt.
	Liver	Heart	Lung	Kidney	Ovary	
0	5.11b	1.08b	1.21a	1.81a	5.69a	123.51a
37.2	5.96a	1.22ab	1.25a	1.99a	6.12a	128.71a
149	5.50ab	1.11ab	1.33a	2.00a	6.35a	128.37a
372	5.58ab	1.14a	1.23a	1.89a	6.14a	128.35a

¹Means with the same subscript letters are not significantly different at the 5% level of significance.

²Weights (gms) are the mean of 25 birds.

TABLE 3.18

DUNCAN'S COMPARISON FOR ORGAN AND BODY WEIGHT MEANS (GMS)
FOR BIRDS AT 14 WEEKS OF AGE BETWEEN SENCOR
TREATMENTS WITHIN GENERATION III^{1,2}

Treatment (ppm)	Organ					Body wt.
	Liver	Heart	Lung	Kidney	Ovary	
0	5.60b	1.12a	1.16a	1.85a	4.95a	125.80a
37.2	7.26a	1.15a	1.18a	1.89a	5.78a	125.05a
149	6.02b	1.08a	1.24a	1.92a	5.45a	127.41a

¹Means with the same subscript letters are not significantly different at the 5% level of significance.

²Weights (gms) are the mean of 25 birds.

37.2, 149 & 372 ppm) between generation I, II and III, no significant differences were found in egg production (Fig. 3:11, 3:24, 3:25, 3:26, Table 3:14).

IV. (a) Organ and body weight comparisons between Sencor treatments within the three generations of Japanese quail

Table 3:15 presents the F values for the analysis of variance between treatment levels of generation I, II and III for organ and body weights measured at fourteen weeks of age. Tables 3:16 to 3:18 are the mean weights for each organ and body weight in each generation. Tables 3:16 to 3:17 also contain the results of a Duncan's test on the means between the different treatments.

Liver

In generation I results for the treatments were erratic. Figure 3:27 and Table 3:16 demonstrate this point. For instance the control has a significantly larger mean weight than the 37.2 ppm Sencor treatment, yet it has a significantly smaller mean weight than the 149 ppm treatment while showing no difference from the 372 ppm treatment. In effect deviation from a normal pattern seems to be caused by the 37.2 ppm treatment which gave a very low mean liver weight. This pattern changes in generation II and III (Tables 3:17, 3:18) and develops a curvilinear relationship. In both generation II and III there is a slight but significant increase in liver weight from the control to the 37.2 ppm Sencor treatment.

