

THE EFFECTS OF TWO CARBAMATE INSECTICIDES  
ON BOBWHITE (COLINUS VIRGINIANUS) BIOENERGETICS

by

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## INTRODUCTION

This study was a continuation of investigations at Kansas State University on bobwhite (Colinus virginianus) bioenergetics. Previous studies have involved bobwhite energy requirements, metabolizable energy in natural foods, and quail habitat improvement in Kansas. Investigations have recently been directed toward the effects of insecticides on bobwhite behavior and bioenergetics. Although numerous insecticidal studies have involved avian behavior and reproduction, few have looked at insecticidal effects on the bioenergetics of the test animal.

Carbamate insecticides were chosen for this energetic study since they are becoming increasingly popular as replacements for the more persistent organochlorine insecticides. Through 1971 and 1972, 287,471 acres in Kansas were treated with 438,525 pounds of the carbamate, Sevin® (Kansas State Board of Agriculture 1971 and 1972). Through these years, 97,957 acres were treated with 75,735 pounds of Furadan®, another carbamate. The worldwide use of Sevin® has increased from 1 million pounds in 1959 to 25 million pounds in 1970. The U. S. use of the organochlorine, DDT, from 1959 (80 million pounds) to 1970 decreased by 2/3 (Frear 1972). The use of DDT was restricted by the Environmental Protection Agency in December, 1972.

The purpose of my study was to determine the influences of two insecticidal carbamates on bobwhite bioenergetic variables: gross energy intake, metabolized energy, excretory energy, efficiency of metabolism, and fecal energy.

## LITERATURE REVIEW

### Sevin® and Furadan®

The first insecticidal carbamates (n-dimethylcarbamates) were developed in Switzerland in 1947 (O'Brien 1967). Sevin® (1-naphthyl-n-methylcarbamate), the first widely used insecticidal carbamate, was introduced by Union Carbide, New York, in 1958 (Nisbet 1971). Sevin® can be described as a broad spectrum, quick knock-down, rapidly metabolized, nonphytotoxic insecticide of low avian and mammalian toxicity. Furadan® (2,2-dimethyl-2, 3-dihydrobenzofuranyl-7-n-methylcarbamate), a highly toxic carbamate, was introduced by Niagara Chemical Company, New York, about 1968.

The literature on Sevin® (Sevin) and Furadan® (Furadan) consists of three types: metabolism and residues, toxicity, and effects on reproduction.

#### Metabolism and Residues

Studies of domestic mammals and domestic fowl indicate that Sevin is rapidly metabolized to water soluble conjugates and excreted through the urine. Paulson and Feil (1969) dosed white leghorn hens (Gallus domesticus) orally with 10 mg/kg radio-labeled Sevin. The labeling was done as both ring-<sup>14</sup>C and carbonyl-<sup>14</sup>C. The hens were surgically modified in order to collect feces and urine. Via the urine, 94.1 percent of the ring-<sup>14</sup>C and 32.2 percent of the carbonyl-<sup>14</sup>C was excreted. Radioactive metabolites were excreted most rapidly during the first 6 hours after dosing. The major portion (50 percent) of the carbonyl-<sup>14</sup>C was eliminated via respiration. No ring-<sup>14</sup>C Sevin was eliminated through respiration. Relatively minor amounts of both ring-<sup>14</sup>C (8.0 percent) and carbonyl-<sup>14</sup>C (6.4 percent) were excreted in the feces. Paulson and Feil also noted that food and water intake of the hens remained normal after the insecticide was administered.



Dorough (1967) dosed lactating cows (Bos taurus) with 0.25 and 3.05 mg/kg radio-labeled Sevin. With the 0.25 and 3.05 mg/kg doses, 70 percent and 58 percent of the doses, respectively, were excreted through the urine. Eleven percent and 15 percent of the respective doses were excreted via the feces. Trace amounts of unaltered Sevin and its hydrolysis product, 1-naphthol, were found in the urine. Of the total radioactive metabolites measured in the urine, 95 percent were excreted during the first 24 hours after dosage. Small amounts of radioactivity in the urine were noted through 120 hours.

Furadan is also known to be metabolized to water soluble conjugates and excreted through the urine. Furadan is more slowly metabolized (hydrolyzed) than Sevin. Hicks et al. (1970), using carbonyl-<sup>14</sup>C Furadan with white leghorn hens, noted that 54 percent of the dose was hydrolyzed after 6 hours, 72 percent after 24 hours, 77 percent after 4 days and 80 percent after 10 days. All body tissues contained residues at 6 and 24 hours. Residues in the liver and kidney were 2.6 ppm at 6 hours but declined to 0.2 ppm at 24 hours. Residues were found in the eggs also.

Ivie and Dorough (1968) dosed lactating cows with radio-labeled Furadan. Of the total radioactive metabolites in the urine, 94 percent was detected within 72 hours. The symptoms of salivation, tearing, hyperactivity, loss of balance, and diarrhea were most severe at 2 hours, but at 4 hours only slight loss of balance was noted. Approximately 0.2 percent of the Furadan dose was found in the milk. Metcalf et al. (1968) found that mice eliminated 50 percent of the Furadan metabolites in 24 hours.

#### Toxicity

No studies were found that determined the acute oral toxicity of Sevin to bobwhites. Heath et al. (1972) determined the LC<sub>50</sub> of Sevin for bobwhites

to be greater than 5,000 ppm in the feed for 5 days. Tucker and Crabtree (1970) reported that the acute toxicity ( $LD_{50}$ ) of Sevin for mallards (Anas platyrhynchos) was 2179 mg/kg, pheasants (Phasianus colchicus) 2000 mg/kg, coturnix quail (Coturnix coturnix japonica) 2290 mg/kg, and for sharp-tailed grouse (Pedioecetes phasianellus) 780-1700 mg/kg. Tucker and Crabtree also noted that Sevin was a relatively fast-acting chemical. Tucker and Haegelle (1971) determined the acute toxicity of four carbamates, zectran, mobam, Baygon®, and landrin, to mallards, pheasants, and chukar partridge (Alectoris graeca).

The Furadan acute  $LD_{50}$  for bobwhites as determined by Tucker and Crabtree (1970) was 5.04 mg/kg. The  $LD_{50}$  for mallards, pheasants, and fulvous tree ducks (Dendrocygna bicolor) were 0.397 mg/kg, 4.15 mg/kg, and 0.238 mg/kg, respectively. Furadan was noted to have little or no cumulative toxicity. Sherman et al. (1967) reported the acute  $LD_{50}$  of white leghorn chicks as 6.3 (5.5-7.2) mg/kg Furadan.

Sherman and Ross (1969) conducted both chronic and acute Furadan toxicity studies with coturnix. Sexual differences in susceptibility were not noted in acute tests (males 1.9 mg/kg, and females 1.7 mg/kg), while the male was more susceptible to poisoning than females in chronic studies. Growth depression was noted the first 2 weeks with 200 ppm Furadan in the feed.

#### Reproduction

Collins et al. (1971) studied rats (Rattus rattus) and gerbils (Meviones unguiculatus) given 2,000 to 10,000 ppm Sevin in the feed for three generations. Dose-related decreases were noted for the number of live young born, litter size, number of young weaned, and survival of young during the first 4 days. The doses used were extremely high, and the animals were fed the treated feed over long periods.

Weil et al. (1972) have summarized numerous reports on the reproductive effects of Sevin. The many conflicting reports, as to what dosage levels cause certain reproductive effects, are due to authors using different dosing methods. Weil et al. cited only two studies concerning game birds, a 1961 study by J. B. Dewitt and C. M. Menzie with bobwhites and pheasants, and Khera (1966) with duck embryos. The former study was never published. Khera found congenital foot deformities of duck embryos when Sevin was injected into the eggs.

The Fish and Wildlife Service (1960) reported that 1000 ppm and 100 ppm Sevin in the diets of bobwhites and pheasants, respectively, would cause a 25 percent decrease in reproductive rates. Little detailed data were reported. Reproduction rates were measured as the number of viable chicks produced. Haegelle and Tucker (1974) reported temporary egg shell thinning when coturnix quail were given an oral dose of 1000 mg/kg Sevin. No appreciable egg shell thinning was noted in eggs of mallards dosed with 1000 mg/kg. The authors stated that the shell thinning was probably caused by the temporary reduction in food consumption by treated quail. Egg shell thinning was observed in eggs from coturnix starved for 36 hours and receiving no insecticidal dosing.

Sherman and Ross (1969) found 200 ppm Furadan in coturnix feed decreased egg production, fertility, and hatchability. There were no abnormal embryos that failed to hatch or abnormal hatched chicks.

#### Insecticide-Bioenergetics

The metabolic requirements of an animal may be determined by measuring its oxygen consumption ( and/or CO<sub>2</sub> production) or by measuring its food energy intake and energy excreted. The oxygen consumption method measures the animal's basal metabolic rate, and is conducted when the animal is at rest, in the dark, and in a postabsorptive state. The food consumption method allows

measurement of metabolized energy, existence energy, efficiency of metabolism, food energy intake, and excretory energy. The food consumption method is conducted on unconstrained animals under conditions of ad libitum food and water, and a natural photoperiod.

Insecticides can cause oxygen consumption changes in treated animals. Lindane, DDT, toxephene, methoxychlor, and DNOC cause increased oxygen consumption in test animals, while arsenicals and rotenone decrease oxygen consumption (O'Brien 1967). The increase of oxygen consumption usually peaked 1-2 hours after animals were dosed.

Lustick et al. (1972) used the oxygen consumption method to study the effect of DDT on bobwhite basal metabolism. The quail feed was treated with 10, 50, 100, and 150 ppm of DDT. The metabolic rates for the treated quail were higher than that of the controls. The metabolic rates did not increase with increased DDT levels. DDT also decreased the quails' thermoneutral zone and upper critical temperature. Lustick et al. stated that the increased oxygen consumption was due to increased thyroid activity and not just an increase of nervous activity.

No comprehensive insecticide-energetic studies were found that measured food intake, energy excreted, and metabolized energy. Insecticide toxicity and reproductive studies have noted changes in the food intake of the test animals. Cope (1971:345) stated that, for birds, "Several investigators have shown reduced egg production after the feeding of pesticides, but lowered food consumption and not the pesticide have been held responsible for the reductions."

Azevedo et al. (1965) fed pheasants DDT treated feed (10, 100, and 500 ppm). The controls consumed 61 grams of feed per day while the feed consumption of birds, on DDT treated feed, decreased 10-13 grams. Pheasant egg production was directly related to this decrease. Linduska and Springer (1957) showed, in

one study, that bobwhites consumed DDT treated feed as readily as nontreated feed. In another study, DDT caused a decrease in bobwhite food consumption. Linduska and Springer also found that chloradane, parathion, and toxephene decreased food consumption of quail, but this was due to the "repellent effect" (1957:3) of the treated feeds.

Atkins and Linder (1967) dosed pheasants weekly with dieldrin (0, 2, 4, and 6 mg/kg). The 4 and 6 mg/kg dieldrin dosed pheasants showed a significant 2.9 g and 64.4 g, respectively, decrease in food intake. Baxter et al. (1969) dosed the second generation pheasants (of Atkins and Linder's pheasants) with 6 and 12 mg/kg dieldrin and found that food intake decreased between 3.75 and 12.64 grams per day. Even the second generation pheasants not dosed with dieldrin (parents had received 6 mg/kg) showed a lower food consumption than the controls.

Sevin and Furadan have been shown to reduce the food intake of birds. Nir et al. (1966) dosed white leghorn hens daily with Opigal (a powder of 50 percent Sevin) for 60 days. Doses of 180 mg/kg and 540 mg/kg reduced food intake by 10 g and 30 g, respectively (controls consumed 65 g/day). Sevin given to white leghorns at 250 ppm and 500 ppm in the feed caused no changes in mean food consumption (Lillie 1973). Sherman and Ross (1969) gave Furadan to Japanese quail at 50, 100, 200, 400, and 800 ppm in the feed. All treatment levels decreased the food intake of quail. No studies were found concerning the palatability of Furadan in feed.

No studies were found concerning insecticidal effects on efficiency of metabolism. The efficiency of metabolism is somewhat considered when animal food conversions (food consumed/unit weight gain) are discussed. Sherman and Ross (1969) treated coturnix with Furadan and found conversion factors of 4.35 for the controls and 7.07 for the treated quail. McDonald et al. (1964) food conversions of 3.95 and 4.04 for controls and malathion fed white leghorns, respectively.

## MATERIALS AND METHODS

### Bird Care

Male bobwhites were obtained from the Kansas Forestry, Fish and Game Commission's quail farm, Pittsburg, Kansas. Each quail was leg banded and placed in a 48 X 25 X 13 cm polypropylene cage. Cages had bottoms and sliding tops of 0.6 or 1.3 cm hardware cloth. Treatment groups were shelved vertically in a walk-in environmental chamber to expose each group to the same vertical distribution of light.

Caged birds were provided ad libitum food and water. The feed was a balanced mash (P-18) prepared by the Department of Grain Science and Industry at Kansas State University (Table 1). To provide uniform particle size, the food was passed through a 7 mesh (2.0 mm openings) screen, then collected in a 20 mesh (0.833 mm openings) screen. This both minimized selection of particle size by birds and aided separation of split food and feces. Quail were not provided grit since Bisset (1972) showed no significant differences in any energetic variable when quail were or were not given grit with a P-18 diet.

Two weeks after being placed in the environmental chamber, quail were weighed every two days to the nearest 0.1 gram. All weighings and feedings began 0.5 hour before the photoperiod (light) began. All food and water containers were removed before the weighing began. When the majority of birds became weight-stable (less than 1.0 percent weight change between weighings) the insecticide-energetic studies began.

### Insecticidal Doses

The insecticidal dosages were prepared with technical grade

Table 1. Feed formulation of P-18 standard mash.

INGREDIENTS <sup>1</sup>	PERCENT	CUMULATIVE PERCENT
Soybean meal	26.4	26.4
Corn	30.0	56.4
Sorghum grain	30.0	86.4
Alfalfa meal	5.0	91.4
Distillers solubles	2.0	93.4
Fish meal	3.0	96.4
Dicalcium phosphate	1.0	97.4
Ground limestone	1.5	98.9
Salt	0.5	99.4
Vitamin A (10,000 IU/g)		
Vitamin D <sub>3</sub> (15,000 IU/g)		
Vitamin B <sub>12</sub> (20 mg/lb)		
Methionine		
Baciferin 10		
		100.0

<sup>1</sup> Expressed as a percent on a weight basis.

Sevin<sup>1</sup> or Furadan<sup>2</sup> (power forms) and corn oil. Corn oil was used as a carrier since these insecticides were not water soluble. When mixed with corn oil, the insecticides formed a temporary suspension. The corn oil-insecticide suspension was administered directly into the quail crop, via the esophagus, with Plastipak 1 cc Disposable Syringes. No regurgitation of the suspension occurred following the dosage.

#### Sevin® LD<sub>50</sub> Study

To determine the LD<sub>50</sub> of Sevin for bobwhites, nine 4-bird groups were dosed once with a 1.0 ml corn oil-Sevin suspension (Table 2). The mean weight of a quail group was used to calculate and prepare that group's insecticidal dose. The observation period, after a group was dosed, was 4 days. The insecticidal symptoms, and the dosage levels at which they occurred, were recorded. A simple linear regression (Snedecor and Cochran 1972), using probits versus log doses, determined the LD<sub>50</sub>.

#### Bioenergetic Data Collection

For the Sevin study, 52 adult (approximately 1 year of age), male quail were randomly assigned to four treatment groups (Fig. 1) and held under constant environmental conditions: 25 C temperature, 14.5L:9.5D photoperiod, and 65 percent relative humidity. These conditions were yearly June, July, and August means for eastern Kansas (personal communication, Dr. L. D. Bark, Kansas State University Meteorologist). These summer conditions were used since Sevin may be applied to the crop any time during the growing season, whenever pest infestation occurs. It was estimated that bobwhites in the wild could

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<sup>1</sup>Union Carbide Corporation, 270 Park Avenue, New York 17, New York.

<sup>2</sup>Niagara Chemical Division, FMC Corporation, Middleport, New York.



Figure 1. Experimental designs used for the Sevin and the Furadan bobwhite energetic studies.  
Each period was two days.

SEVIN					FURADAN				
	Corn Oil Control	Sevin Low	Sevin Medium	Sevin High	Water Control	Corn Oil Control	Furadan Low	Furadan Medium	Furadan High
Period <sup>1</sup>	(4) <sup>1</sup> not dosed (16)		not dosed (16)		(4) <sup>1</sup> not dosed (4)		(16)	not dosed (16)	
Period <sup>2</sup>	1.0 ml corn oil (4)	10 mg/kg Sevin in corn oil (16)	50 mg/kg Sevin in corn oil (16)	90 mg/kg Sevin in corn oil (16)	0.5 ml water (4)	0.5 ml corn oil (4)	.25 mg/kg Furadan in corn oil (16)	1.13 mg/kg Furadan in corn oil (16)	2.0 mg/kg Furadan in corn oil (16)
Period <sup>2</sup>	1.0 ml corn oil (4)	10 mg/kg Sevin in corn oil (12)	50 mg/kg Sevin in corn oil (12)	90 mg/kg Sevin in corn oil (12)	0.5 ml water (4)	0.5 ml corn oil (4)	.25 mg/kg Furadan in corn oil (12)	1.13 mg/kg Furadan in corn oil (12)	2.0 mg/kg Furadan in corn oil (12)
Period <sup>2</sup>	1.0 ml corn oil (4)	10 mg/kg Sevin in corn oil (8)	50 mg/kg Sevin in corn oil (8)	90 mg/kg Sevin in corn oil (8)	0.5 ml water (4)	0.5 ml corn oil (4)	.25 mg/kg Furadan in corn oil (8)	1.13 mg/kg Furadan in corn oil (8)	2.0 mg/kg Furadan in corn oil (8)
Period <sup>2</sup>	1.0 ml corn oil (4)	10 mg/kg Sevin in corn oil (4)	50 mg/kg Sevin in corn oil (4)	90 mg/kg Sevin in corn oil (4)	0.5 ml water (4)	0.5 ml corn oil (4)	.25 mg/kg Furadan in corn oil (4)	1.13 mg/kg Furadan in corn oil (4)	2.0 mg/kg Furadan in corn oil (4)

<sup>1</sup> Sample size shown parenthetically.

<sup>2</sup> Four quail from each insecticide group were killed for cholinesterase determinations.

consume 6.1 mg/kg Sevin a day based on the recommended field application rate of 2 lb/acre, and the surface area (possible contact with Sevin) of the vegetable matter a quail could consume a day. The dosage levels of this study were 10, 50, 90 mg/kg Sevin. The 10 mg/kg level was chosen to approximate the 6.1 mg/kg estimate. It may be possible for a quail to consume 50 mg/kg a day, but it is doubtful that 90 mg/kg a day could be consumed in the field. One milliliter of the corn oil-Sevin suspension was used for each dose.

For the Furadan study, 56 subadult (8 months of age), male quail were randomly assigned to five treatment groups (Fig. 1) and held under constant environmental conditions: 20 C temperature, 14.0L:10.0D photoperiod, and 50 percent relative humidity. These conditions simulated spring in eastern Kansas (April, May, June), since Niagara (1973) and Brooks and Gates (1973) recommend that Furadan be applied to croplands at time of planting. It was estimated that a quail could consume 4.6 mg/kg Furadan a day based on the recommended application rate of 0.75 lb/acre, and the surface area of the vegetable matter consumed by a quail. The study dosage levels (0.25, 1.13, and 2.0 mg/kg Furadan) were relatively low compared to the 4.6 mg/kg estimate, because the Furadan LD<sub>50</sub> to bobwhites is 5.04 mg/kg (Tucker and Crabtree 1970). One half milliliter of the corn oil-Furadan suspension was used for each dose.

The addition of the water control allowed the determination of effects of corn oil on energetic variables. From these data, an estimate was made of the efficiency of corn oil metabolism by bobwhites.

The Sevin and Furadan studies were conducted for five 2-day periods each (Fig. 1). The quail were not dosed during period 1, but were dosed at the beginning of each subsequent period.

At the beginning of each period, all quail were weighed, dosed, placed in clean cages, and then given new feed. This procedure was accomplished

within 0.5 hours before and 2.0 hours after the start of the photoperiod. Gross separation of the spilt feed and feces in the quail cages was accomplished within 12 hours after cages were removed from the chamber. The amount of feed consumed was recorded and the feces was collected. Two feed samples each were collected for caloric and moisture content determinations during periods 1, 3, and 5. Percent dry weight and caloric content of feed was used to calculate gross energy intake. Fecal samples and feed samples were stored at -20 C in labeled glass vials. Fecal samples were later dried at 65 C in open petri dishes for 14 hours, after which the remaining spilt feed was removed by sifting the sample with the same screens used earlier for the feed. Feed and feces remaining in the second screen (20 mesh) were separated by hand. The fecal sample was then dried (65 C) to constant weight (3.5 days). Dried samples were weighed then ground in a Wiley Model micro mill using a 20 mesh (0.51 mm openings) screen.

Caloric measurements of the ground samples were performed in a Parr Series 1200 adiabatic calorimeter, using a Parr 1101 oxygen bomb under 30 atmospheres oxygen. Approximately 30 percent of the fecal samples, of the Sevin and Furadan studies, were replicated to determine the consistency of the bombing method.

The energetic variables determined for each period were:

(1) Gross energy intake (kcal/bird/day):

$$\frac{\text{dry weight of food ingested per period} \times \text{kcal per gram food} \times \text{kcal of corn oil}}{2 \text{ days per period}} =$$

(2) Excretory energy (kcal/bird/day):

$$\frac{\text{dry weight feces per period} \times \text{kcal per gram feces}}{2 \text{ days per period}} =$$

This variable consists of the unabsorbed food energy from the large intestine and the unmetabolized energy excreted by the kidneys (Kendigh 1949).

(3) Metabolized energy (kcal/bird/day):

$$\text{gross energy} - \text{excretory energy} =$$

This is existence energy when the animal studied remains weight stable.

(4) Fecal energy (kcal/gram feces).

(5) Efficiency of metabolism (percent):

$$\frac{\text{metabolized energy}}{\text{gross energy}} \times 100 =$$

### Bioenergetic Data Analysis

Data were analyzed on an IBM S/370 Model 158 computer using a two-way analysis of variance ( $\alpha = .05$ ) with unequal subclasses. Unequal subclasses were used because data from quail gaining weight could not be used for energetic determinations.

For those quail losing weight, Clement (1970) determined a correction factor of 0.356 kcal/g weight loss, to be subtracted from the excretory energy. The 0.356 kcal/g weight loss was used in the insecticide studies to correct excretory energy, metabolized energy and efficiency of metabolism.

The two-way analysis of variance was used for quail during periods 1-5, 1-4, and 1-3. Data analysis of periods 1-5 used a sample size of four quail per group, while period 1-4 and 1-3 analysis were based on a sample size of eight and twelve birds per group. In an attempt to compromise the number of periods and sample size I decided to present all data based on periods 1-4 (sample size of eight quail per group).

When presenting the results of each energetic variable measured, two figures are discussed. The first figure is the actual kilocalories measured,

periods 1 through 4 (example, Fig. 5a). To eliminate period 1 differences between treatment groups, all data were changed to ratios. Gross Energy example:

$$\frac{\text{GROSS ENERGY}}{\text{GROSS ENERGY}} \frac{\text{bird 102, period 2, 3, or 4}}{\text{bird 102, period 1}} = \text{RATIO} \quad \text{Bird 102, period 2, 3, or 4}$$

The same method was used for metabolized energy, excretory energy, efficiency of metabolism, and fecal energy for each quail during each period. A two-way analysis of variance was performed on these ratio data ( $\alpha = .05$ ). Since the corn oil control quail experienced changes in energetic variables, it was desirable to subtract the corn oil effects from the corn oil-insecticide effects:

$$\left( \begin{array}{c} \text{GROSS ENERGY} \\ \text{TREATMENT} \\ \text{RATIO MEAN} \\ \text{period 2, 3, or 4} \end{array} - \begin{array}{c} \text{GROSS ENERGY} \\ \text{CONTROL} \\ \text{RATIO MEAN} \\ \text{period 2, 3, or 4} \end{array} \right) \text{GROSS ENERGY} \text{ period 1 mean} =$$

This subtraction was multiplied by the period 1 mean so to give kilocalorie results. This is the second figure presented for each energetic variable.

#### Cholinesterase Determinations

Four quail were sacrificed from each insecticidal treatment group 48 hours after the first, second, third, and fourth dosings. Birds were killed by squeezing the thoracic cavity. All birds were frozen at -20 C until the brains could be removed and analyzed.

Cholinesterase determination followed that described by Nusz (1971) and Howard (unpublished data). Each whole brain was homogenized in 6 ml Triton X-100 solution (1.0 percent) with a 15 ml all glass tissue grinder. The homogenate was then centrifuged in a Sorvall RC2-B refrigerated centrifuge for 15 minutes at 6,000 g (0-3°C). The supernatant was then spun for 45 minutes at

16,000 g, and the resulting supernatant at 20,000 g for 30 minutes. The final supernatant was analyzed with a Sigma Chemical Kit #420<sup>3</sup> (plasma analysis). Colorimetrically the difference between the optical density readings for a deactivated homogenate sample and an active sample was used with a predetermined calibration curve to determine the cholinesterase level. The curve plotted optical density differences against Rappaport units of cholinesterase. One Rappaport unit of cholinesterase will hydrolyze one micro Mole of acetylcholine in 30 minutes at 25 C and pH of 7.8. Spectral absorption was measured with a Baush and Lomb Spectronic 20 at a wavelength of 435 mu.

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<sup>3</sup>Sigma Laboratory, St. Louis, Missouri

## RESULTS

### Sevin® LD<sub>50</sub>

Symptoms of the heavily dosed (870 mg/kg - 1348 mg/kg) quail were inactivity, inability to stand, head lowered, diarrhea, salivation, lachrimation, gasping, and wing and tail tremors. Quail treated with 147, 250, and 500 mg/kg Sevin® were inactive and their feathers were roughed. Two of the 500 mg/kg quail were the first birds to show difficulty standing. Salivation and diarrhea were first noted with the 870 mg/kg quail. Regurgitation of the corn oil, lachrimation, and oily feces were first noted at 1225 mg/kg Sevin. Death first occurred with the 1057 mg/kg Sevin treatment. The heaviest bird in each of the 1057, 1140, 1225, and 1348 mg/kg groups showed the symptom of inactivity only. All symptoms, except inactivity, dissipated within 24 hours. The corn oil control birds showed none of the above symptoms.

The probit-log dose regression included the 870 mg/kg and all higher dosed groups. The regression equation (Fig. 2) was significant ( $P = .0169$ ). The Sevin LD<sub>50</sub> for bobwhites used in this study was 1193 mg/kg. The 95 percent confidence interval was 1077-1322 mg/kg.

### Sevin® Energetics

The mean feed and corn oil caloric values were 4.350 kcal/g and 9.049 kcal/ml, respectively (Table 4, appendix). The feed percent dry weight used in gross energy determinations was 91.5 percent.

During the study 192 fecal samples were collected. Two oxygen bombings were preformed on each of 60 samples. The mean difference between duplications was 0.547 percent.

Table 2. Sevin ID<sub>50</sub> data. Only the 870-1348 mg/kg data were used for the ID<sub>50</sub> determination.

SINGLE CRAL DOSE (mg/kg) <sup>1</sup>	LOG DOSE	QUAIL WEIGHT	QUAIL WEIGHT	SYMPTOMS	QUAIL DEAD <sup>2</sup>	PROBITS	MEAN WEIGHT (g)	MEAN WEIGHT CHANGES (g) <sup>3</sup>
Control <sup>4</sup>	-	1	183.3			-	181.4	-4.7
		2	173.6					
		3	179.3					
		4	189.2					
147	-	5	191.0	a		-	184.7	-1.8
		6	172.5	a				
		7	181.0					
		8	194.4	a				
250	-	9	161.0	ab		-	171.5	-4.0
		10	167.8	ab				
		11	170.2	ab				
		12	186.8	ab				
500	-	13	178.5	abc		-	177.2	-2.6
		14	186.3	abc				
		15	177.1	ab				
		16	166.8	ab				
870	2.9395	17	177.1	abcde		0.00	182.7	-8.3
		18	191.6	a				
		19	177.5	abcde				
		20	184.6	abc e				
1057	3.0241	21	167.3	abc e		4.33	180.2	-13.7
		22	173.2	bc e				
		23	185.6	cd	X			
		24	194.5	ab				
1140	3.0569	25	204.5	a		4.33	184.3	-7.2
		26	182.4	a cd				
		27	179.2	abc	X			
		28	170.9	a				
1225	3.0881	29	209.5	a		5.67	184.2	-22.8
		30	169.2	c ef	X			
		31	169.7	cde g	X			
		32	188.5	e	X			
1348	3.1297	33	167.6	a cde	X	5.67	189.0	-20.7
		34	212.1	a				
		35	183.3	abc e g	X			
		36	193.0	abc e	X			

<sup>1</sup> Corn oil was mixed with Sevin<sup>®</sup> to give a 1.0ml dose.

<sup>2</sup> All deaths were within 24 hours after the dosing.

<sup>3</sup> Weight change 4 days after the dosing.

<sup>4</sup> Received 1.0 ml corn oil.

a inactivity

b feathers roughed

c inability to stand

d salivation

e diarrhea

f lacrimation

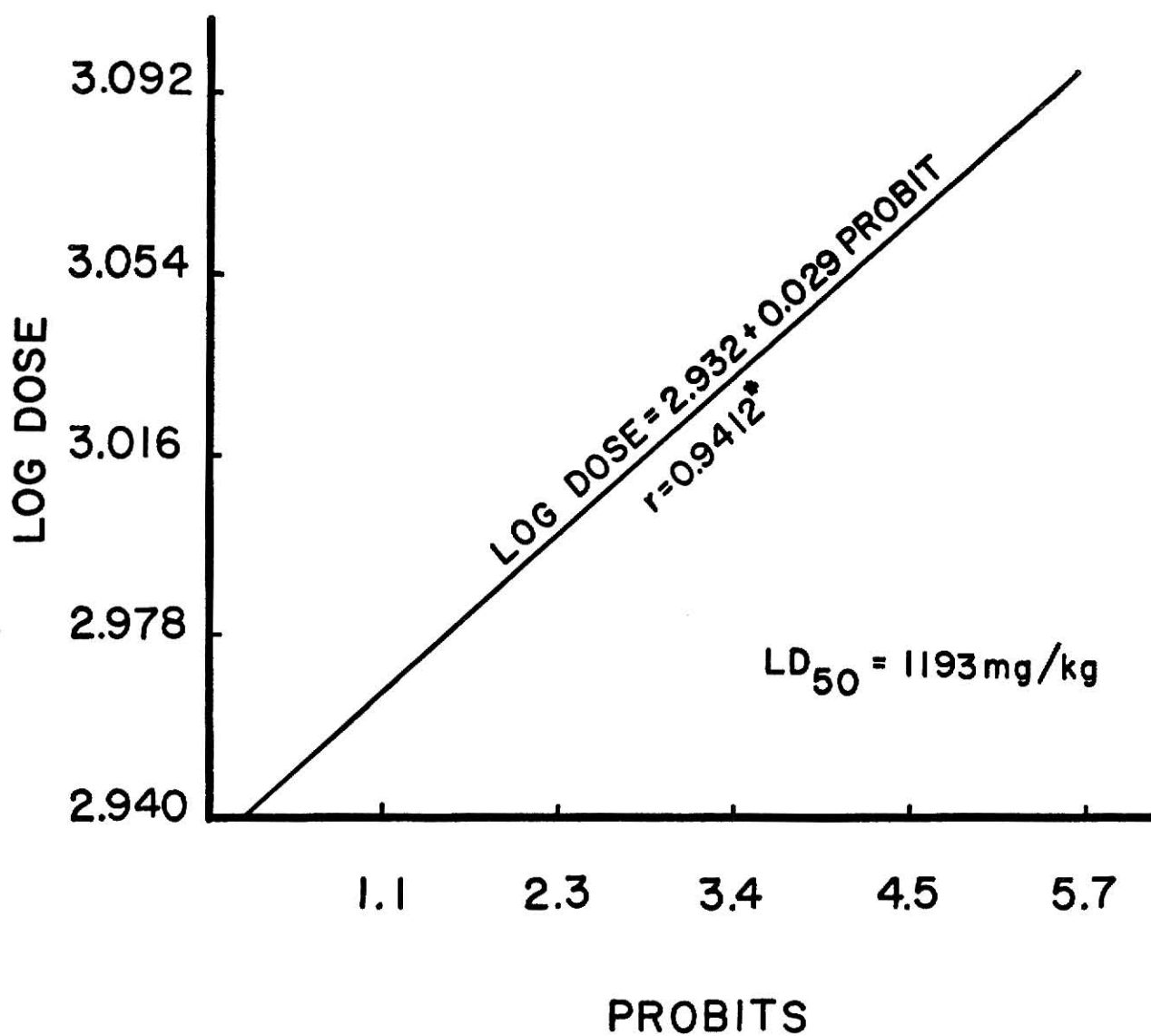
g oil regurgitation



**THIS BOOK  
CONTAINS  
NUMEROUS PAGES  
WITH DIAGRAMS  
THAT ARE CROOKED  
COMPARED TO THE  
REST OF THE  
INFORMATION ON  
THE PAGE.**

**THIS IS AS  
RECEIVED FROM  
CUSTOMER.**

Fig. 2. The probit-log dose regression equation used to determine the LD<sub>50</sub> of Sevin®, in corn oil to bobwhites. The data are listed in Table 2. \* P<.05



A change of fecal odor was noted when quail were first dosed at the beginning of period 2. All birds, including the corn oil control, excreted a watery, yellow feces at 1.5-2.0 hours after being dosed. This feces was oily, but no Sevin particles were noted.

The ratio data for gross energy intake, metabolized energy, excretory energy, efficiency of metabolism, fecal energy, and weight showed no significant treatment-period interaction ( $P>.05$ ) and no significant treatment effects (Table 5a, appendix). All energetic variables except body weight showed a significant ( $P<.05$ ) period effect.

### Weight

Of 112 bird-periods (28 birds X 4 periods) in the Sevin study, eight gained greater than 1.0 percent body weight (weight-gain quail). Twelve birds lost greater than 1.0 percent body weight (weight-loss quail). A chi-square analysis (Snedecor and Cochran 1972) of weight-gain, weight-loss, and weight-stable quail for each treatment (oil control, 10 mg/kg, 50 mg/kg, and 90 mg/kg Sevin) showed no significant deviations from expected values (Table 6, appendix). The Sevin treatments did not cause a greater number of quail to gain or lose more than 1.0 percent body weight. A chi-square analysis of these weight changes for each study period (1, 2, 3, 4, and 5) showed significant deviations. The first doses at period 2 caused a number of quail to lose weight, while at period 4 and 5, quail gained weight (Fig. 3).

The corn oil control, 10 mg/kg, 50 mg/kg, and 90 mg/kg Sevin quail, when first dosed (period 2), lost weight (Fig. 4). The greatest loss was 2.3 g by the 90 mg/kg quail, and the smallest loss was 0.6 g by the corn oil control. These were not significantly different ( $P>.05$ ). All treatment groups, except the 50 mg/kg group, began to gain weight at periods 3 or 4.

Fig. 3. The number of Sevin , weight-unstable quail ( expressed as a percent of the total quail number) for each 2-day period. Quail were first treated at period 2.

<sup>1</sup> Quail that lost greater than 1.0 percent body weight.

<sup>2</sup> Quail that gained greater than 1.0 percent body weight.

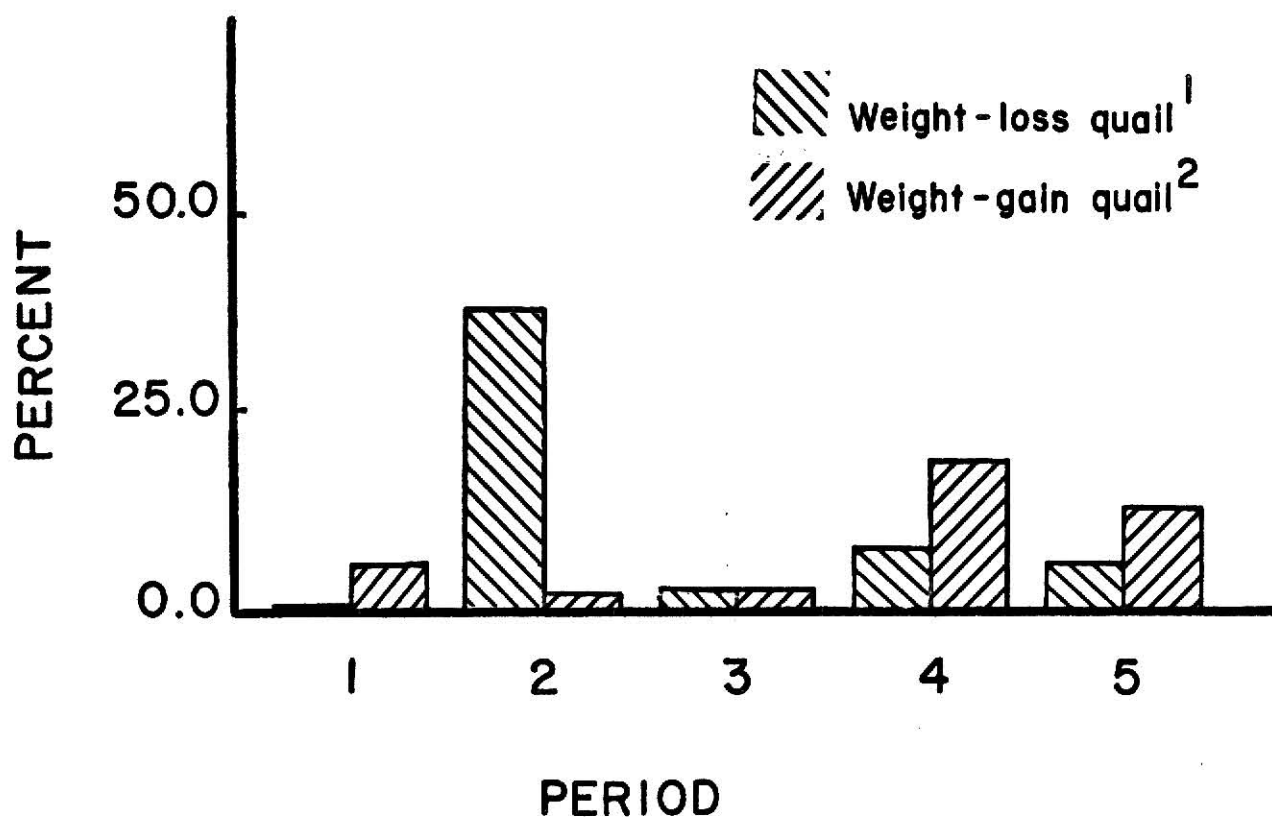
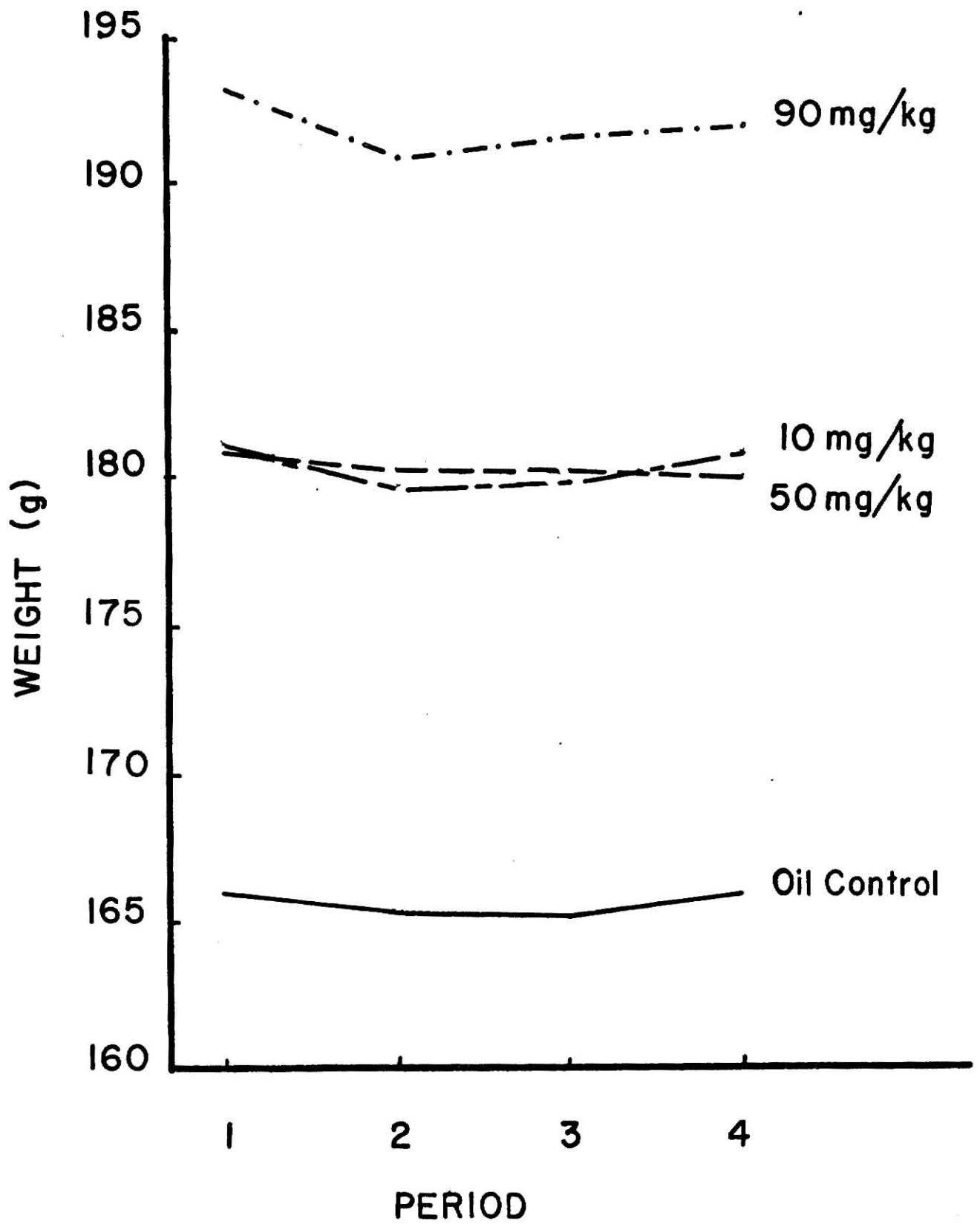




Fig. 4. Body weights of bobwhites treated with Sevin<sup>®</sup> and/or corn oil at periods 2 to 4. Quail were not treated during period 1. Each period was 2 days.