PERCENT HEN-DAY PRODUCTION

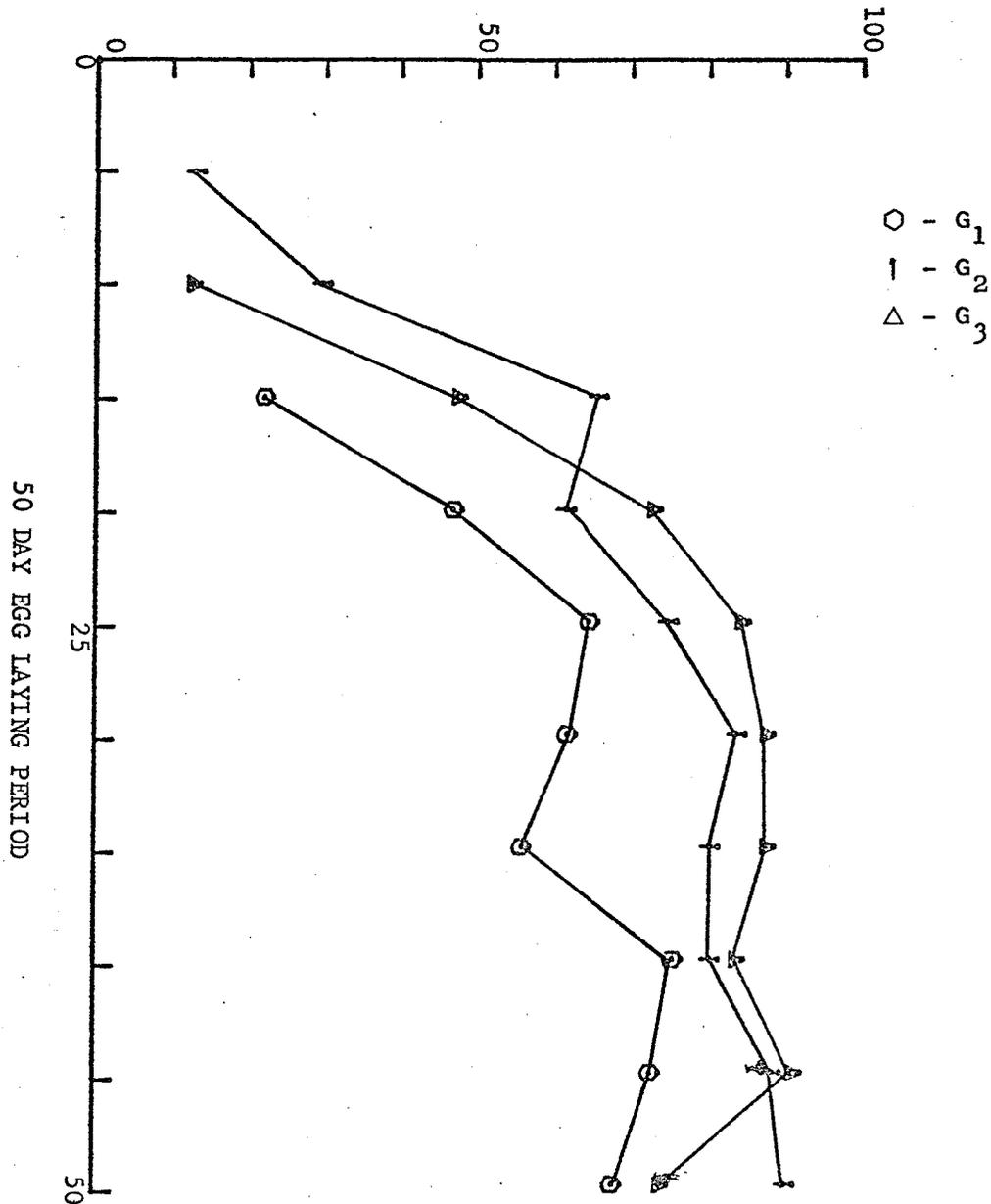


Fig. 3:24.--Egg production of Japanese quail fed Sencor at 37.2 ppm for three generations