## Gross Energy

The two-way analysis of variance for gross energy intake ratios showed nonsignificant treatment-period interaction and treatment effects (Table 5a, appendix).

The period 1 (no treatment) gross energy intake mean was 40.327 kcal. When first dosed (period 2) the corn oil control, 10 mg/kg, 50 mg/kg, and 90 mg/kg Sevin quail decreased gross energy intake 9.124 kcal, 4.210 kcal, 11.108 kcal, and 14.887 kcal, respectively (Fig. 5a). The ratio data of period 2 (0.761, 0.867, 0.731, and 0.676, respectively) showed no significant dose related decreases.

The gross energy intake of all treatment groups increased at periods 3 and 4, even though quail were dosed at these periods. The rates at which each quail group increased were not significantly different. The corn oil control was the only treatment group to increase above its period 1 value at period 4 (period 4 ratio of 1.003). The 90 mg/kg Sevin treatment group, at period 4, showed the lowest ratio of 0.918, which was not significantly different from the oil control ratio.

A significant period (all treatment groups combined) effect was noted. With the first dose the period 1 mean of 40.327 kcal gross energy intake decreased significantly to 30.494 kcal at period 2. From period 2, two significant increases brought the gross energy to 38.692 kcal (period 4). This was not significantly different from period 1.

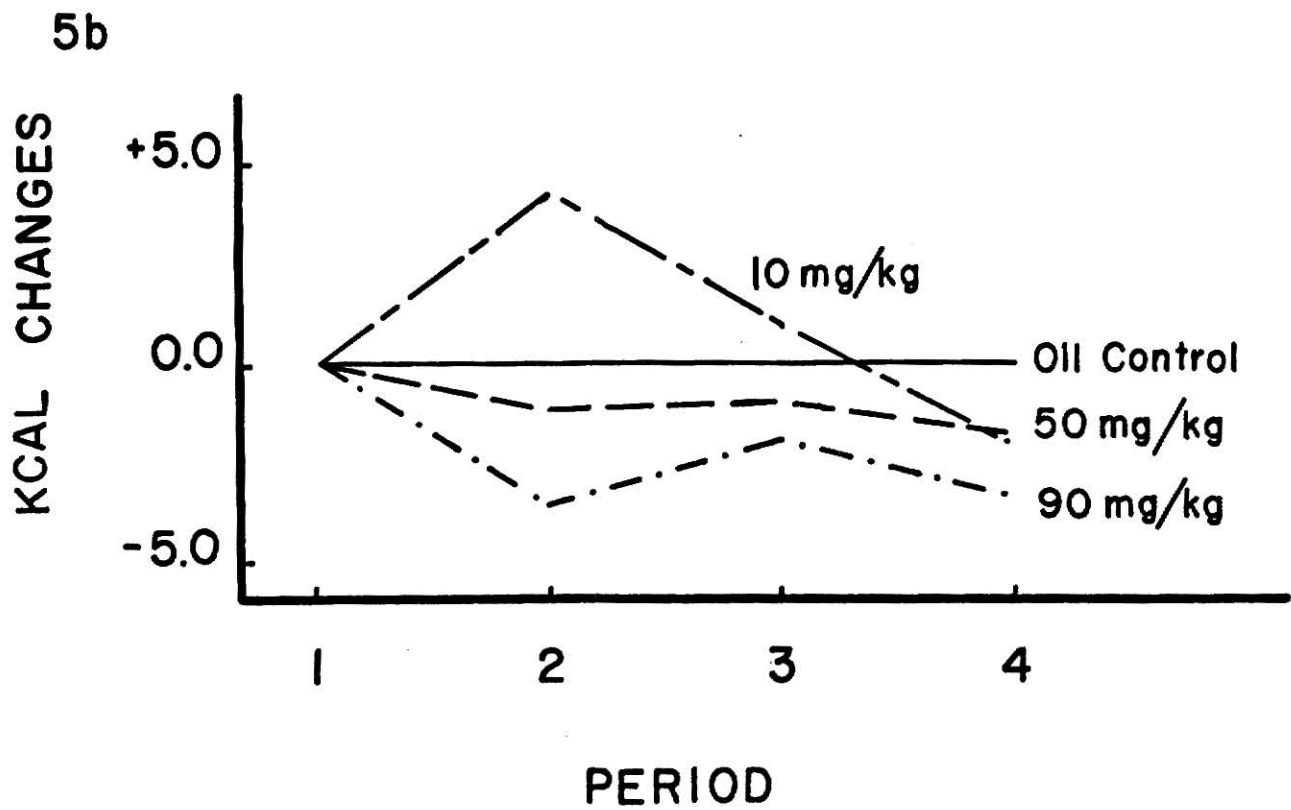
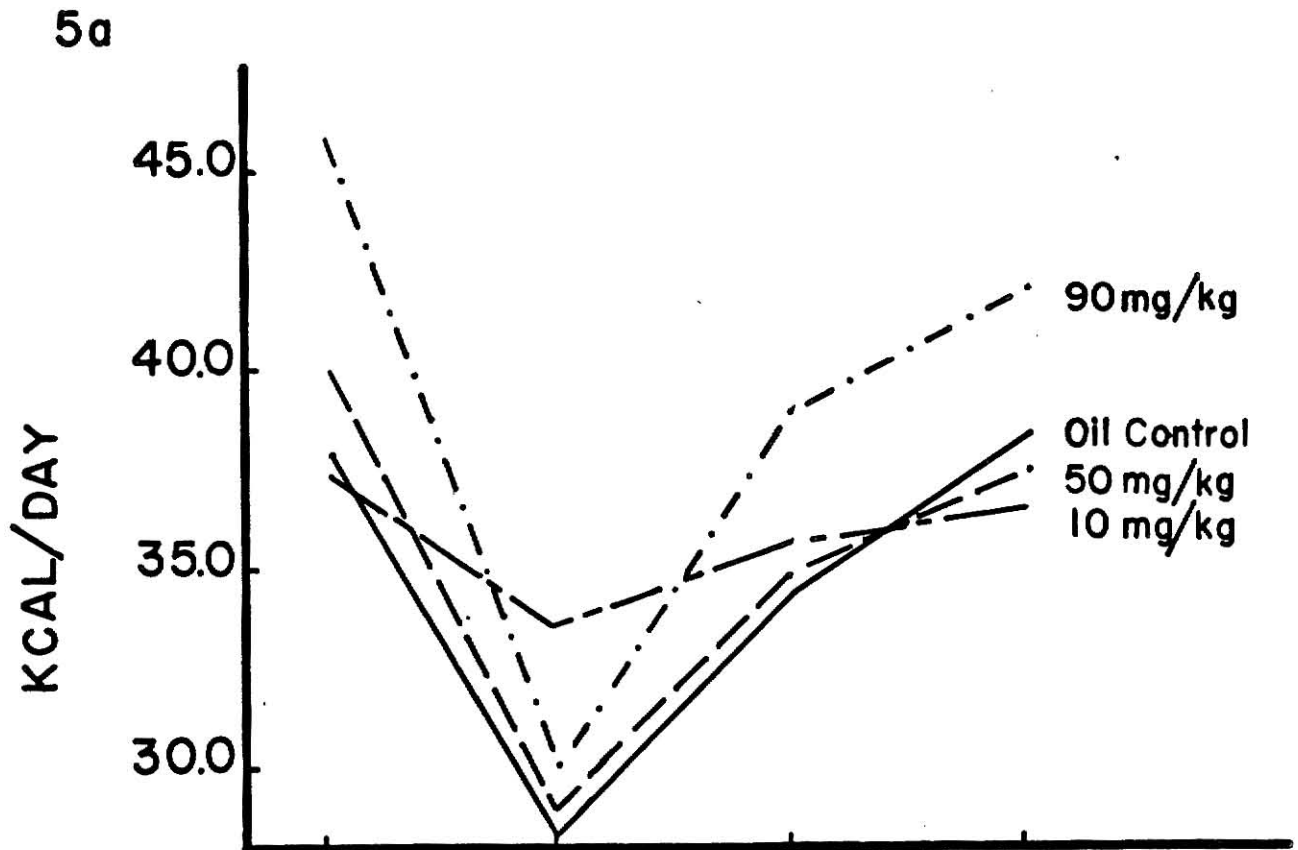
While figure 5a shows the gross energy intake of the 10 mg/kg quail as decreasing after the first treatment, these quail increased their gross energy intake in relation to the corn oil control (Fig. 5b). This period 2 increase of 4.275 kcal was significantly greater than the control. The period 2 decreases of 1.210 kcal and 3.428 kcal for the 50 mg/kg and 90 mg/kg quail, respectively, were not significantly lower than the control.





- Fig. 5a. Gross energy intake of bobwhites treated with Sevin<sup>®</sup> and/or corn oil at periods 2 to 4. Quail were not treated during period 1. Each period was 2 days.
- 5b. Gross energy intake changes of Sevin<sup>®</sup> treated quail in relation to the corn oil control.

## GROSS ENERGY INTAKE (SEVIN)



The overall (period 1-4) changes of +1.089 kcal for the 10 mg/kg quail, -1.271 kcal for the 50 mg/kg quail, and -2.944 kcal for the 90 mg/kg quail were not statistically separate from the control (Table 8, appendix).

#### Metabolized Energy

The two-way analysis of variance for metabolized energy ratios showed nonsignificant ( $P > .05$ ) treatment-period interaction and nonsignificant treatment effect (Table 5a, appendix).

The period 1 metabolizable energy mean was 30.299 kcal. The metabolizable energy of all treatment groups decreased after the first dosing (Fig. 6a): the corn oil control, 7.580 kcal; the 10 mg/kg Sevin dosed quail, 4.500 kcal; the 50 mg/kg quail, 9.271 kcal; and the 90 mg/kg quail, 12.664 kcal. The difference in the decreases of metabolized energy between corn oil treated and 90 mg/kg Sevin treated quail was significant.

Period 1-2 metabolized energy changes for all treatment groups were combined and found highly correlated ( $r = 0.9810$ ) with period 1-2 changes in gross energy intake (Table 7, appendix).

Metabolizable energy increased during period 3 and 4 (Fig. 6a). The rates at which all treatment groups were increasing were not significantly different because of the nonsignificant treatment-period interaction. The corn oil control was able to surpass, at period 4, its period 1 value of 28.520 kcal (period 4 ratio of 1.026). The 90 mg/kg quail were least able to reach, at period 4, their period 1 value of 34.876 kcal (0.902 ratio). This 0.902 ratio was significantly lower than the corn oil control ratio, 1.026.

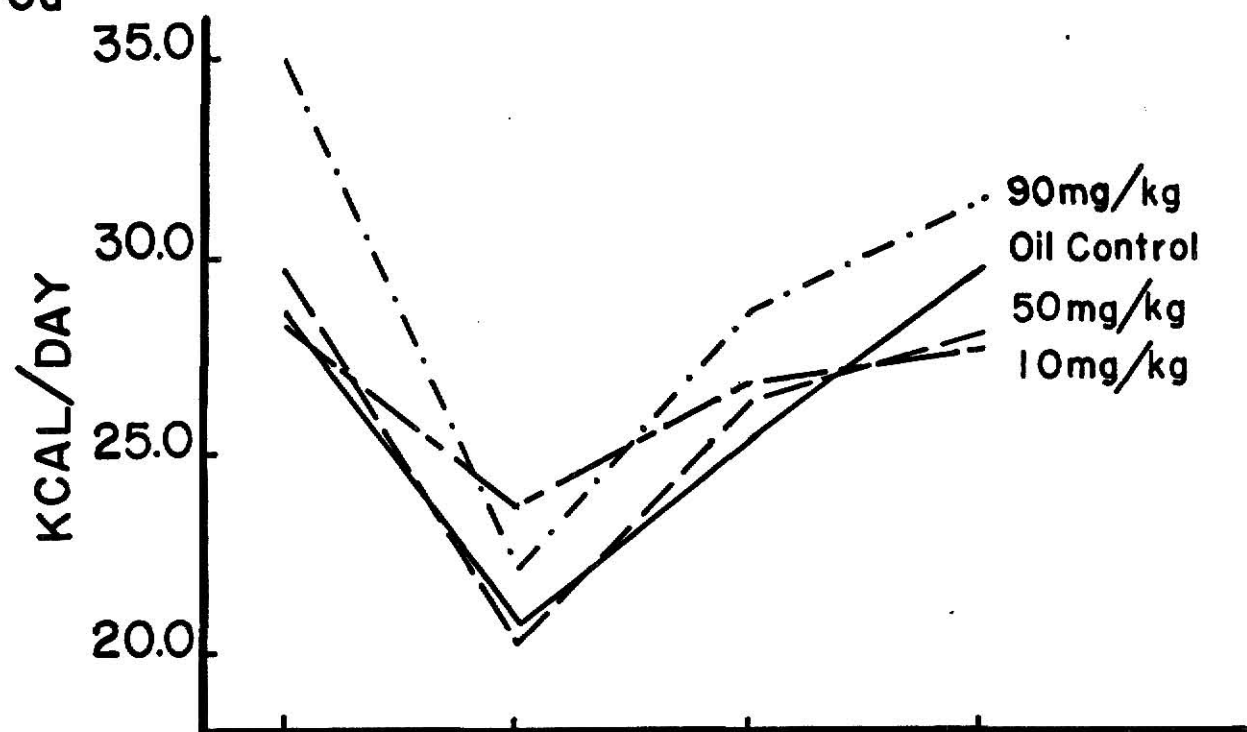
A significant period effect was noted. The period 1 mean of 30.299 kcal metabolized energy decreased significantly to 21.795 kcal after the first dosing at period 2. Significant increases from period 2 gave a metabolizable energy of 29.260 kcal at period 4. This was nonsignificantly different from



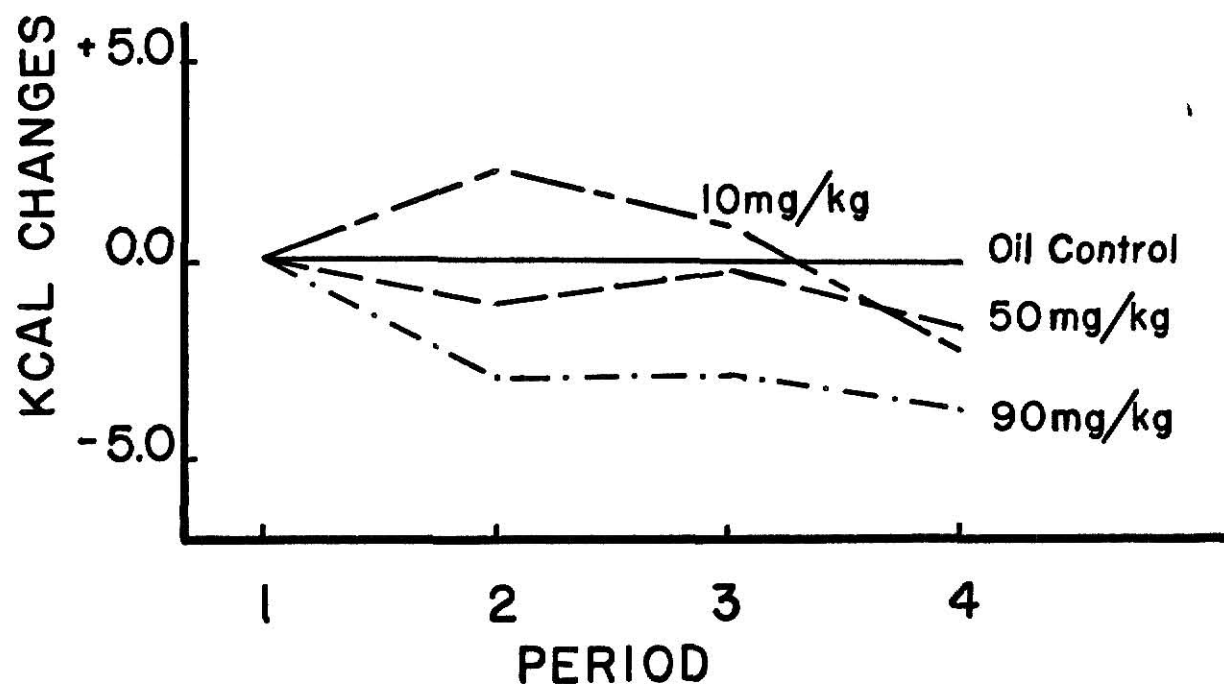
- Fig. 6a. Metabolized energy of bobwhites treated with Sevin<sup>®</sup> and/or corn oil, periods 2 to 4. Quail were not treated during period 1.
- 6b. Metabolized energy changes of Sevin<sup>®</sup> treated quail in relation to the corn oil control.

## METABOLIZED ENERGY (SEVIN)

6a



6b



period 1. Figure 6b shows the 10 mg/kg quail increasing metabolized energy nonsignificantly at period 2 by 2.212 kcal (in relation to the corn oil control). The 50 and 90 mg/kg Sevin treatment decreased metabolized energy 1.121 and 3.000 kcal, respectively. Only the 90 mg/kg treatment decreased metabolized energy significantly lower than the corn oil control at period 2. The overall means though were not statistically different from the control (Table 8, appendix).

### Excretory Energy

The two-way analysis of variance for excretory energy ratios showed nonsignificant treatment-period interaction and treatment effect (Table 5a, appendix).

The period 1 mean excretory energy was 9.976 kcal. The excretory energy of the control, 10 mg/kg, 50 mg/kg, and 90 mg/kg Sevin dosed quail decreased (at period 2) by 1.545 kcal, 0.062 kcal, 1.837 kcal, and 2.223 kcal, respectively (Fig. 7a). These decreases were not significantly different.

The overall period 1 to period 2 changes (all treatments combined) in gross energy intake, and metabolizable energy were highly correlated ( $P < .01$ ) with period 1 to period 2 changes in excretory energy (Table 7, appendix).

All treatment groups, at period 4, had not recovered to period 1 values. The significant period effect though showed the period 4 mean excretory energy (9.451 kcal) as not statistically different from the period 1 mean (9.976 kcal).

The 10 mg/kg Sevin dosed quail increased excretory energy significantly by 1.597 kcal, at period 2, in relation to the corn oil control (Fig. 7b). The 50 mg/kg and 90 mg/kg quail decreased 0.080 kcal and 0.349 kcal, respectively. The overall changes (period 1 through 4) of the 10, 50, and 90 mg/kg quail were +0.718 kcal, -0.249 kcal, and +0.060 kcal, respectively. These

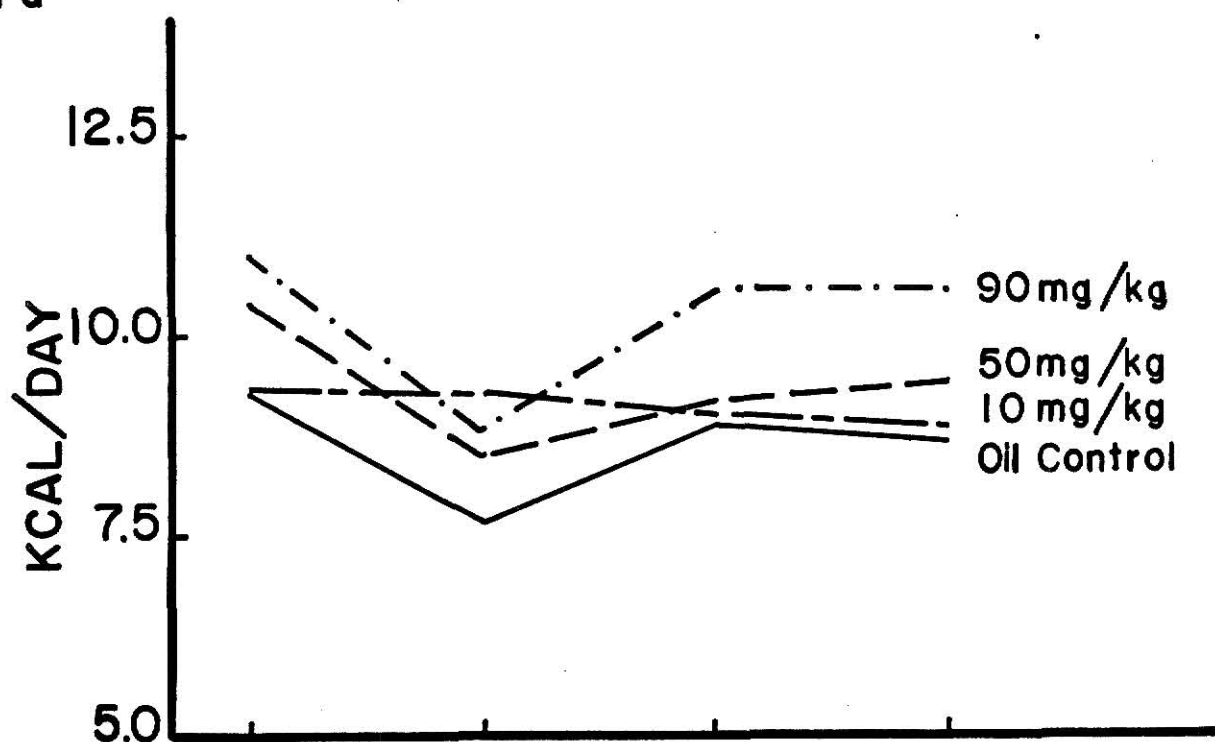




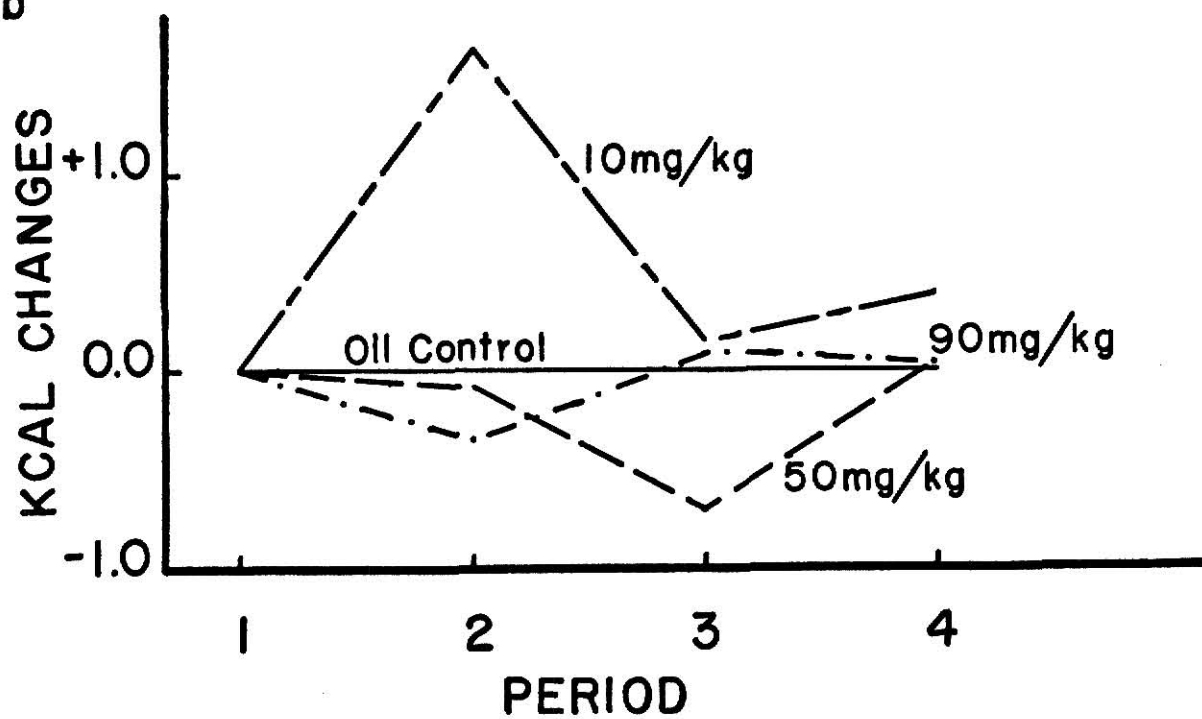
- Fig. 7a. Excretory energy of bobwhites treated with Sevin® and/or corn oil, periods 2 to 4. Quail were not treated during period 1.
- 7b. Excretory energy changes of Sevin® treated quail in relation to the corn oil control.

## EXCRETORY ENERGY (SEVIN)

7a



7b



were not significantly different from the control or among themselves (Table 8, appendix).

#### Efficiency of Metabolism

The two-way analysis of variance for efficiency ratios showed nonsignificant treatment-period interaction and treatment effects (Table 5a, appendix).

The period 1 mean efficiency of metabolism was 75.23 percent. Period 1 to period 2 decreases were observed for all treatment groups (Fig. 8a): control, 2.13 percent; 10 mg/kg Sevin, 2.99 percent; 50 mg/kg Sevin, 4.32 percent; and 90 mg/kg Sevin, 3.52 percent. The 90 mg/kg quail change was significantly greater than the control change. The overall changes (all treatments combined), from period 1 to period 2, in efficiency of metabolism and metabolized energy were statistically correlated (Table 7 appendix).

The general period 1-2 decrease and period 2-4 increase of efficiency for all treatments was reflected in the significant period effect. The period 1 value of 75.23 percent decreased with the first dose to 72.00 percent. The efficiency then increased significantly to a period 4 value of 75.59 percent, which was nonsignificantly greater than period 1.

In relation to the corn oil control quail, the Sevin quail had a lower efficiency of metabolism (Fig. 8b). The overall treatment changes were -1.13 percent for the 10 mg/kg treatment, -0.53 percent for the 50 mg/kg treatment, and -2.26 percent for the 90 mg/kg treatment. No significant differences existed (Table 8, appendix).

#### Fecal Energy

The two-way analysis of variance for fecal energy showed nonsignificant treatment-period interaction and treatment effects (Table 5a, appendix).

The period 1 mean fecal energy was 3.239 kcal/g. The fecal energy (Fig. 9a) increased with the first treatment doses: 0.321 kcal/g for the oil control,



Fig. 8a. Efficiency of metabolism of bobwhites treated with Sevin® and/or corn oil, periods 2 to 4. Quail were not treated during period 1.

8b. Efficiency of metabolism changes of Sevin® treated quail in relation to the corn oil control.

## EFFICIENCY OF METABOLISM (SEVIN)

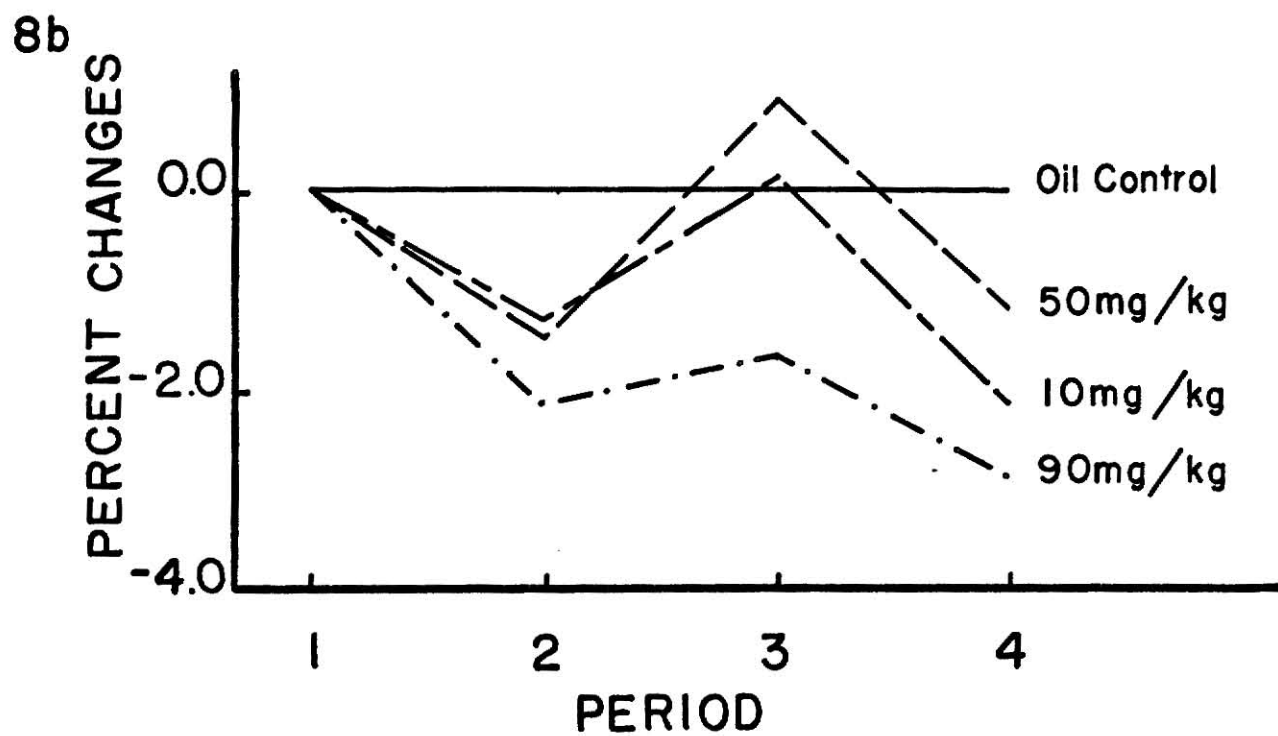
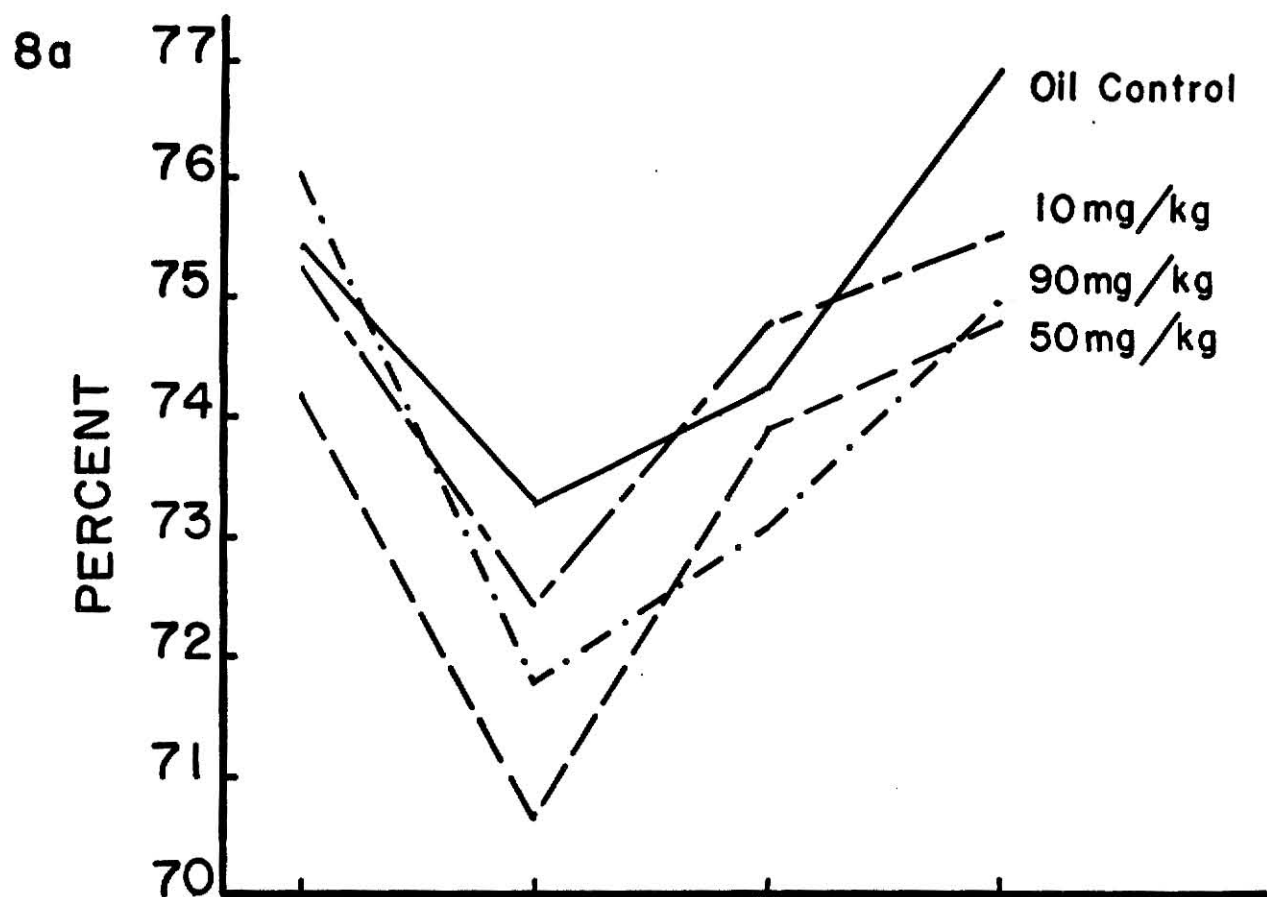
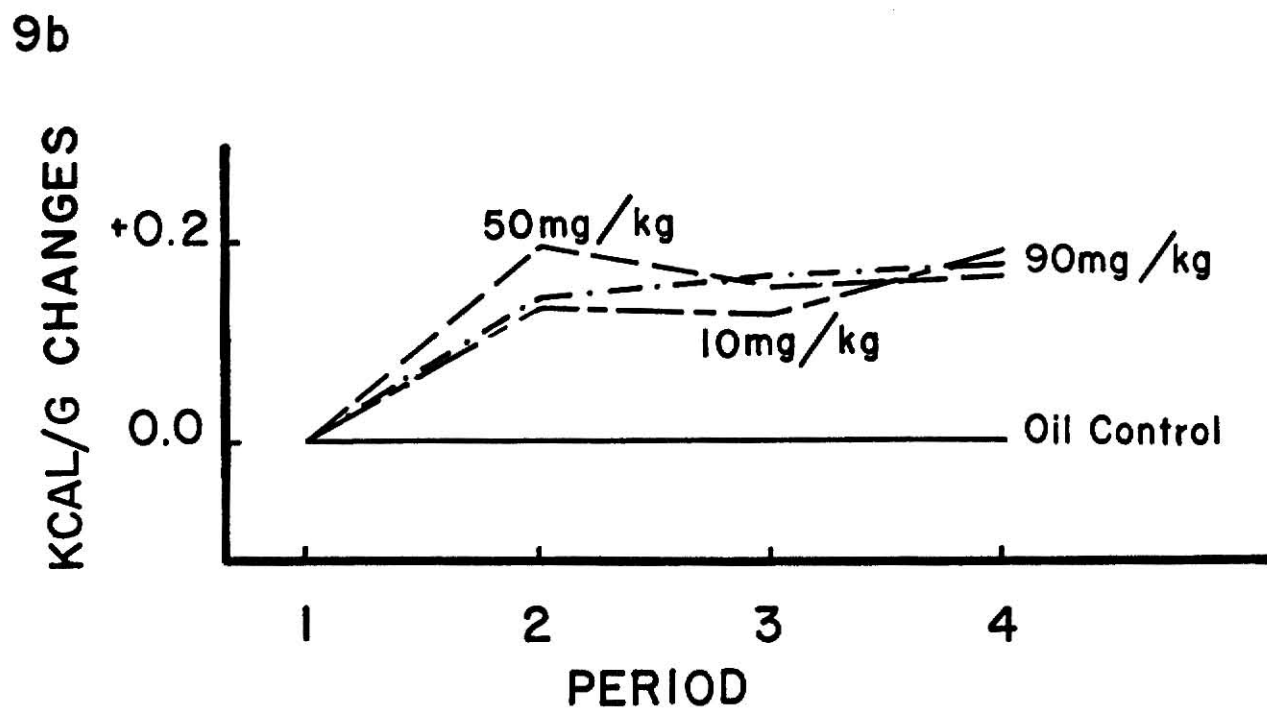
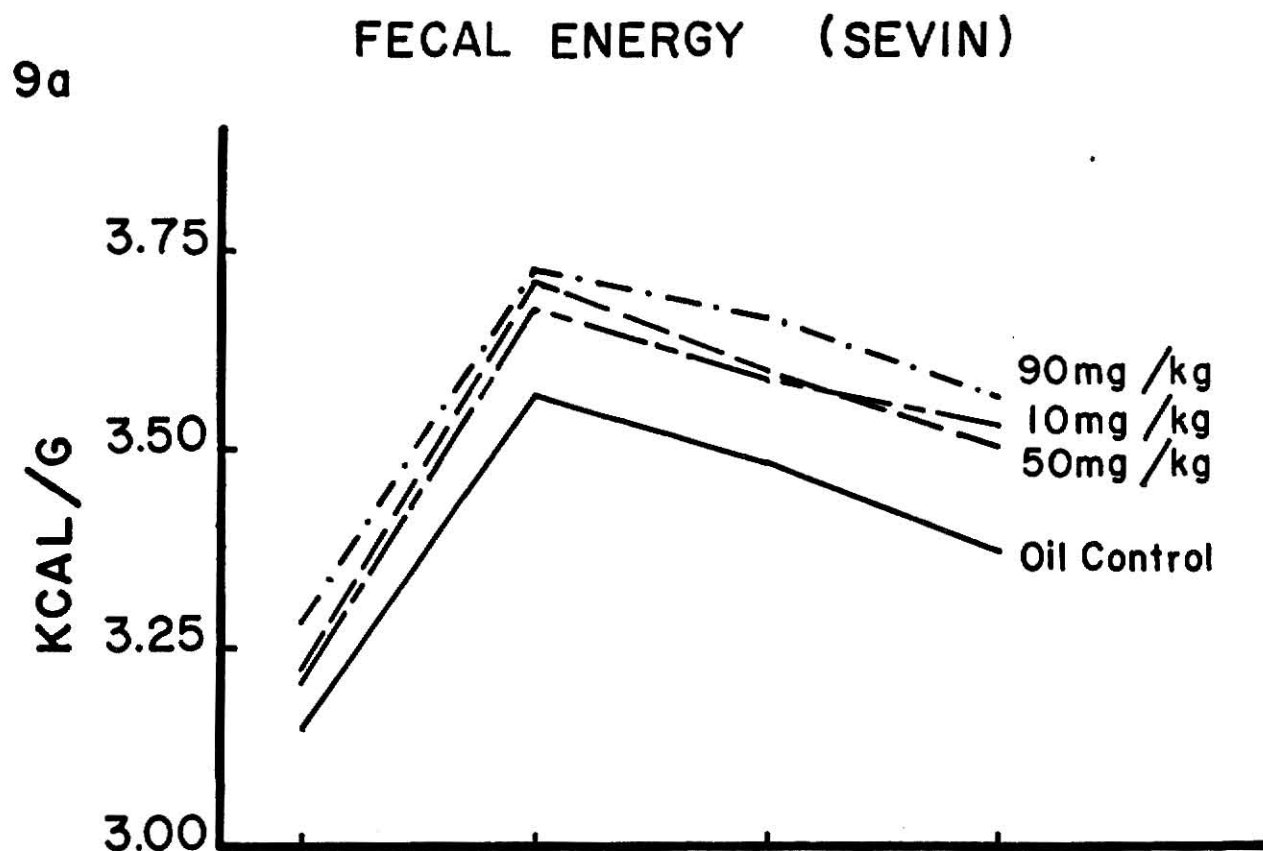






Fig. 9a. Fecal energy of bobwhites treated with Sevin<sup>®</sup> and/or corn oil, periods 2 to 4. Quail were not treated during period 1.