PERCENT HEN-DAY PRODUCTION

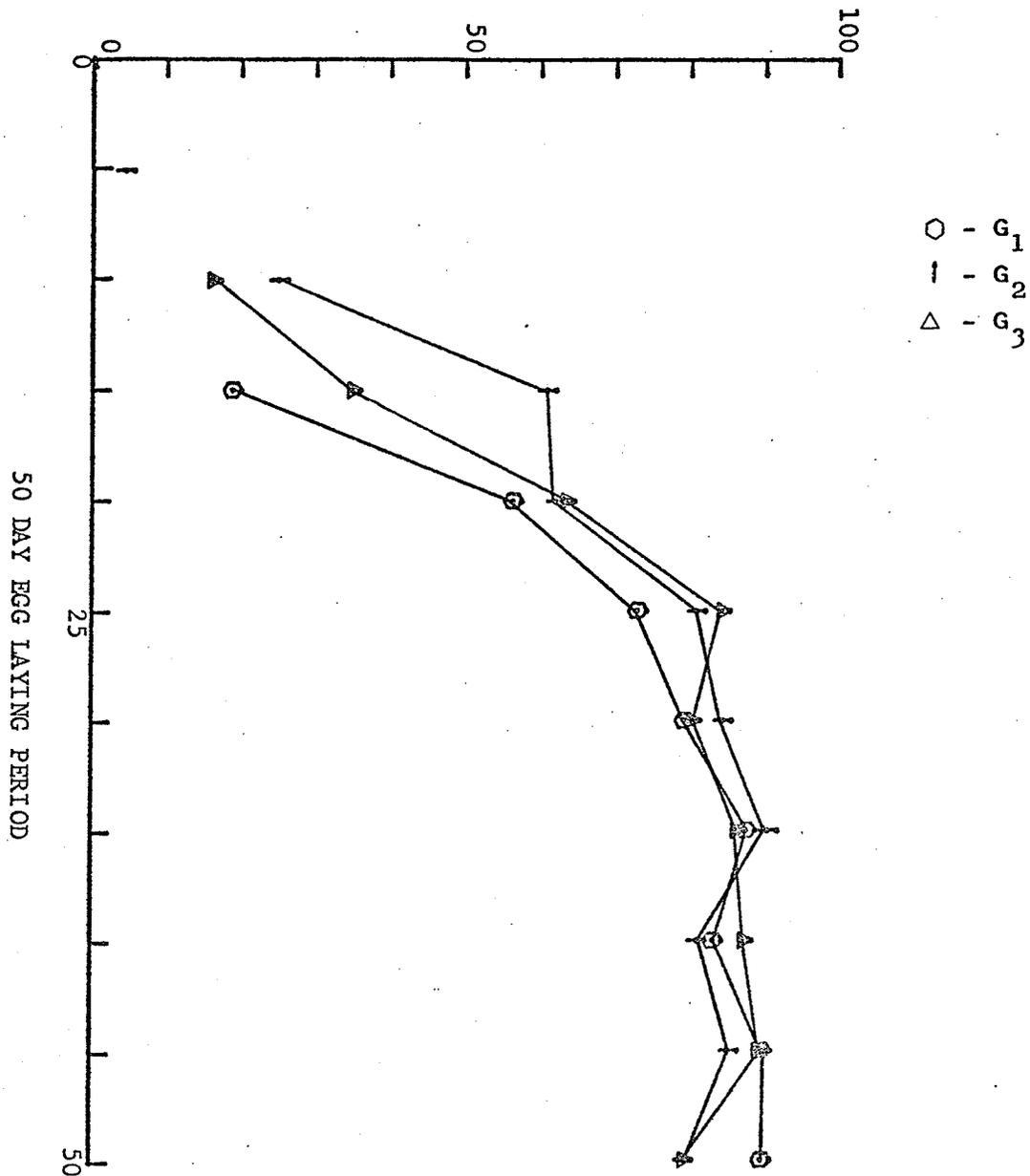


Fig. 3:25.--Egg production of Japanese quail fed Sencor at 149 ppm for three generations

PERCENT HEN-DAY PRODUCTION

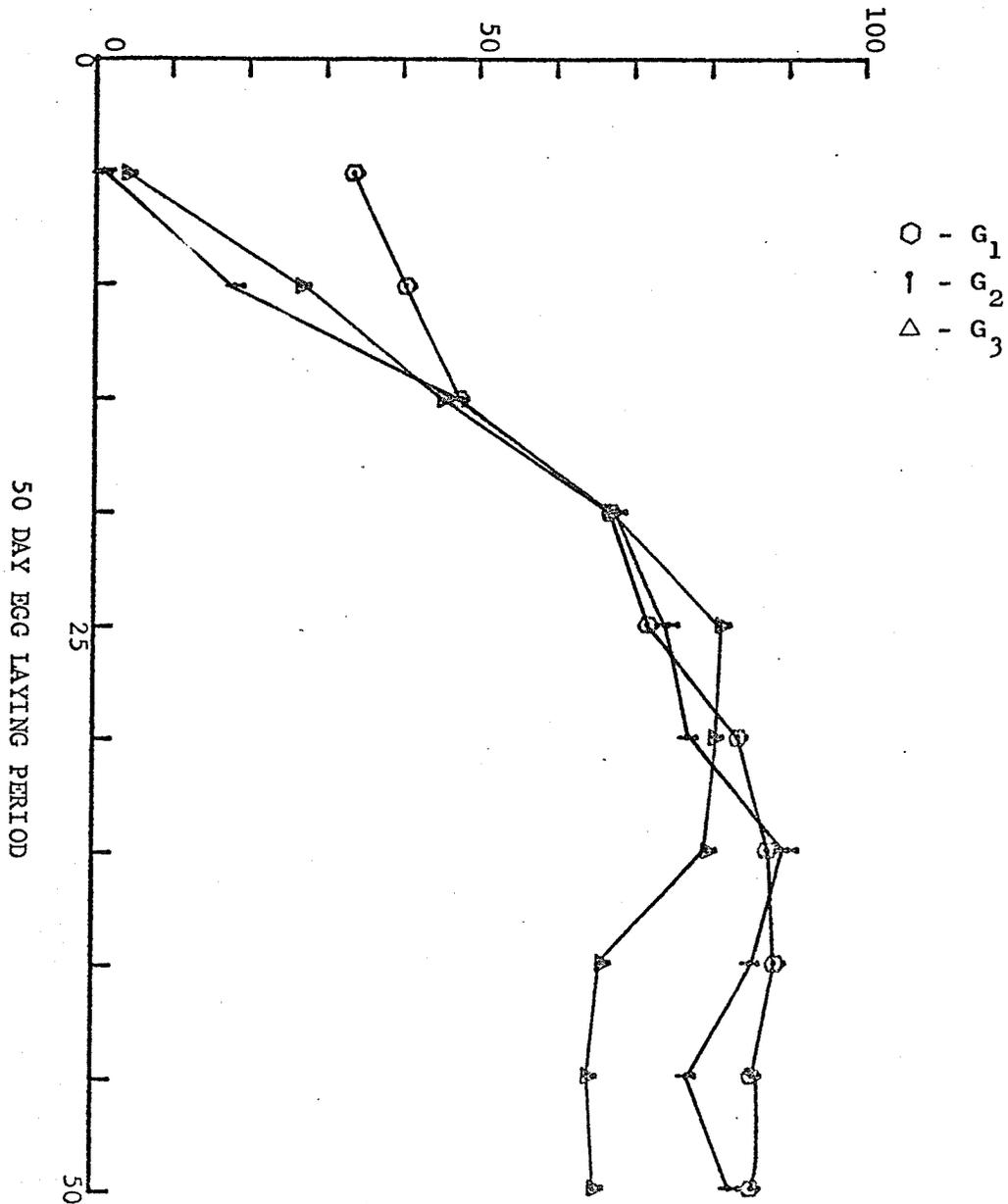


Fig. 3:26.--Egg production of Japanese quail fed Sencor at 372 ppm for three generations