9b. Fecal energy changes of Sevin<sup>®</sup> treated quail in relation to the corn oil control.



0.475 kcal/g for the 10 mg/kg Sevin dosed quail, 0.502 kcal/g for the 50 mg/kg quail, and 0.463 kcal/g for the 90 mg/kg quail. The Sevin treated quails' ratios at period 2 (10 mg/kg quail, 1.141; 50 mg/kg quail, 1.156; and 90 mg/kg quail, 1.142) were significantly greater than the corn oil control ratio (1.099), but were not significantly different among themselves.

The significant period effect showed that the fecal energy increased with the first dosing, 3.239 kcal/g to 3.679 kcal/g (period 2). Two significant decreases at periods 3 and 4 lowered this variable to 3.496 kcal/g, which was still significantly greater than period 1.

In relation to the corn oil control (Fig. 9b), the 10 mg/kg, 50 mg/kg, and 90 mg/kg quail's overall fecal energy changes were +0.152 kcal/g, +0.168 kcal, and +0.155 kcal, respectively. The 0.168 kcal/g increase was significantly greater than the control (Table 8, appendix).

#### Furadan® Energetics Study

The mean caloric content and mean moisture content of the food was 4.275 kcal/g and 4.7 percent, respectively (Table 4, appendix). Mean caloric content of the corn oil was 9.395 kcal/g.

Duplicate oxygen bombings were performed on 50 of 184 fecal samples. The mean percent differences between two bombings of the same sample was 0.496 percent.

A two-way analysis of variance for the energetic variable ratio data showed no significant treatment-period interaction. Gross energy intake, metabolized energy, fecal energy, and weight showed significant treatment effects (Table 5b, appendix). All variables, except weight, showed a significant period effect.

## Weight

Of the 128 bird-periods (32 quail X 4 periods) eight gained greater than 1.0 percent body weight (weight-gain quail) and 14 lost greater than 1.0 percent body weight (weight-loss quail). A chi-square analysis of weight-loss, weight-gain, and weight-stable quail for the five dosing treatments (water control, oil control, 0.25 mg/kg, 1.13 mg/kg, and 2.0 mg/kg Furadan®) showed no significant deviations from expected values (Table 6, appendix). The different treatments did not cause quail to gain or lose greater than 1.0 percent body weight. The chi-square of weight changes and periods (1 through 5) showed significant deviations. The greatest deviation occurred when 14 quail lost weight during period 1.

The water control, corn oil control, low (0.25 mg/kg) Furadan, and medium (1.13 mg/kg) Furadan treatment quail gained weight periods 1 through 3 and lost weight at period 4 (Fig. 10a). The corn oil control had the largest period 1 to period 2 increase of 2.3 grams. The high (2.0 mg/kg) Furadan treatment quail lost weight during periods 1 through 3, and gained during period 4.

Quail weights decreased in relation to the corn oil control (Fig. 10b). The overall mean weights (periods 1-4) for the low, medium, and high Furadan dosed quail decreased 1.5, 2.2, and 3.3 grams, respectively. The 2.2 and 3.3 gram losses were statistically different from the oil control, and were statistically separate themselves (Table 10, appendix).

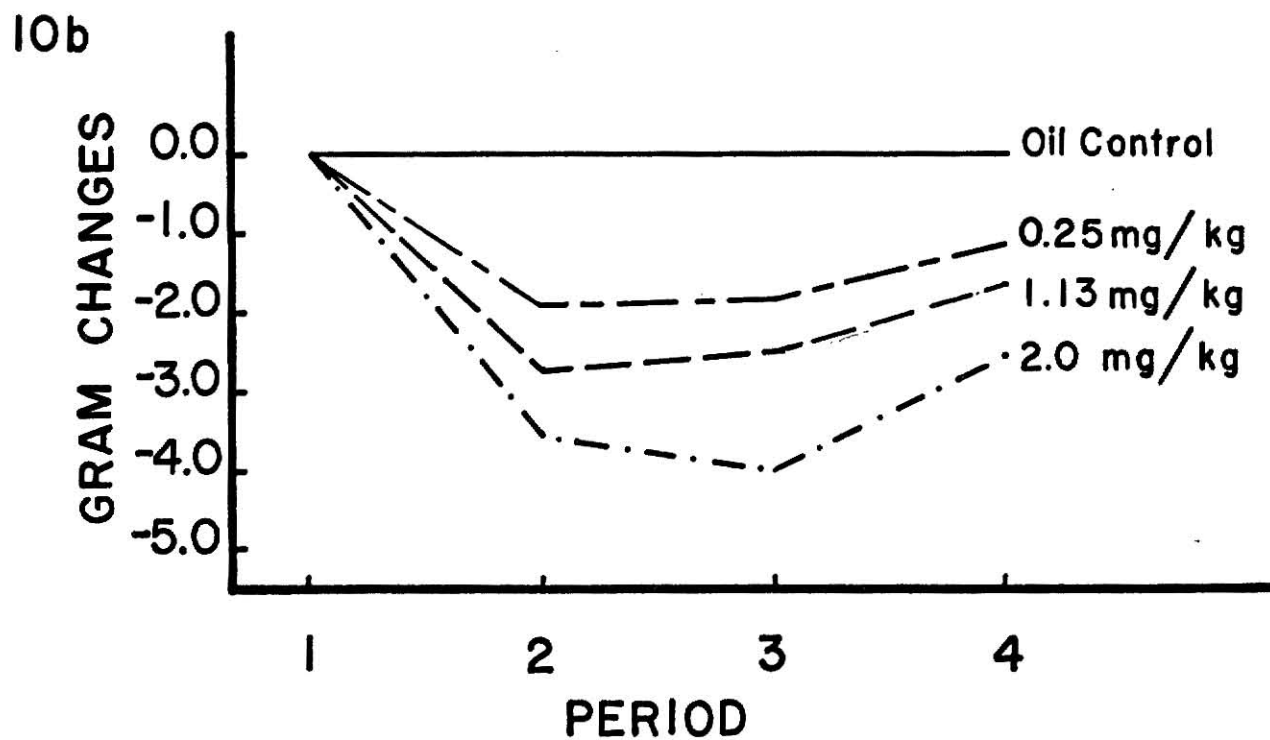
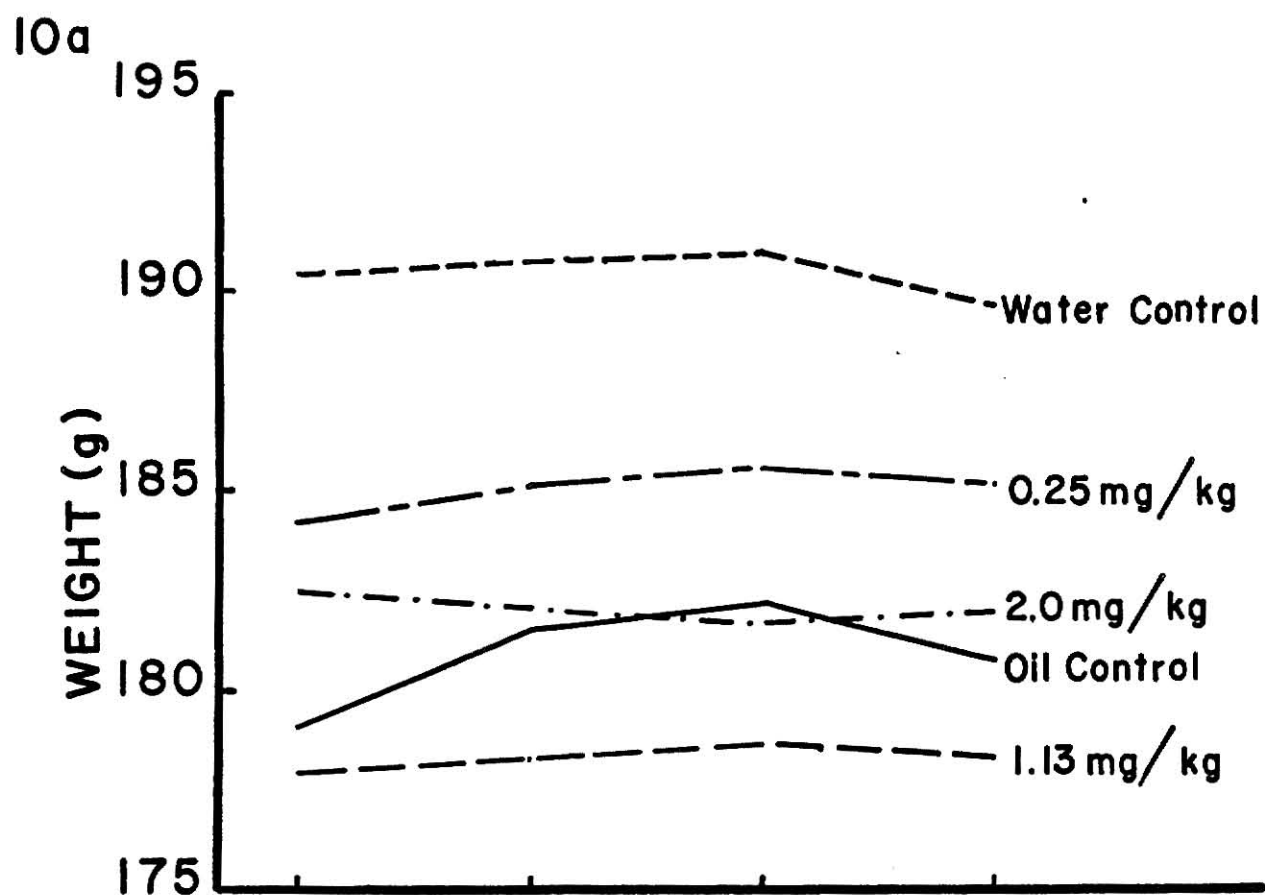
## Gross Energy Intake and Metabolized Energy

The gross energy intake and the metabolized energy results showed identical significance patterns (Table 5b, appendix). The gross energy intake and metabolized energy values were highly correlated (Table 9, appendix).



Fig. 10a. Body weights of bobwhites treated with Furadan<sup>®</sup> and/or corn oil, and water, during periods 2 to 4. Quail were not treated during period 1.

10b. Body weight changes of Furadan<sup>®</sup> treated quail in relation to the corn oil control.



The period 1 means for gross energy intake and metabolized energy were 40.568 kcal and 30.210 kcal, respectively. The corn oil control, for both variables, was the only quail group to show an increase with the first dosing (Fig. 11a and 12a). The corn oil control increases of 4.019 kcal gross energy and 2.861 kcal metabolized energy were not significant. The water control, and the low, medium, and high Furadan groups decreased gross energy intake and metabolized energy after the first dosing. The high Furadan dosed quail decreased significantly, 5.119 kcal gross energy intake and 4.517 kcal metabolized energy. All quail groups increased these two variables during periods 2 to 4.

All three Furadan dosages, in relation to the corn oil control (Fig. 11b and 12b), decreased the period 2 gross energy intake and metabolized energy significantly. The low (0.25 mg/kg) Furadan dose effect and the medium (1.13 mg/kg) Furadan dose effect were not significantly different. The low and medium treatment gross energy intake decreases at period 2 were 7.018 and 6.856 kcal, respectively. The low and medium treatment metabolized energy decreases at period 2 were 5.045 and 5.317 kcal, respectively. The high (2.0 mg/kg) Furadan treatment decreases in gross energy (10.913 kcal) and metabolized energy (8.429 kcal) at period 2, were significantly greater than the low and medium Furadan treatment decreases. Similar patterns of significance were noted with subsequent dosings at periods 3 and 4 (Fig. 11b and 12b).

The overall treatment means (Fig. 11b and 12b) for gross energy intake and metabolized energy showed that the medium and high Furadan treatments were significantly below the oil control (Table 10, appendix) and significantly different themselves. The medium and high Furadan treatments showed respective gross energy intake decreases of 5.355 and 9.331 kcal, and respective metabolized energy decreases of 4.380 and 7.281 kcal. The low Furadan treatment decreases were not significantly below the control.

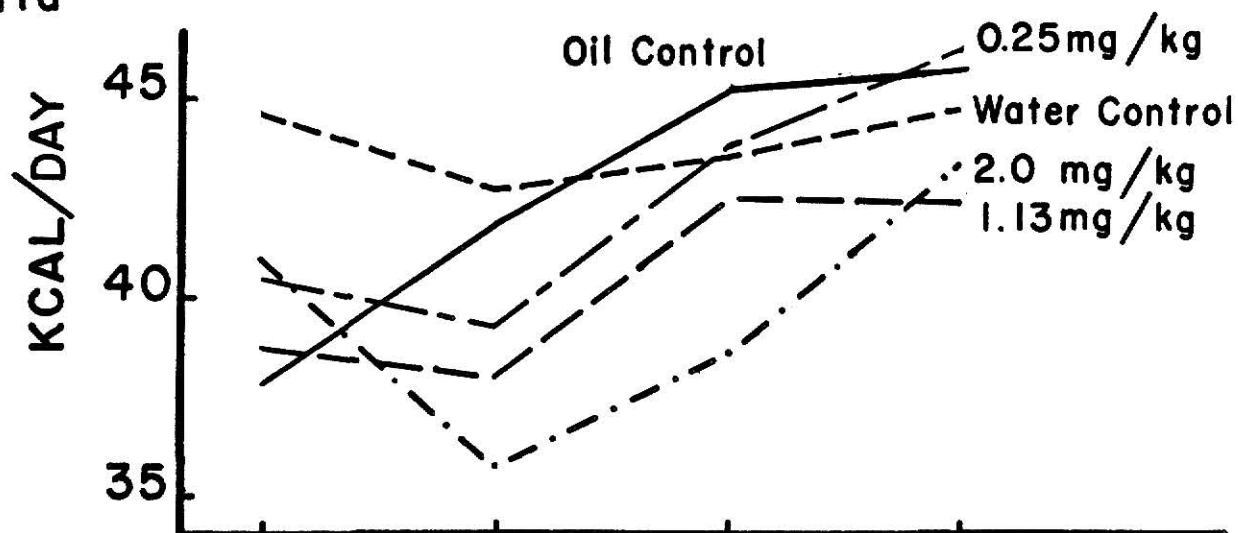




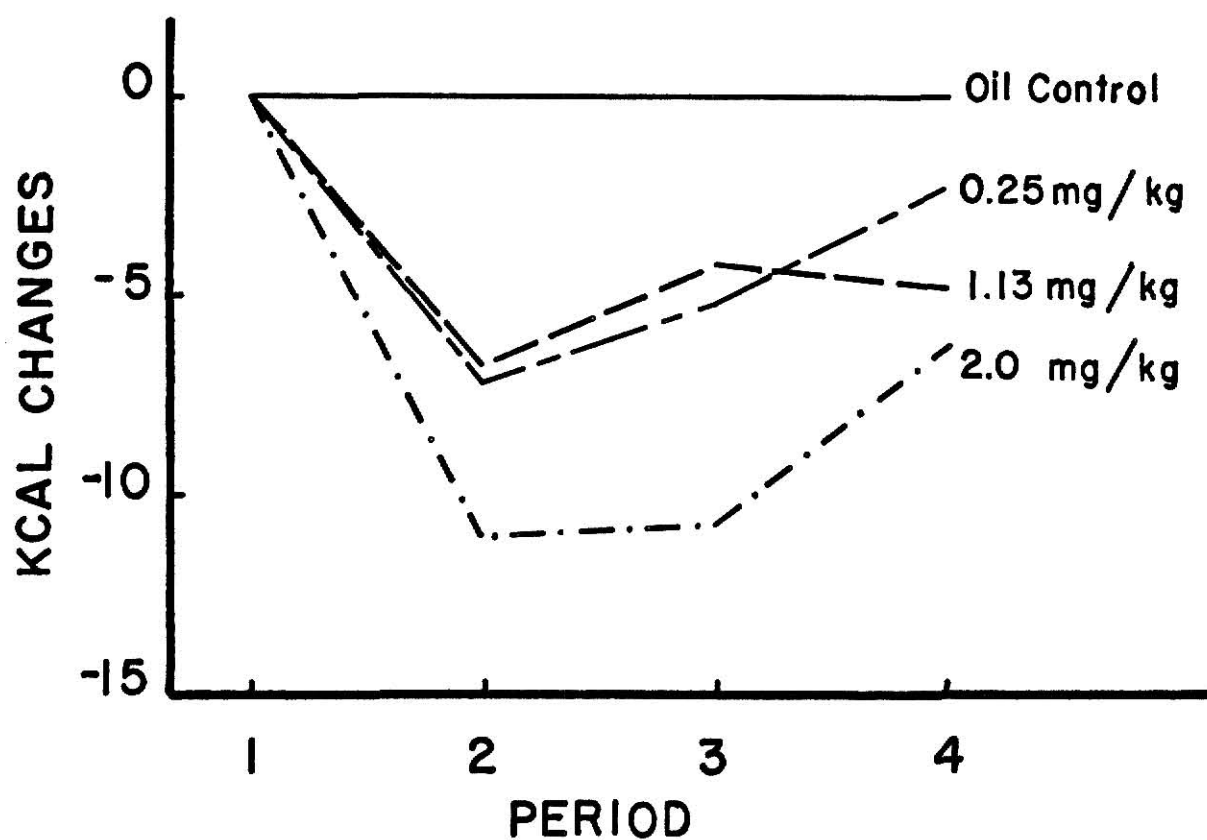
- Fig. 11a. Gross energy intake of bobwhites treated with Furadan<sup>®</sup>, and/or corn oil, and water during periods 2 to 4. Quail were not treated during period 1.
- 11b. Gross energy intake changes of Furadan<sup>®</sup> treated quail in relation to the corn oil control.