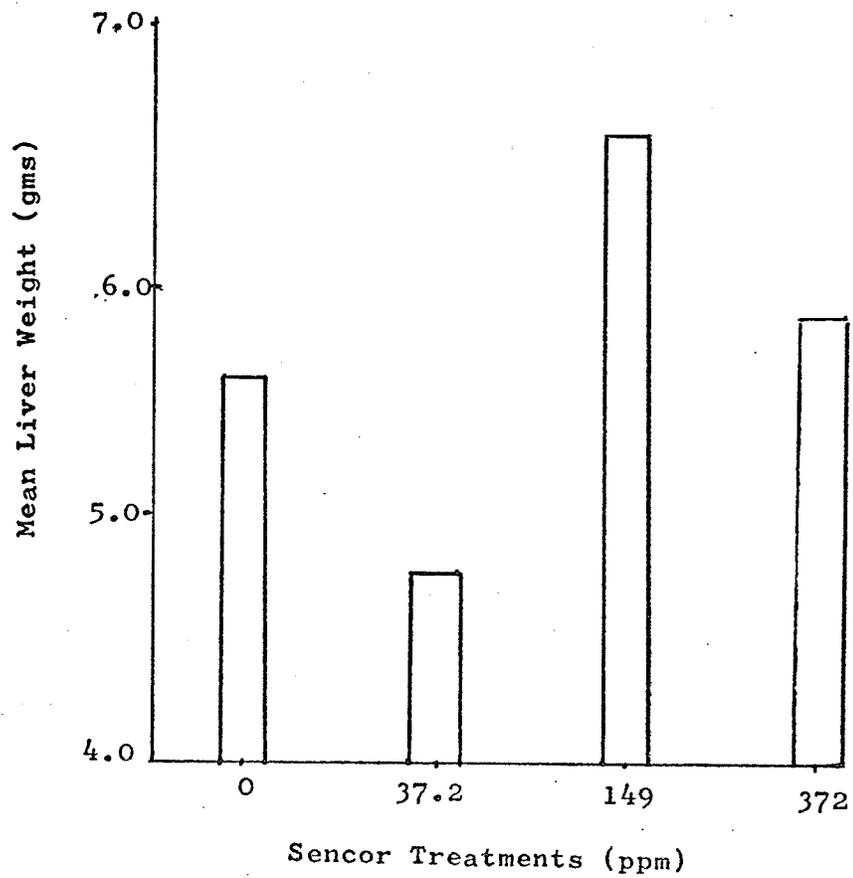


Fig. 3:27.--Mean liver weights at 14 weeks of age for generation I birds treated with Sencor

and a fall in the liver weight as the concentration increases to 149 ppm Sencor. The decrease in mean liver wieght from treatment 37.2 ppm to treatment 149 ppm is significant in generation III but not in generation II (Tables 3:17 to 3:18).

Heart

Of the three generations tested, generation I and II showed significant differences (Table 3:16 and 3:17) while generation III showed no significant differences (Table 3:18). In both generation I and II the 372 ppm treatment produced the largest mean heart weights, of the four treatments (0, 37.2, 149 and 372 ppm Sencor).

Lung

Results showed no significant variation between any treatments in the three generations studied (Tables 3:16 to 3:18).

Kidney

Results of mean weight comparisons showed significance only in generation I (Table 3:16) between treatments of 37.2 and 372 ppm Sencor. As in the liver weights of generation I for Sencor (Table 3:16) no consistent pattern developed for the pesticide effect.

Ovary

Although differences occurred between the ovary weights of the Sencor treatments in generation I (Table 3:16), as in the liver weights there is no consistent trend. For generation

II and III no significant results occurred (Table 3:17 and 3:18).

Body Weight

Of the three generations tested only generation I showed significance (Table 3:16). Here the dietary level 149 ppm had a very low body weight which does not seem normal even for this level of herbicide intake; and as such, can only be treated cautiously as a significant effect. The birds appeared healthy and no reason was found for the lower than normal weight.

(b) Comparison of Internal Organ and Body weight between the three generations of Japanese quail within treatments of Sencor

Table 3:19 is the analysis of variance between the three generations within Sencor treatments for each organ and body weight. Duncan's tests were performed on all significant data from Table 3:19 as shown in Table 3:20 where the organ and body weight means are given.

The 372 ppm treatment in the third generation was not produced and as such a Duncan's test was not required, only a one way analysis of variance, in order to show which generation (I or II) had a lower mean weight. Means and lettering similar to a Duncan's method are provided in Table 3:20.