## GROSS ENERGY INTAKE (FURADAN)

IIa



IIb

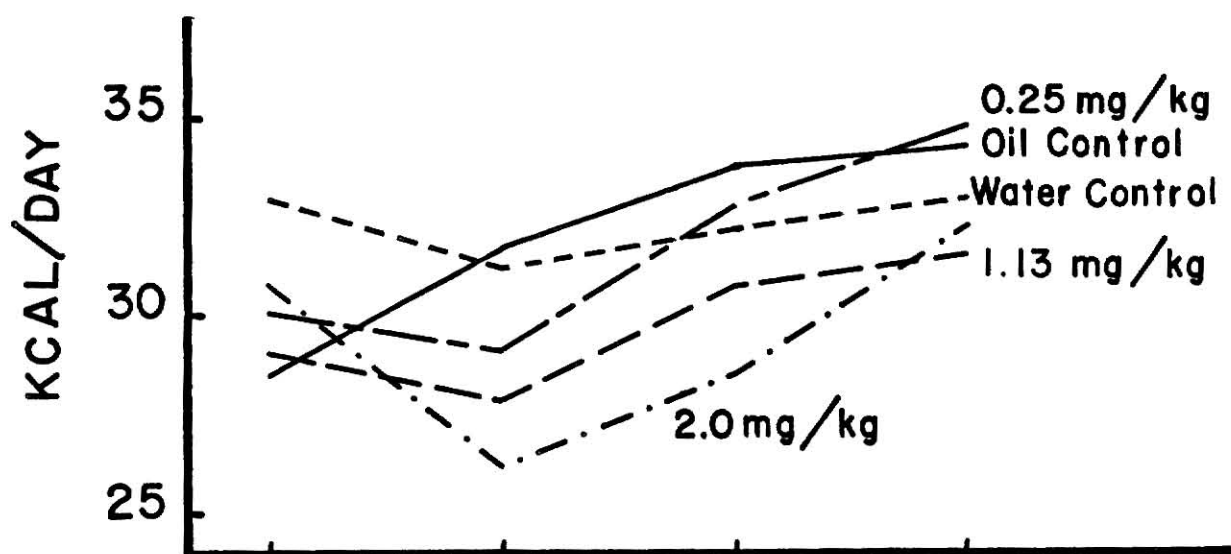




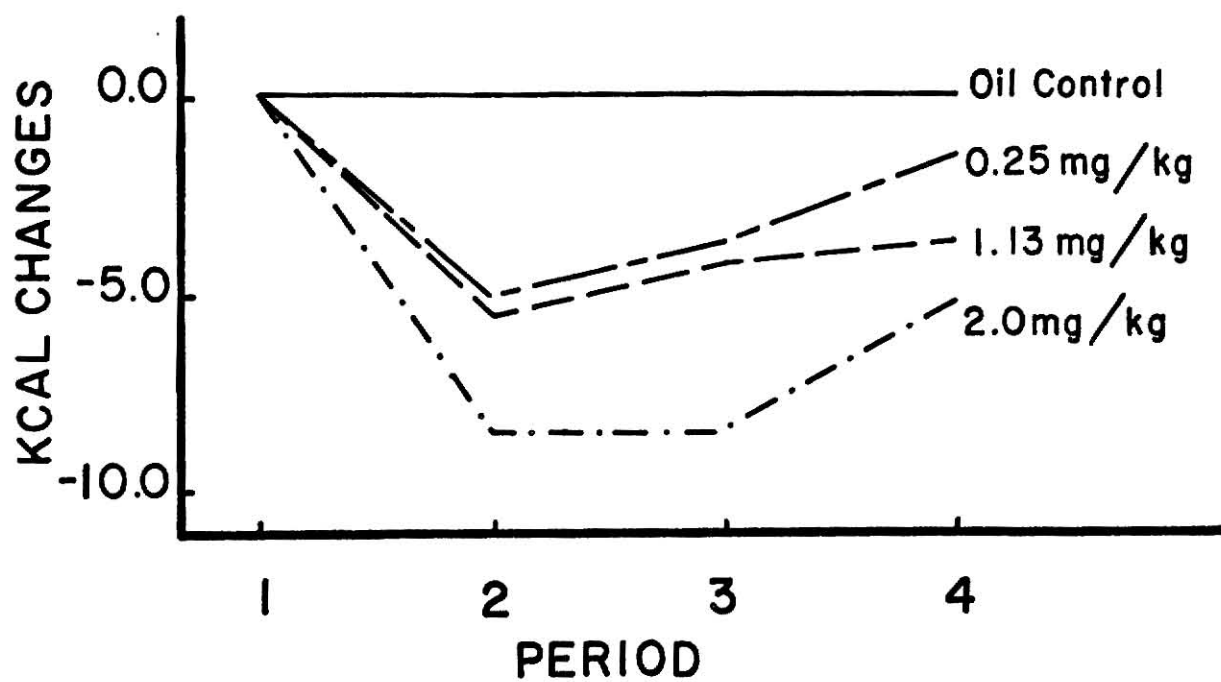
- Fig. 12a. Metabolized energy of bobwhites treated with Furadan<sup>®</sup>, and/or corn oil, and water during periods 2 to 4. Quail were not treated during period 1.
- 12b. Metabolized energy changes of Furadan<sup>®</sup> treated quail in relation to the corn oil control.

## METABOLIZED ENERGY (FURADAN)

12a



12b

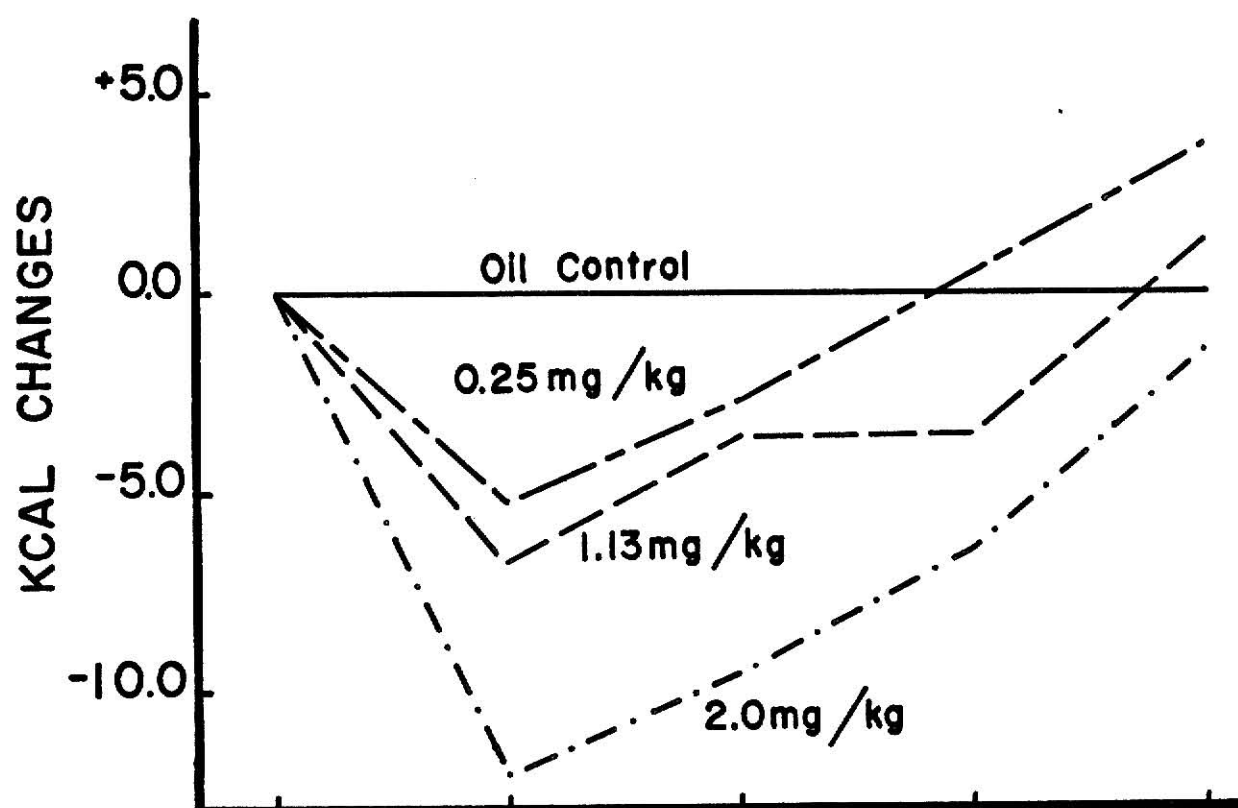




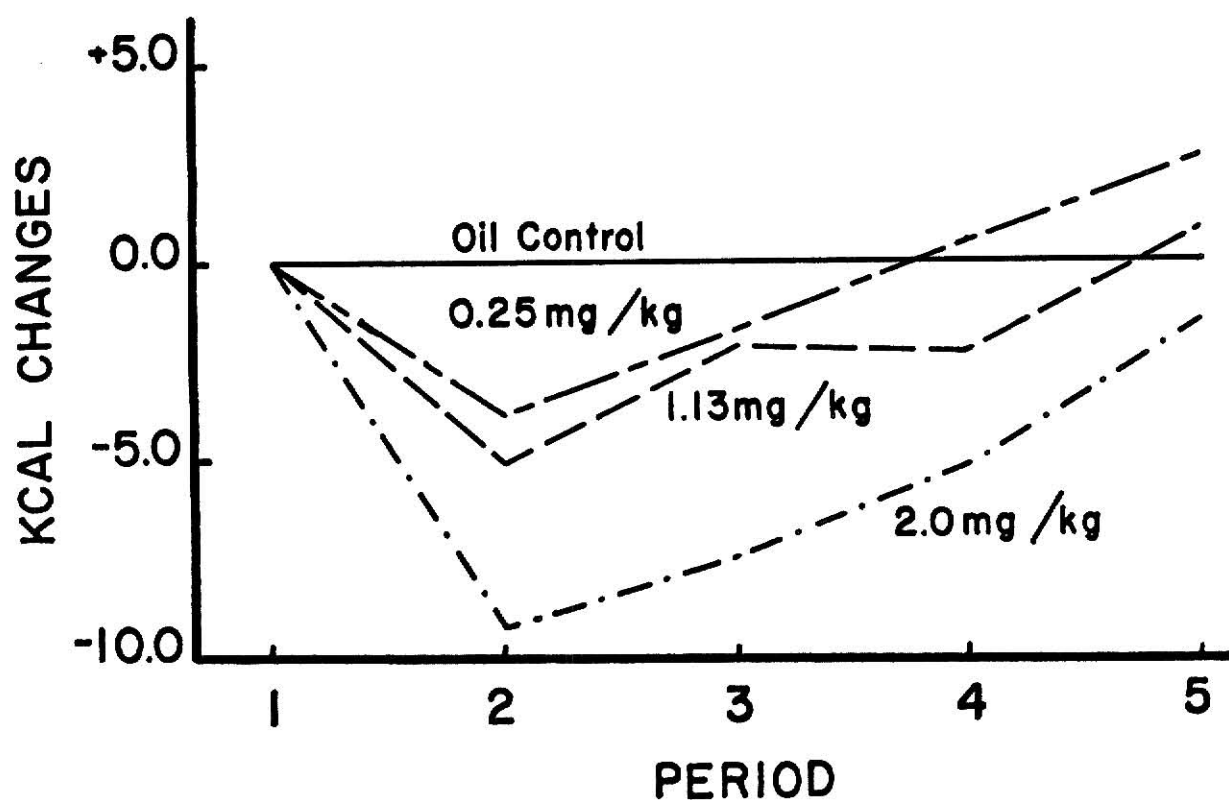
- Fig. 13a. The gross energy intake changes of Furadan® treated quail (periods 1 to 5), in relation to the corn oil control.
- 13b. The metabolized energy changes of Furadan® treated quail (periods 1 to 5), in relation to the corn oil control.



## 13a GROSS ENERGY INTAKE (FURADAN)



## 13b METABOLIZED ENERGY (FURADAN)



Figures 11b and 12b show gross energy intake and metabolized energy changes approaching 0.000 (corn oil control), at periods 2 to 4. To determine if the treatment groups reached the control at period 5, the period 1-5 data (page 15) were analyzed on the ratio basis (page 15). The low and medium Furadan quail were able to surpass the control at period 5, while the high Furadan quail did not (Fig. 13a and 13b). The high Furadan dosed quail at period 5 were not significantly below the control.

#### Excretory Energy

The period 1 to period 2 excretory energy changes (Fig. 14a) were all nonsignificant ( $P > .05$ ). The corn oil control and medium Furadan quail increased excretory energy by 1.156 kcal and 0.276 kcal, respectively. The low and high Furadan quail decreased excretory energy by 0.136 kcal and 0.640 kcal, respectively.

The three Furadan dosed quail groups, in relation to the corn oil control, decreased excretory energy significantly at period 2 (Fig. 14b). The low, medium, and high Furadan groups decreased excretory energy by 2.001 kcal, 1.576 kcal, and 2.519 kcal, respectively. The nonsignificant overall (period 1-4 ratios) treatment effects showed only the high Furadan treatment as statistically lower (2.104 kcal) than the corn oil control (Table 10, appendix). The low and medium Furadan treatments decreased excretory energy, overall, by 1.493 kcal and 1.244 kcal, respectively.

#### Efficiency of Metabolism

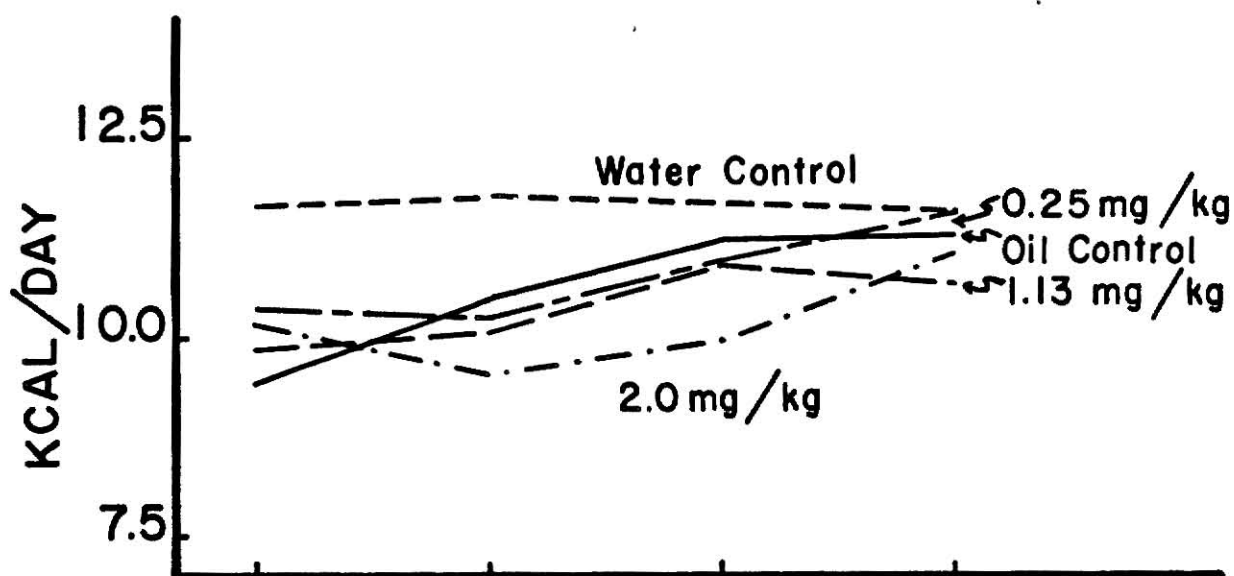
All quail groups decreased efficiency of metabolism when first dosed (Fig. 15a). The medium and high insecticide doses caused significant decreases of 1.44 percent and 1.90 percent, respectively. These were not statistically different themselves. All efficiencies increased from period 2 to period 4.



- Fig. 14a. Excretory energy of bobwhites treated with Furadan<sup>®</sup>, and/or corn oil, and water during periods 2 to 4. Quail were not treated during period 1.
- 14b. Excretory energy changes of Furadan<sup>®</sup> treated bobwhites in relation to the corn oil control.

## EXCRETORY ENERGY (FURADAN)

14a



14b

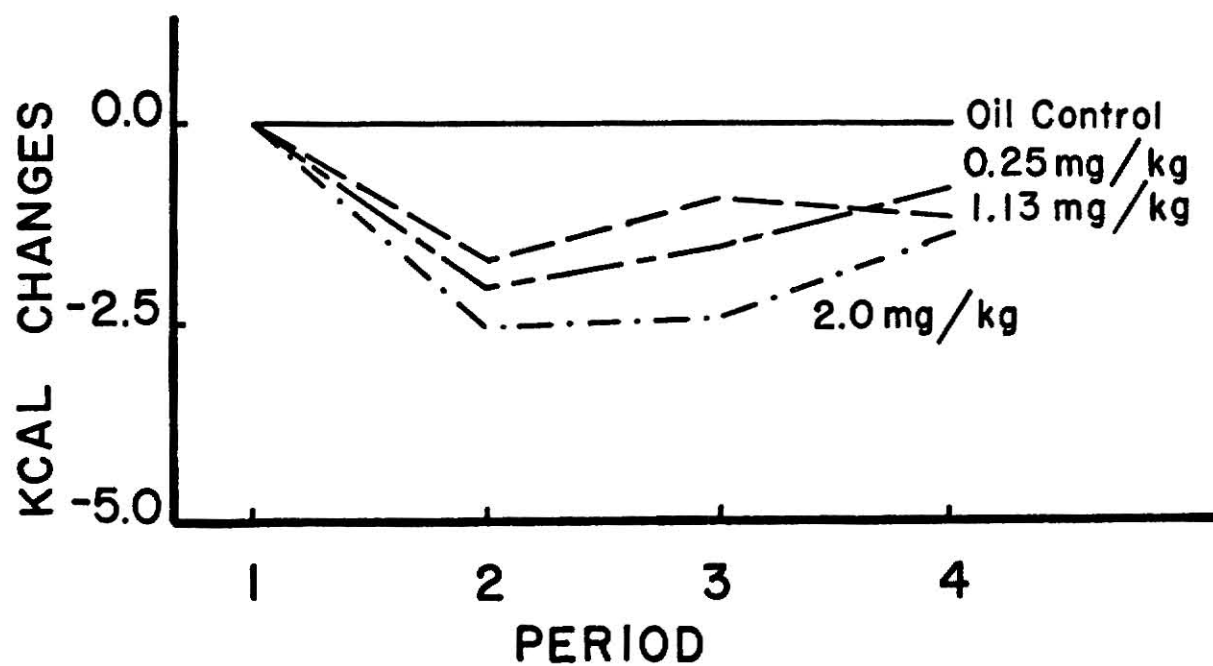
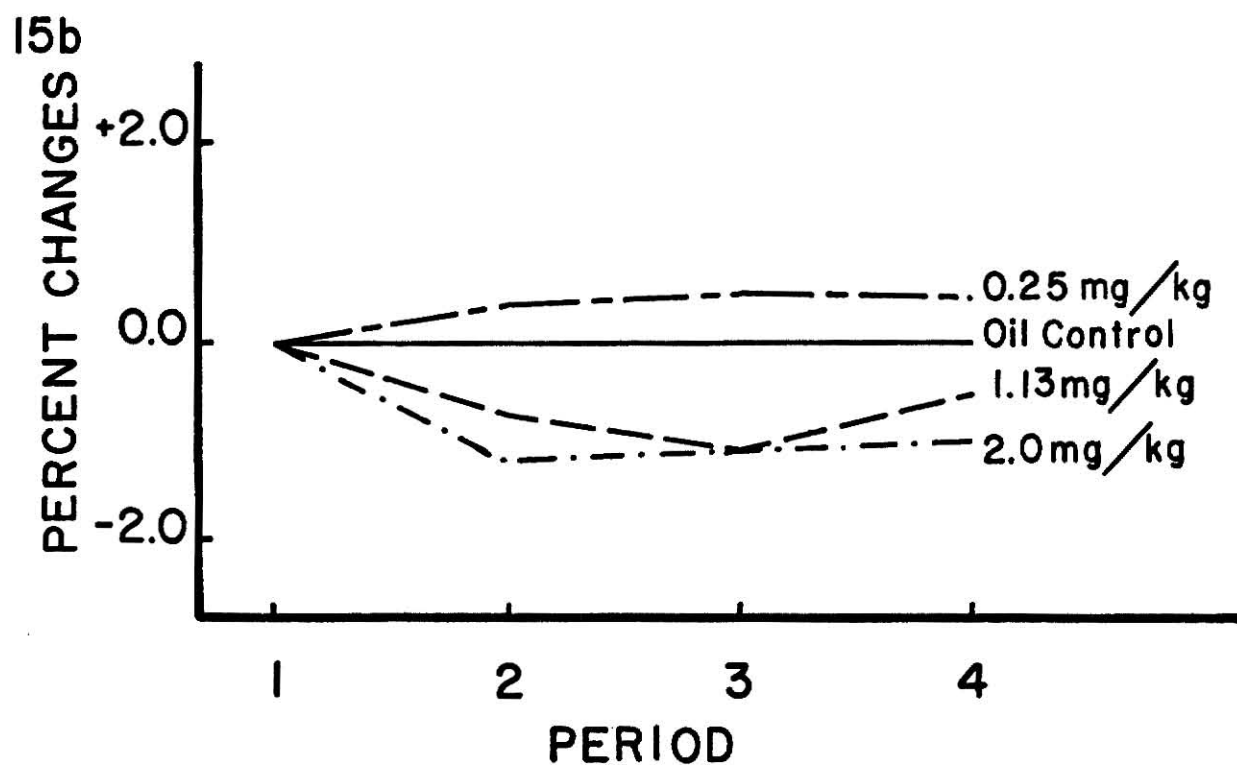
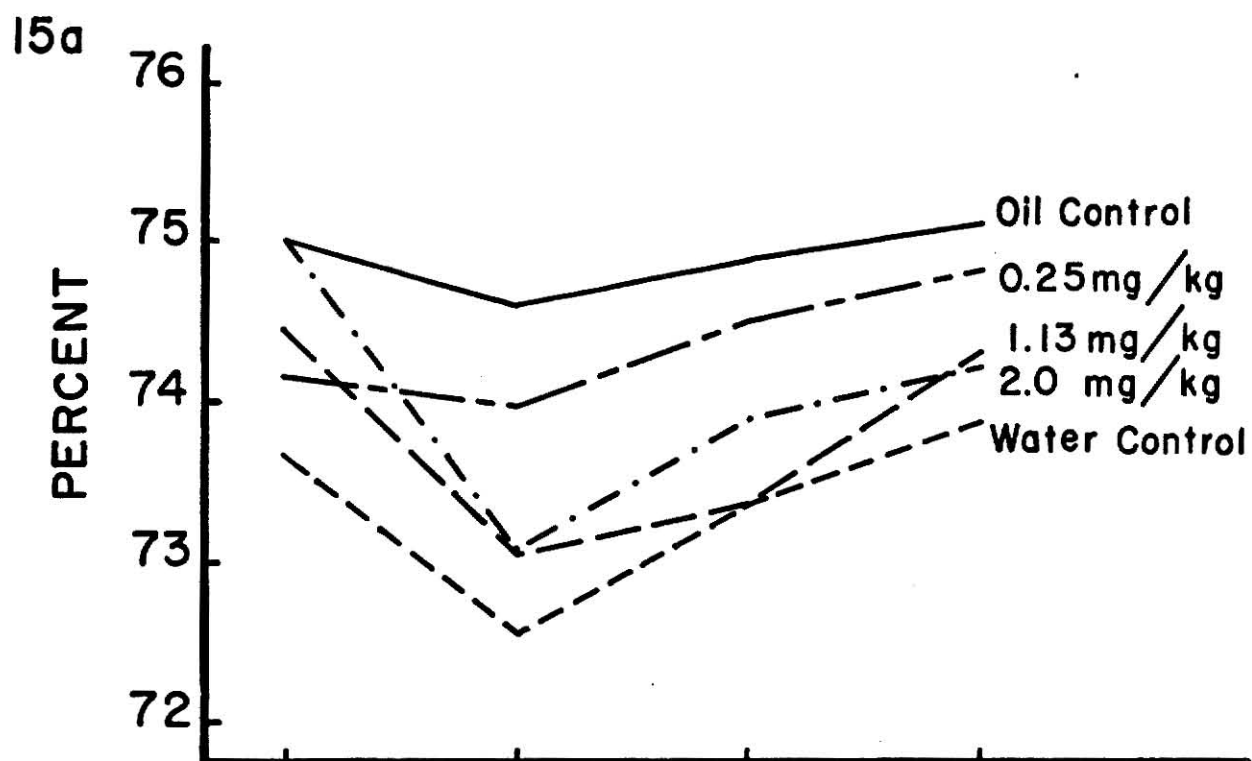