When comparisons were made between the three generations the most common factor when significance occurred was generation I organs being consistently smaller in weight than those in either

TABLE 3:19

ANALYSIS OF VARIANCE, F VALUES, FOR ORGAN AND BODY WEIGHTS
 AT 14 WEEKS OF AGE BETWEEN GENERATIONS WITHIN
 TREATMENTS FOR SENCOR TREATED JAPANESE QUAIL¹

Treatment (ppm)	Organ					
	Liver	Heart	Lung	Kidney	Ovary	Body wt.
0	2.27	10.32*	4.13*	.24	2.29	.39
37.2	66.21*	10.31*	.96	9.54*	2.27	7.58*
149	6.93*	19.23*	7.96*	4.52*	102.78*	44.61*
372	1.68	4.74*	.84	.63	1.47	4.27*

¹F values with asterisk are significant at the 5% level.

TABLE 3:20

DUNCAN'S COMPARISONS FOR ORGAN AND BODY WEIGHT MEANS (GMS) BETWEEN GENERATIONS WITHIN SENCOR TREATMENTS¹

Organ	Gener- ation	Control	Mean Weight of Organ (gms)		
			37.2 ppm	149 ppm	372 ppm
Liver	I	5.57a	4.75a	6.60b	5.83a
	II	5.10a	5.96a	5.48a	5.68a
	III	5.60a	7.24b	5.98a	
Heart	I	0.92b	0.93b	0.90b	1.10b
	II	0.08a	1.14a	1.11a	1.24a
	III	1.11a	1.15a	1.10a	
Lung	I	1.45b	1.15a	1.09b	1.20a
	II	1.21a	1.23a	1.33a	1.25a
	III	1.17a	1.18a	1.24a	
Kidney	I	1.77a	1.62b	1.75a	1.94a
	II	1.81a	1.93a	2.00ab	2.01a
	III	1.85a	1.89a	1.92b	
Ovary	I	4.72a	5.92a	1.79b	5.66a
	II	5.68a	6.10a	6.35a	6.12a
	III	4.95a	5.45a	5.79a	
Body Weight	I	123.21a	120.96b	105.63b	123.28b
	II	123.51a	128.35a	128.41a	128.75a
	III	125.76a	128.04a	127.41c	

¹Means with the same subscript letter are not significantly different at the 5% level of significance.

generation II or III, while generation II and III were seldom different from each other. The above statement holds true for significant tests on heart (149 ppm), lung (149 ppm), ovary (149 ppm), body weight (37.2 and 372 ppm), kidney (37.2 ppm) and on the heart for the control population (Table 3:20). In three cases of significance this does not hold true. The livers from the 37.2 ppm treatment for generation I and II are significantly smaller than those from generation III. When the kidneys at treatment level 149 ppm were tested generation I was found to be significantly smaller than generation III but not smaller than generation II. The third exception was found for the lung in the control, when it was compared. Here generation II and III were significantly smaller than generation I but they are not significantly different from each other (Table 3:20).

4. DISCUSSION AND CONCLUSION

The study demonstrated that Japanese quail have a high tolerance for Abate and even higher tolerance for Sencor. When the total pesticide consumption is calculated for the experimental period it is apparent that these birds can tolerate intakes that cumulatively are many fold greater than the single acute oral LD₅₀ dose. For instance, the LD₅₀ for Abate is 69.1 mg/kg but birds on the 32 ppm diet consumed a quantity equal to about 10 times a single LD₅₀ dose over the 14 week treatment period with no ill effects. The birds fed Sencor at 372 ppm in their diet consumed 25 times the single acute oral LD₅₀ dose over the 14 week period, again with no ill effects. This suggests that over a 24 hour period the birds were capable of metabolizing and/or excreting the majority of the ingested pesticide which prevented a cumulative effect from occurring.

Data from the literature suggests that excretion of Abate in the rat is about 50% of the intake in 24 hours and 90% in 48 hours at dosages up to 6.2 mg/kg (Blinn 1969). Thus the 32 ppm Abate treatment did not exceed the birds metabolic and/or excretory capacity to protect itself. But the 80 ppm Abate treatment did exceed this metabolism-excretion capacity of the birds since mortalities and decreased egg production occurred.

For Sencor the literature on triazines suggests that the

majority of a dose is excreted in 24 - 72 hours (Bakke et al 1972, Bakke et al 1971, Bakke et al 1967, Larson and Bakke 1971). In the present study 372 ppm was the highest level of Sencor fed to the birds and the intake did not exceed the bird's metabolic excretion capacity to protect itself.