Fig. 15a. Efficiency of metabolism of bobwhites treated with Furadan<sup>®</sup>, and/or corn oil, and water during periods 2 to 4. Quail were not treated during period 1.

15b. Efficiency of metabolism changes of Furadan<sup>®</sup> treated quail in relation to the corn oil control.

## EFFICIENCY OF METABOLISM (FURADAN)





The low Furadan dosed quail, in relation to the corn oil control (Fig. 15b), increased efficiency of metabolism, while birds dosed with higher levels decreased efficiency of metabolism. The overall (period 1-4) efficiency changes for the low, medium, and high Furadan dosed quail were +0.45, -0.75, and -1.12 percent, respectively. These were not statistically different from the control (Table 10, appendix).

#### Fecal Energy

The amount of energy in the feces of the water control quail and corn oil control quail remained relatively constant throughout the study (3.249 kcal and 3.225 kcal, respectively). The low, medium, and high Furadan quail increased fecal energy significantly (period 2) by 0.081 kcal, 0.115 kcal, and 0.160 kcal, respectively (Fig. 16a). The three Furadan dosed quail groups decreased fecal energy significantly during period 2 to 3.

In relation to the corn oil control (Fig. 16b), the medium and high Furadan treatments increased fecal energy significantly at period 2 (0.104 kcal and 0.149 kcal, respectively). The overall means of the medium and high treatments were significantly greater than the corn oil control (0.071 kcal and 0.110 kcal, respectively) and significantly different themselves (Table 10, appendix).

#### Corn Oil Effects and Metabolism

Corn oil control and water control data were used to determine corn oil effects on energetic variables, and to estimate efficiency of corn oil metabolism by bobwhites. Figures 17a, b, c, d, e, and f were determined using the equation on page 15. The corn oil control changes were calculated in relation to the water control.

Gross energy intake, metabolized energy, and excretory energy were significantly ( $P < .05$ ) increased by 0.5 ml corn oil doses (Fig. 17a, and c). The overall

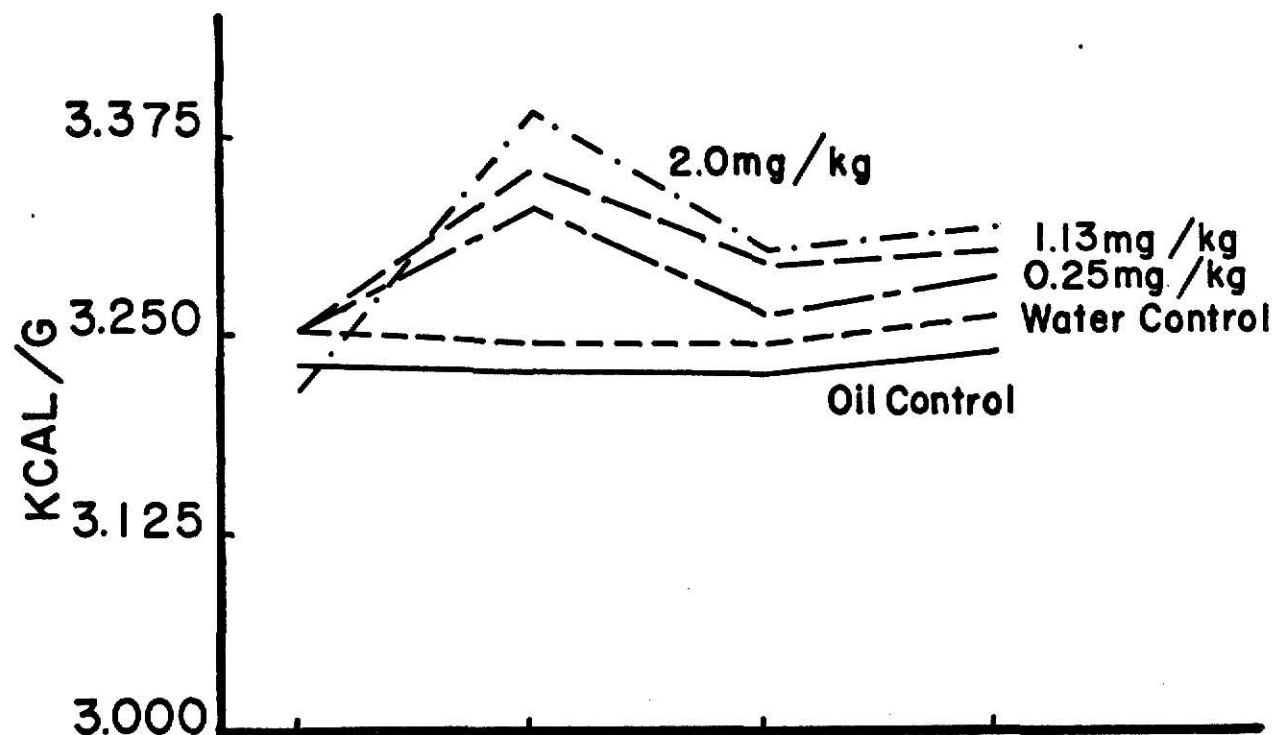


Fig. 16a. Fecal energy of bobwhites treated with Furadan<sup>®</sup>, and/or corn oil, and water during periods 2 to 4. Quail were not treated during period 1.

16b. Fecal energy changes of Furadan<sup>®</sup> treated quail in relation to the corn oil control.

## FECAL ENERGY (FURADAN)

16a



16b

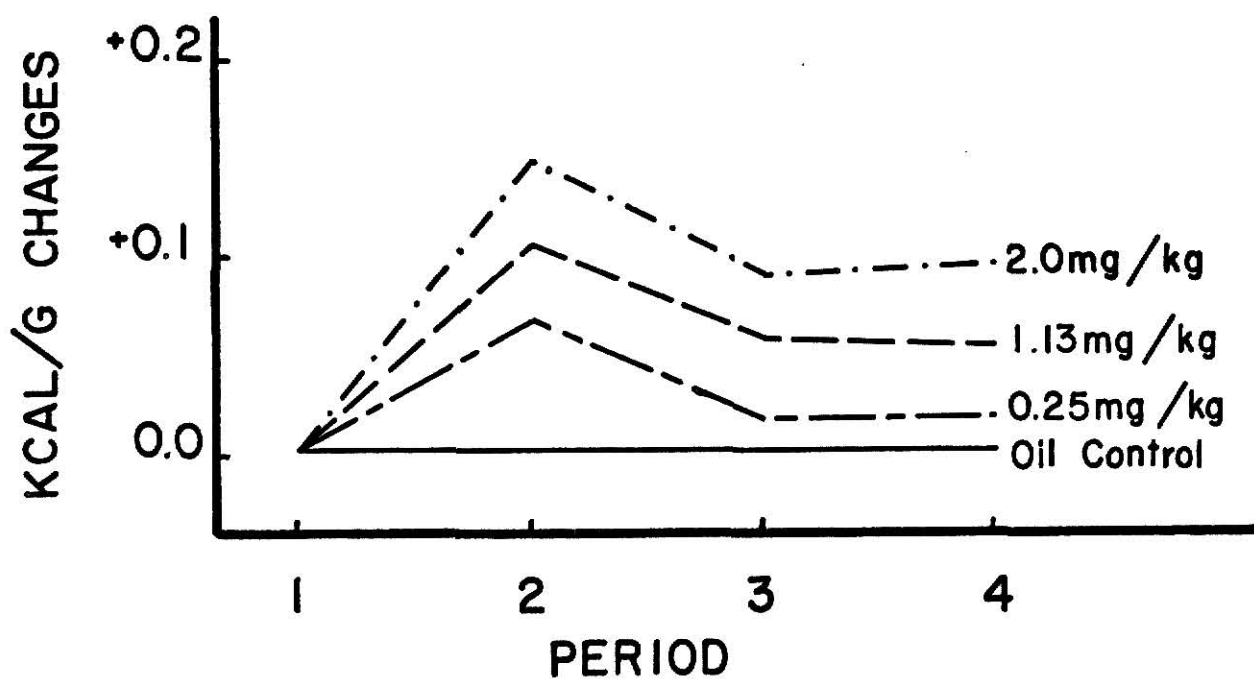
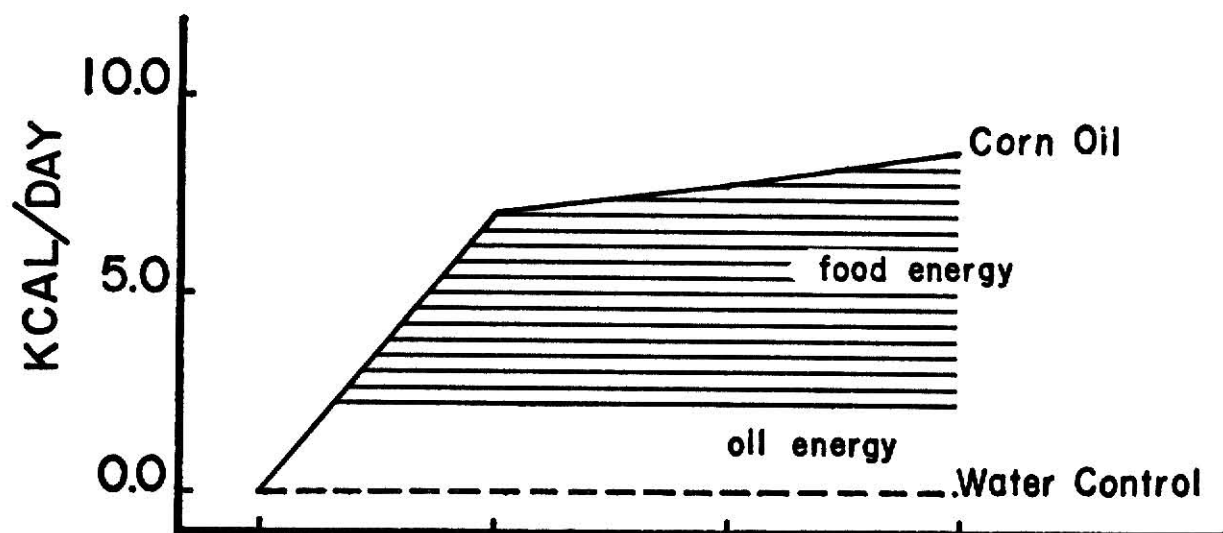




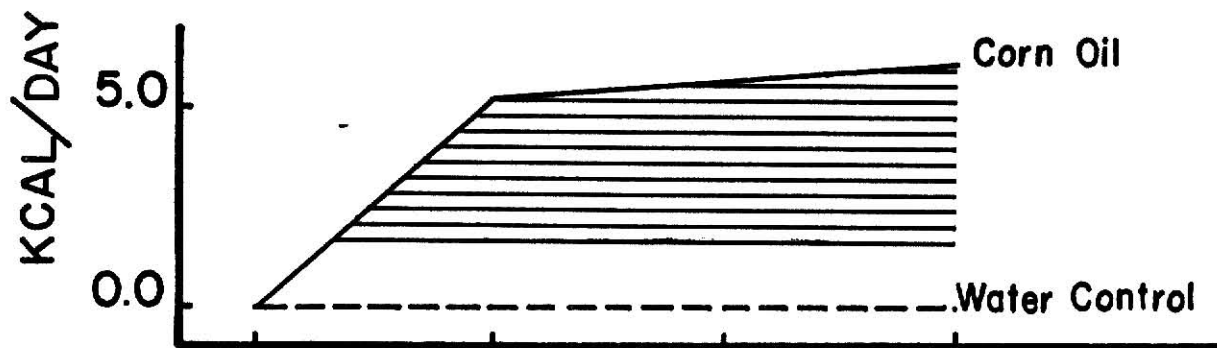
Fig. 17a-f. The effects of 0.5 ml doses of corn oil on the energetic variables, in relation to the water control. Quail were not treated during period 1. Each period was 2 days.

- 17a. Gross Energy
- 17b. Metabolized Energy
- 17c. Excretory Energy
- 17d. Efficiency of Metabolism
- 17e. Fecal Energy
- 17f. Weight