Environmental intakes of Abate and Sencor (8 and 37.2 ppm in the diet, respectively) did not show any long term effects on the birds. Percent hatch, fertility, and weight gain were all unaffected, which is in agreement with work done by Gough et al 1967. In the present study no differences were found in mortality between chicks and adults fed Abate or Sencor unlike Sherman et al 1972 who found using the insecticide S4087 that there was a higher mortality for mature Leghorns than for chicks. In fact, Fig. 3:1 to 3:3 and 3:15 to 3:17 show a consistent and normal weight gain of the Japanese quail from chick to adult. Based on the design of this experiment these two pesticides will not cause any harm to birds in the environment when applied at the rates recommended for pest control. Furthermore, when one considers the natural degradative forces that act upon such chemicals in the natural environment (bacteria, sunlight, water, etc.) the chance of them adversely affecting birds is extremely remote.

In the summer of 1972 air conditioning ducts were installed in the bird room, resulting in extremely warm and noisy conditions for 2 to 3 weeks. This subjected the birds to a secondary stress

factor. The birds fed Abate in generation I were reared at this time and show differences in weight gain and egg production that were not evident in generation II or III birds, which were not subjected to this secondary stress factor. The primary stress factor, Abate, did not affect the birds' weight gain or egg production during the first 11 weeks of the experiment, however, when the secondary stress factor was introduced at the beginning of the twelfth there was a sharp reduction in weight gain and egg production (Table 3:1). Egg production began to recover once the secondary stress was removed but the Abate treated birds recovered at a slower rate than the controls.

Birds on the Sencor treatments were only subjected to the primary pesticide stress. Results for between generations within treatments, indicate a significantly lower weight gain for birds in generation I than in generations II and III on two of the four treatments (37.2 and 149 ppm) (Table 3:12). Thus some adaptation either physically or metabolically to the pesticide Sencor may have taken place by generation II with no further adaptation by generation III.

Egg production is one of the more responsive biological parameters for measuring pesticide effects in birds and changes in this parameter will be discussed in conjunction with changes in ovary weight. The general indication from this experiment is that as the concentration of Abate in the diet increased there was a

decrease in ovary weight and egg production. This concurs with the results of Sherman et al, 1971, Gough et al 1967, Sherman and Herrick 1966, and Shellenberger 1966 and 1965.

The most significant reduction in egg production occurred in generation I and II for the birds fed Abate at 80 ppm. This high level of intake was significantly affecting the female birds. The fertility of the eggs produced in generation I was sufficient to produce generation II but the few eggs produced in generation II were very low in fertility. This can be attributed to the observation that throughout this generation the males experienced great difficulty in copulating with the females. They did not have the proper muscle coordination. Thus this high level of Abate intake also affected the males. As a result there was no third generation of birds produced on the 80 ppm Abate treatment.

The feeding of progressively higher levels of Sencor did not consistently affect ovary weight and/or egg production in the birds.

The birds fed Abate at 0, 8 and 32 ppm experienced a few significant differences in organ and body weights but there was no consistent trends indicating overall biological deterioration. These differences occurred even though the birds were randomly selected in setting up the treatments and 25 birds were used in each treatment group. The birds receiving 80 ppm Abate demonstrated a consistent trend towards lower organ and body weights in each

generation. This clearly demonstrates that at 80 ppm in the diet Abate adversely affects the Japanese quail.

Significant differences occurred between organ and body weights of the Sencor fed birds (0, 37.2, 149 and 372 ppm) in each generation. The differences occurred randomly and do not establish any relationship between level of intake and effect. As occurred with the Abate fed birds, random selection and large sample size (25 birds) did not prevent these differences from occurring.

In conclusion the data indicate that Japanese quail can tolerate the environmental levels of Sencor and Abate that would occur following a pest control application at recommended rates. Some selection or adaptation may have taken place from generation to generation, by physical or metabolic means. For instance there could have been selection for hardier birds. Since there was no significant death rate in any of the adults in generation I of any treatment this could have happened in three possible ways; birds affected produced fewer eggs, eggs of more susceptible birds failed to hatch, or chicks of generation I birds that were susceptible succumbed a few days after hatch and the deaths of these chicks did not show up in the statistics. Any of these events would have subsequently led to a more resistant bird population.

Although Japanese quail can safely tolerate high pesticide intakes 32 ppm Abate and 372 ppm Sencor some effort must be taken to simulate a birds environment, because the laboratory intake that

birds can tolerate may bear no relationship to what they can safely ingest in their natural environment. That is other stresses may compound the effect of the pesticide or the rate of breakdown in the environment may allow the birds to be safely exposed to much higher rates of pesticide application than laboratory work would indicate.

5. SUMMARY

1. Japanese quail can ingest high levels of Abate (32 ppm in the diet) through three generations without any measurable physiological effect. When this level of ingestion was continued over a period of 14 weeks it amounted to a total consumption of 10 times the single acute oral LD₅₀ dose.
2. Japanese quail can ingest high levels of Sencor (372 ppm in the diet) through three generations without any measurable physiological effect. When this level of ingestion was continued over a period of 14 weeks it amounted to a total consumption of 25 times the single acute oral LD₅₀ dose.
3. At an intake rate of 80 ppm Abate in the diet some birds died and others had slower growth rates, lower egg production, decreased fertility and lower organ weight.
4. When Abate and Sencor were fed in the diet from day 1 to week 14 no differences in pesticide effect were noted in immature versus adult birds.
5. Pesticide adaptation may have occurred in the quail from generation I to generation II.
6. When Japanese quail are exposed to pesticides a consistent exposure to secondary stress factors can produce an additive effect upon the birds.
7. When testing pesticides other stress factors should be included in the experiment for meaningful results.

7. APPENDIX I

Hen-day production - total eggs laid divided by total hen-day and expressed as a percentage (North 1972 and Poultry Division 1972).

$$\% \text{ hen-day production} = \frac{\text{Number eggs produced} \times 100}{\text{No. live hens}}$$

6. LITERATURE CITED

- Abbott, Ursula K. and R.M. Craig, 1960. Observations on hatching time in three avian species. *Poultry Sci.* 39:827.
- Angelucci, R., D. Artini, A. Cresseri, P.N. Giralardi, W. Logemann, G. Nannini and G. Volzell, 1965. Some aspects of the metabolism of triazine derivatives active in experimentally induced virus infections. *Brit. J. Pharmacol.* 24:274.
- Bakke, J.E., J.D. Larson, C.E. Price, 1972. Metabolism of Atrazine and 2-Hydroxyatrazine by the rat. *J. Agr. Food Chem.* 20:602.
- Bakke, J.E., J.D. Robbins, and V.J. Feil, 1971. Metabolism of 2-methoxy-4-ethylamino-6-sec-butylamino-s-triazone by the dairy cow and the goat. *J. Agr. Food Chem.* 19:462.
- Bakke, J.E., J.D. Robbins, and V.J. Feil, 1967. Metabolism of 2-Chlor-4-, 6-bis (isopropylamino)-s-(Propazine) and 2-Methoxy-4, 6-bis (isopropylamino)-s-triazine (Prometone) in the rat. Balance study and urinary metabolite separation. *J. Agr. Food Chem.* 15:628.
- Bitman, J., H.C. Cecil, H.C. Helene and G.F. Fries, 1969. DDT induces a decrease in eggshell calcium. *Nature* 224:44.
- Blinn, R.C. 1969. Metabolic fate of Abate, insecticide in the rat. *Jour. Agr. Food Chem.* 17:118.
- Bohme, C. & F. Bar, 1967. Uber den Abbau von Traizin-Herbiciden im tierischen organismus. *Fd. Cosmet. Toxicol.* 5:23.
- Daniels, G.L., 1968. Ovulation and longevity in the Japanese quail (*Coturnix coturnix japonica*) under constant illumination. *Poultry Sci.* 47:1875.
- Dunachie, J.F. and W.V. Fletcher, 1970. The toxicity of certain herbicides to hens eggs assessed by the egg injection technique. *Ann. Appl. Biol.* 66:515.
- Dunachie, J.F., and W.W. Fletcher, 1967. Effects of some herbicides on the hatching rate of hens' eggs. *Nature* 215:1406.

- Goly, F.B., 1960. Energy dynamics of a food chain of an old-field community. *Ecological Monographs*. 30:187.
- Gough, B.J., T.E. Shellenberger and L.A. Escuriex, 1968. Responses of Japanese quail fed purified diets. *Quail Quart.* 5:2.
- Gough, B.J., L.A. Escuriex and T.E. Shellenberger, 1967. A comparative toxicologic study of a phosphorodithioate in Japanese and Bobwhite Quail. *Toxicology & Applied Pharmacology*. 10:12.
- Hickey, J.J. and D.W. Anderson, 1968. Chlorinated hydrocarbons and eggshell changes in raptorial and fish eating birds. *Science* 162:271.
- Howes, J.R., 1964. Hatchery management and disease prevention for Coturnix quail. *Animal Digest*. 3:12.
- Leigh, E., St. John Jr., D.G. Wagna and D.J. Lisk, 1964. Fate of Atrazine, Kuron, Silvex and 2,4,5-T in the Dairy Cow. *Jour. Dairy Sci.* 47:1267-1270.
- Johnson, A.E., K.R. Vankampen and W. Binns, 1972. Effects on cattle and sheep of eating hay treated with the Triazine herbicides Atrazine and Prometone. *Am. J. Vet. Res.* 33:1433.
- Kawahara, T., 1967. Wild Coturnix quail in Japan. *Quail Quart.* 4:62.
- Larson, J.D., and J.E. Bakke, 1971. Metabolism of plant metabolites of s-triazines herbicides in the rat. *Proc. N.D. Acad. Sci.* 24:178.
- Latshaw, J.D., and L.S. Jensen, 1970. Comparison of purified & practical diets for reproduction in Japanese quail. *Poultry Sci.* 49:1599.
- North O. Mack, 1972. *Commercial Chicken Production Manual*. The AVI. Publishing Inc., West Port, Connecticut.
- Odum, E.P., 1959. *Fundamentals of Ecology*, 2nd ed. Philadelphia, Saunders, 1959.
- Oliver, W.H., G.S. Born, P.L. Ziemer, 1969. Retention, distribution and excretion of Ametryne (2-Methylmercaptar-4-ethylamino-6-isopropylomino-s-triazine) in the rat. *J. Agr. Food Chem.* 17:1207.

- Padgett, C.S. and W.D. Ivey, 1959. Coturnix quail as a laboratory research animal. *Science*. 129:267.
- Peakall, D.B., 1970. Pesticides and the reproduction of birds. *Scientific American*. 222:84.
- Porter, R.D. and N.W. Stanley, 1969. Dieldrin and DDT: Effects on sparrow hawk eggshells and reproduction. *Science*. 165:199.
- Poultry Division, Production and Marketing Branch Agricultural Final Report 17th Central Production test, C.D.A. 1972.
- Shellenberger, T.E., 1966. Cholinesterase inhibition and toxicological evaluation of two organophosphate pesticides in Japanese quail. *Toxicology & Applied Pharmacology*. 8:22.
- Shellenberger, T.E., G.W. Newell, R.M. Bridgeman and J. Barbaccia, 1965. A Subacute toxicity study of N-(2-Mercaptoethyl) Benzenesulfonamide S-(OO-Diisopropyl Phosphorodithioate and Phthalimidomethyl-o-o-dimethyl Phosphorodithioate with Japanese quail. *Toxicology and Applied Pharmacology*. 7:550.
- Shellenberger, T.E., 1965. Toxicological evaluations of agricultural chemicals with Japanese quail. *Laboratory Animal Care*. 15:119.
- Sherman, M., and R.B. Herrick, 1972. Chronic toxicity and fly control from feeding S-4087 o-p-Cyanophenyl O-Ethyl Phenylphosphonothioate) to laying hens. *Poultry Sci*. 5:1064.
- Sherman, M., E. Ross, and J.R. Yate III, 1971. Comparative toxicity of four halogenated organophosphorous insecticides to chicks, Japanese quail and Diptera. *Jour. of Economic Ent.* 64:814.
- Sherman, M., E. Ross and M.T.Y. Chang, 1964. Acute and subacute toxicity of several organophorous insecticides to chicks. *Toxicology and Applied Pharmacology*. 6:147.
- Sherman, M. and R.B. Herrick, 1966. Acute and subacute toxicity of Apholate to the chick and Japanese quail. *Toxicology and Applied Pharmacology*. 9:279.
- Smith, S.E., C.W. Weber and B.L. Reid, 1969. The effect of high levels of dietary DDT on egg production, mortality, fertility, hatchability and pesticide content of yolks in Japanese quail. *Poultry Sci*. 48:1000.

- Steel, R.G.D. and J.H. Torrie, 1960. Principles and Procedures of Statistics. McGraw-Hill Book Company Inc., New York.
- Tucker, R.K., M.A. Haegele and M.A. Huegele, 1970. Comparative acute oral toxicity of pesticides to six species of birds. Toxicology and Applied Pharmacology. 20:57.
- Vagt, H. and L. Steinke, 1971. Observations on the effect of sexual relationships and age on fertility and hatchability in Japanese quail. Arch. for Geflugelkunde. 34:1.
- Vohra, P., M.J. Davis and R.M. Craig, 1970. The improvement of hatchability of coturnix (Japanese quail) eggs by EDTA. Poultry Sci. 49:780.
- Walker, A.I.T., C.H. Neill, 1969. The toxicity of Dieldrin (HEOD) to Japanese quail (Coturnix, coturnix, japonica). Toxicology and Applied Pharmacology. 15:69.
- Wetherbee, David Kenneth, 1961. Investigation in the life history of the common Coturnix. Am. Midland Naturalists. 65:168.
- Wilson, W.O., K. Ursula and Abbott & Hans Abplanalp, 1961. Evaluation of Coturnix (Japanese quail) as pilot animal for poultry. Poultry Sci. 40:651.
- Woodard, A.E., and H. Abplanalp, 1971. Longevity and reproduction in Japanese quail maintained under stimulatory lighting. Poultry Sci. 50:688.
- Woodard, A.E. and H. Abplanalp 1967. The effect of mating ratio on fertility and hatchability in Japanese quail. Poultry Sci. 46:383.
- Yo, W.C. and J.C. Gilbreath, 1971. The effects of Ronidzile on reproductive performance of Japanese quail. Poultry Sci. 50:977.
- Zins, G.R., 1965. The in vivo production of a potent, long acting hypotensive metabolite from diallylmelamine. J. Pharmacol. Exp. Therap. 150:109.