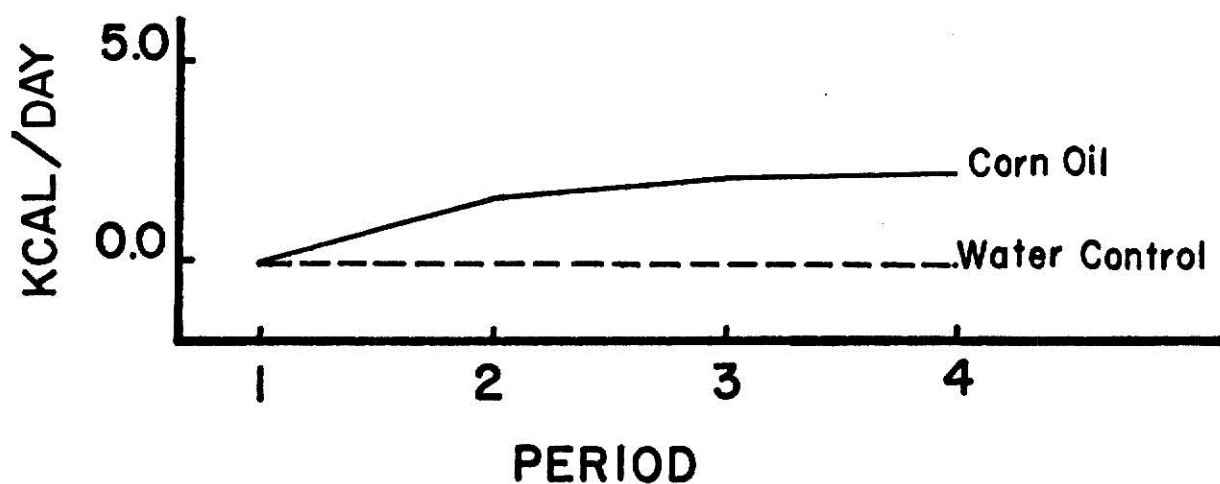
## 17a GROSS ENERGY INTAKE



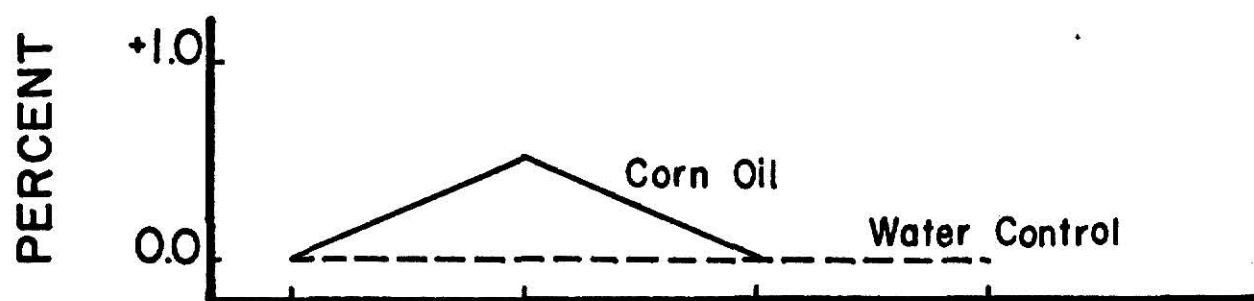
## 17b METABOLIZED ENERGY



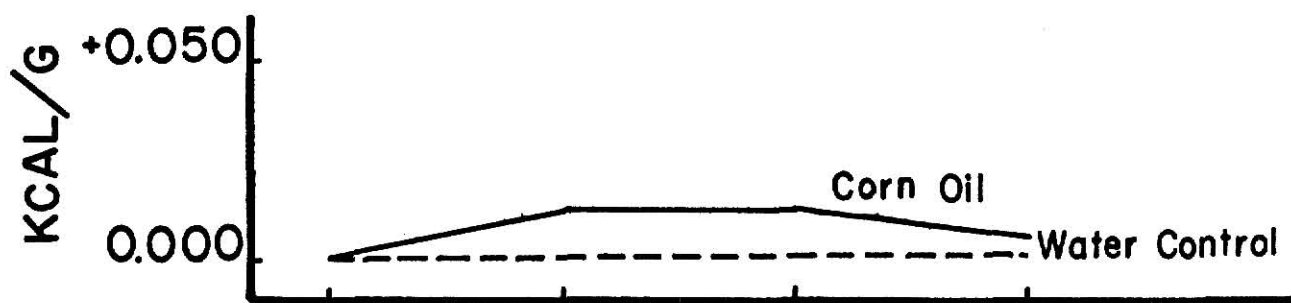
## 17c EXCRETORY ENERGY



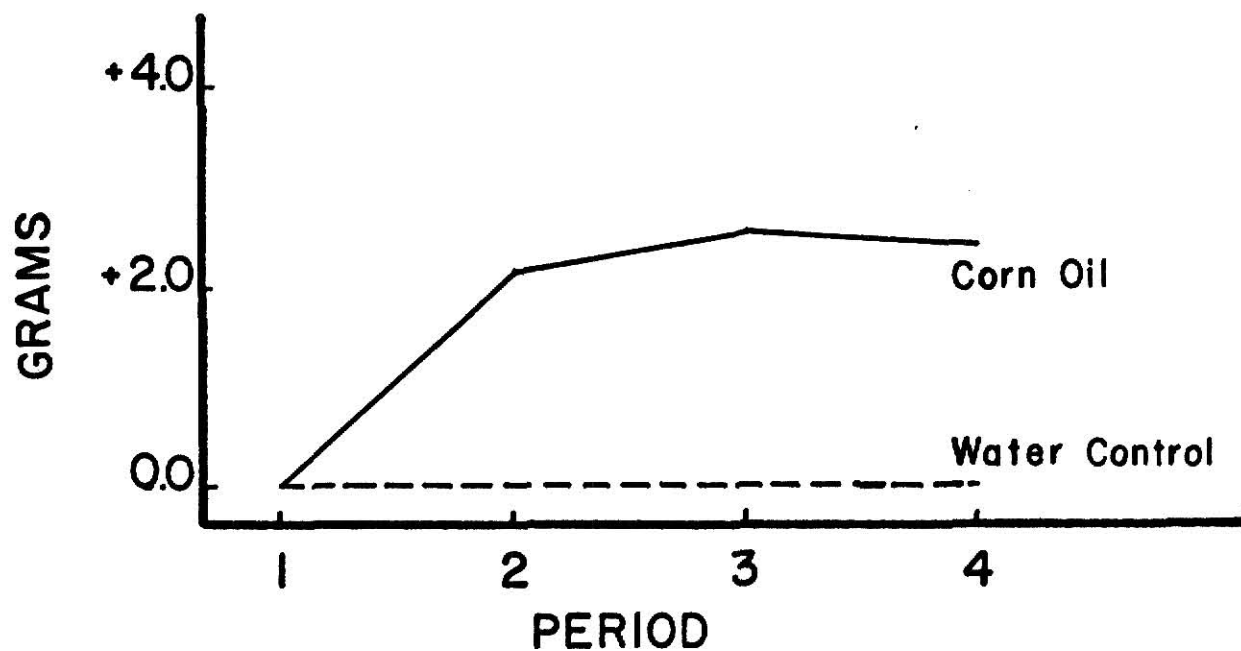
## 17d EFFICIENCY OF METABOLISM



## 17e FECAL ENERGY



## 17f WEIGHT





(period 1-4) increases in gross energy intake, metabolized energy, and excretory energy were 7.951 kcal, 5.951 kcal, and 2.042 kcal, respectively. No significant corn oil effects were observed on efficiency of metabolism (+0.22 percent, Fig. 17d) or fecal energy (+0.010 kcal, Fig. 17e).

For determining the efficiency of corn oil metabolism it was noted that the increase in gross energy intake, 7.951 kcal/day, was greater than the kilocalories available from the corn oil, 2.254 kcal/day. The shaded area under the gross energy curve (Fig. 17a) is therefore the added food intake (7.951 kcal-2.254 kcal = 5.697 kcal) above that supplied by the corn oil. Because the corn oil had no effect on efficiency of metabolism, the increase (5.697 kcal) of gross energy intake above that supplied by the corn oil was metabolized at the control period 1 efficiency of 74.48 percent. The energy metabolized from the 5.697 kcal food energy was 4.243 kcal ( $.7448 \times 5.697$  kcal). Subtraction of the 4.243 kcal from the total metabolized energy of 5.591 kcal yields the metabolized corn oil of 1.708 kcal. The 1.708 metabolized corn oil is then divided by the total energy available from the corn oil, 2.254 kcal, to determine the efficiency of corn oil metabolism, 75.78 percent.

#### Brain Cholinesterase Determinations

Two brain cholinesterase determinations were performed on each of the first 12 brain samples. The mean difference between duplicated determinations was 3.08 Rappaport units (7.46 percent). The mean Rappaport units for all quail groups are listed in Table 3.

#### Sevin® Results

A one-way analysis of variance of Sevin® control and treatment Rappaport units showed nonsignificant treatment differences ( $P=0.0536$ ). The Rappaport

Table 3. Mean Rappaport Units of brain cholinesterase for bobwhites treated with Sevin<sup>R</sup> and Furadan<sup>R</sup>. The quail were dosed every 2 days.

<u>SEVIN</u>		<u>FURADAN</u>	
Treatment	Mean $\pm$ S.D.	Treatment	Mean $\pm$ S.D.
Controls		Controls	
nothing <sup>1</sup>	47.25 $\pm$ 6.80		40.75 $\pm$ 5.62
water <sup>2</sup>		(0.5 ml)	39.00 $\pm$ 5.35
corn oil <sup>2</sup> (1.0 ml)	32.25 $\pm$ 5.68	(0.5 ml)	43.50 $\pm$ 5.74
10 mg/kg		0.25 mg/kg	
dose 1	39.50 $\pm$ 2.89		38.50 $\pm$ 4.43
dose 2	43.00 $\pm$ 5.72		37.75 $\pm$ 5.06
dose 3	44.00 $\pm$ 13.04		39.25 $\pm$ 5.50
dose 4	39.75 $\pm$ 7.93		41.75 $\pm$ 3.95
50 mg/kg		1.13 mg/kg	
dose 1	39.00 $\pm$ 7.30		39.00 $\pm$ 4.16
dose 2	52.00 $\pm$ 7.07		39.50 $\pm$ 4.95
dose 3	36.67 $\pm$ 6.43		39.50 $\pm$ 4.20
dose 4	41.25 $\pm$ 5.12		39.75 $\pm$ 2.06
90 mg/kg		2.0 mg/kg	
dose 1	42.25 $\pm$ 4.35		35.75 $\pm$ 7.14
dose 2	33.25 $\pm$ 4.50		41.00 $\pm$ 5.48
dose 3	43.50 $\pm$ 2.12		45.25 $\pm$ 5.85
dose 4	44.00 $\pm$ 5.66		46.50 $\pm$ 4.93
1057 mg/kg <sup>3</sup>	16.00		
1140 mg/kg <sup>3</sup>	7.00		
1225 mg/kg <sup>3</sup>	6.67		
1348 mg/kg <sup>3</sup>	12.50		

<sup>1</sup> Quail were killed at the beginning of the study.

<sup>2</sup> Quail received four dosings, one every 2 days.

<sup>3</sup> Quail killed by Sevin in the LD<sub>50</sub> study.

units ranged from 32.25 units for the corn oil control, to 52.00 units for the second dose of 50 mg/kg Sevin. The corn oil control was significantly lower than those quail killed at the beginning of the study (47.25 units).

Cholinesterase determinations were also performed on the quail which died of insecticidal poisoning during the Sevin LD<sub>50</sub> study. The Rappaport unit range was 0.0 to 25.0 (7 quail). The mean ( $\pm$  standard deviation) of  $9.71 \pm 9.14$  units was significantly below that of the energetic study's corn oil control ( $32.25 \pm 5.68$ ).

#### Furadan® Results

A one-way analysis of variance of Furadan® control and treatment Rappaport units showed nonsignificant treatment differences ( $P=0.2814$ ). The Rappaport units ranged from 35.75 units for the first dose of 2.0 mg/kg Furadan, to 46.50 units for the fourth dose of 2.0 mg/kg Furadan. No significant differences were found between the three controls: water doses, 39.00; corn oil doses, 43.50; and no doses, 40.75 units. No significant differences were found between the controls and the Furadan treatments.

## DISCUSSION

### Sevin® LD<sub>50</sub>

Even though the dosage levels and number of quail used to determine the bobwhite LD<sub>50</sub> of Sevin® were less than recommended by Heath and Stickel (1965), i.e. 4 dosage levels instead of 6 and 4 birds per treatment instead of 10, the probit-log dose regression provided a strong correlation coefficient ( $r=0.94$ ) which was significant ( $P=0.0169$ ). The Sevin LD<sub>50</sub> for bobwhites in this study was 1193 mg/kg, substantially less than the 2000 mg/kg and 2179 mg/kg, respectively, for pheasants and mallards as reported by Tucker and Crabtree (1970). These data substantiate the findings of Heath et al. (1972) that quail are more susceptible to certain insecticidal poisonings than pheasants and mallards. Heath et al. showed that the carbamates Baygon® and methomyl are more toxic to bobwhites than to mallards, and that Ortho 11775 is more toxic to bobwhites than to mallards and pheasants (all were chronic LC<sub>50</sub>).

### Energetics

Bobwhites gaining more than 1.0 percent body weight, between test periods, were not used in data analysis. Energetic variable measurements are based on the existence requirements of an organism. If the organism gains weight, its energy intake is used for both existence and construction of body tissue. This construction energy must be removed so that existence requirements are measured. The energetic variables for weight-loss quail were corrected according to Clement (1970). There were eight weight-gain quail during the Sevin study and eight in the Furadan® study. If the insecticides had increased all quail weights then the data could not have been analyzed. Before further insecticide-energetic studies are conducted, it would be beneficial to determine correction

factors for both weight-loss and weight-gain test animals. Kendiegh et al. (1969) determined for sparrows (Passer domesticus) the energy cost of constructing body tissue, and the energy gained from tissue metabolism.

Corn oil was used as the insecticide carrier since it formed a suspension with Sevin and Furadan, and it was assumed to be readily metabolized. An oily, yellow feces was excreted by the corn oil controls and the Sevin and Furadan treatment groups 1.5 to 2.0 hours after being dosed. Bisset (1972) determined that 1.5 to 2.0 hours are required for bobwhites to pass food through the alimentary tract. Since the quail did not have access to food for 2 hours after being dosed, the oily feces probably consisted of unmetabolized corn oil, with some fecal and intestinal wastes. The efficiency with which corn oil was metabolized by quail was estimated during the Furadan study at 75.78 percent.

Due to this efficiency of corn oil metabolism, the corn oil's effects on energetic variables (gross energy intake, metabolized energy, excretory energy, and weight), and the caloric content of corn oil, future insecticide-energetic studies should use a more suitable carrier. However the influence of the oil on certain energetic variables did not hinder the determination of Sevin and Furadan energetic effects since the actual caloric data were converted to ratio data then to caloric changes relative to the corn oil control.

#### Sevin<sup>®</sup> Energetics

The quail were weighed every two days, before the study began, to acclimate the birds to the handling. The quail were not accustomed to the insertion of the syringe, and dose, into the crop. Therefore the greatest energetic variable changes were noted with the first treatments. After the first dose all quail began to recover toward pretreatment (period 1) control values even though they were dosed a second and third time. This is seen in the significant period effect for gross energy intake, metabolized energy, excretory energy, efficiency

of metabolism, and fecal energy. The nonsignificant treatment effect showed that the majority of the energetic variable responses were due to the corn oil.

Feces energy was the only variable to increase after the first dosings. Since the Sevin treatments showed no significant effect on fecal energy, the greatest portion of the fecal energy increase was probably due to the unmetabolized corn oil. The efficiency of corn oil metabolism was estimated to be 75.78 percent, therefore, of the total energy available in the corn oil (9.049 kcal/2 days) 2.192 kcal would be excreted. The mean increase of fecal energy at period 2 was 0.440 kcal/g for the 5.25 grams of feces produced during that period. Therefore the total increase in excreted energy was 2.310 kcal (0.440 kcal/g X 5.25 g), almost identical to the estimated unmetabolized corn oil energy of 2.192 kcal/2 days.

Because of the increase in fecal energy (period 2), the excretory energy would be expected to increase. However, this was not the case; excretory energy decreased nonsignificantly (Fig. 7a). This decrease was due to decreases in gross energy intake and metabolized energy. Changes in excretory energy were highly correlated with changes in gross energy intake and metabolized energy ( $r = 0.8384$  and  $r = 0.7169$ , respectively).

After the corn oil effects were removed, by converting energetic variables to ratios then to caloric changes, no significant Sevin effects were noted. The 10 mg/kg Sevin treatment increased gross energy intake, significantly, and metabolized energy, and excretory energy nonsignificantly. Paulson and Feil (1969) found no change in white leghorn food consumption when birds received 10 mg/kg Sevin.

Bobwhites receiving the highest dosages (90 mg/kg) in this study experienced a nonsignificant decrease in food intake. Nir et al. (1966) dosed white leghorn hens daily with 180 and 540 mg/kg Sevin for 60 days. After 21 days, the food consumption of the 180 mg/kg and 540 mg/kg dosed hens was reduced by

10 g/day and 30 g/day, respectively. Likewise, Haegelle and Tucker (1974) found that coturnix decreased food consumption following a single oral dose of 1,000 mg/kg Sevin. The dosage levels used in these two studies were higher than what a wild bird might be expected to encounter in the field.

The three Sevin treatments lowered the bobwhites' efficiency of metabolism nonsignificantly. McDonald et al. (1964) fed malathion to white leghorns and found that their ability to convert feed into flesh had decreased; control birds required 3.95 pounds of feed to produce a pound of flesh, while malathion treated birds needed 4.04 pounds of feed per pound flesh. Likewise, Sherman and Ross (1969) treated coturnix with the carbamate SD 8350, and found a decreased food conversion factor; the controls required 4.35 pounds of feed for a pound weight gain, and the SD 8350 (400 ppm) treated birds needed 4.44 pounds feed per pound gain.

The bobwhite's physiological controls of energy intake and utilization seem not to be effected by repeated sublethal doses of Sevin. Lustick et al. (1972) noted that DDT fed bobwhites experienced increased thyroid activity through increased thyroxin secretion (hyperthyroidism). This increased secretion raised the bobwhites basal metabolic rate. The thyroid, which plays an important role in metabolism, is also affected by doses of Sevin. Kusevitskiy et al. (1970) treated rats with Sevin (2.5 and 15 g/kg feed) and found decreased thyroid activity. Shtenberg and Rybakova (1969) noted decreased thyroid activity in Sevin treated rats, but stressed that the deleterious effects were exerted on the adenohypophysis of the pituitary. Shtenberg and Khovaeva (1970) stated that Sevin decreased the thyrotropic hormone activity of the adenohypophysis and caused possible lesions to the hypothalamus in rats.

#### Furadan® Energetics

During the Furadan study, corn oil increased quail gross energy intake,

metabolized energy, and excretory energy. The use of the corn oil-insecticide suspension seemed to suppress these increases. When considering the energetic variables in relation to the corn oil control, decreases were observed. The nonsignificant treatment-period interaction for all energetic variables, showed no differences between dosing the quail one, twice, or three times with Furadan. The overall treatment effect was significant for gross energy intake, metabolized energy, body weight, and fecal energy. The medium (1.13 mg/kg) and high (2.0 mg/kg) Furadan doses caused significant decreases in gross energy intake, metabolized energy and weight, while the low (0.25 mg/kg) Furadan dose decreased these variables nonsignificantly.

The greatest decrease in food consumption and metabolized energy occurred after the first oral dosings. With continued dosing (3 times) of the quail, these variables increased to pretreatment values in 6 days (period 5). Sherman and Ross (1969) found that coturnix decreased food consumption when fed Furadan treated feed. Food intake decreased with increased Furadan levels, and the greatest decrease was noted the first day of the study. Whether decreases in food consumption were due to Furadan's palatability or physiological effects on the birds was not determined.

The food intake of the Furadan treated bobwhites decreased (period 2) with increased dosage levels. Grossman (1962;880) found that cholinergic substances when placed on the hypothalamus "neural elements" caused rigorous drinking in satiated rats and lowered food intake in hungry rats. Furadan, being an anticholinesterase, allows acetylcholine, a cholinergic substance, to accumulate at synaptic junctions. The three Furadan dosage levels may have caused graded responses of the hypothalamus, thus caused graded food intake responses by the bobwhites.

The only variable to increase significantly (period 2) was fecal energy. The greater the Furadan dose used, the greater was the increase in fecal energy.



The fecal energy of the Sevin control increased with a 1.0 ml corn oil dose, but the Furadan corn oil control showed no change with 0.5 ml corn oil dose. Significant treatment effects were observed in the Furadan study fecal energy (overall increase of 0.072 kcal/g) and not the Sevin study fecal energy (overall increase of 0.158 kcal/g).

The metabolic efficiency of bobwhites was not affected by Furadan. Sherman and Ross (1969) noted a decrease in efficiency when coturnix were treated with Furadan, assuming that food conversion factors (units of food needed to produce one unit weight gain) are indicative of metabolic efficiency. The conversion factors for the control and Furadan quail (400 ppm in feed) were 4.35 and 7.07, respectively. This decrease of coturnix efficiency of metabolism may have been due to the high Furadan dosage. The coturnix consumed approximately 13.15 mg/kg (body weight) Furadan a day, while the bobwhites, of my energetics study, received no more than 2.0 mg/kg.

Furadan probably did not increase the basal metabolic rate (BMR) of the bobwhites. The Furadan dosed quail lost 2.67 g body weight (mean of all quail) during period 2 while the quails' metabolized energy decreased 6.264 kcal. The quail therefore lost 0.426 g/kcal metabolized energy decrease. Linduska and Springer (1957) found that bobwhites decreased food consumption when given DDT treated feed; while Lustick et al. (1972) found that DDT treated feed increased bobwhite BMR. If the BMR of the Furadan treated quail had increased, while the metabolized energy decreased, the factor of 0.426 would be greater than that for untreated quail. Bisset (1972) fed bobwhites various seed diets and noted the metabolizable energy of each diet (P-18 control) and the body weight changes. Using Bisset's data, it was calculated that 0.405 g body weight was lost per kilocaloric decrease in metabolized energy. The 0.405 factor for Bisset's untreated quail is similar to the 0.426 of the Furadan treated quail.

### Cholinesterase Determinations

The purposes of the cholinesterase study were to determine if the carbamates Sevin® and Furadan® exhibited cumulative anticholinesterase activity change, and if the overall treatment levels could be correlated with cholinesterase inhibition. No treatment effects were observed in either the Sevin or Furadan studies.

The quail of the Sevin and Furadan energetic studies were sacrificed 48 hours after the insecticidal dose. The nonsignificant treatment effects of Sevin and Furadan may be due to the rapidly reversible inhibition of brain cholinesterase. Carpenter et al. (1961) dosed rats orally with Sevin and found that both erythrocyte cholinesterase and brain cholinesterase levels returned to normal after 24 hours. Vulchovski (1972) using white leghorn chickens dosed with Sevin (250 mg/kg and 500 mg/kg orally) found blood cholinesterase levels returned to normal at 36 hours. Baron et al. (1964) showed that carbamate brain cholinesterase inhibition of mice returned to normal 1 to 24 hours after treatment. Baron et al. (1966) found that 60 mg/kg Sevin given to rats caused maximum brain cholinesterase inhibition 10 minutes after treatment, with little inhibition recorded after 4 hours. Thus sacrificing bobwhites 48 hours after an insecticidal dose was too late to record any anticholinesterase activity, even after repeated insecticide doses. The only significant reduction of brain cholinesterase occurred with quail that died of Sevin poisoning during the LD<sub>50</sub> study. Upon death, these quail were frozen and anticholinesterase activity was noted.

## CONCLUSIONS

Based on data gathered during this study the following conclusions appear justified:

1. Sevin<sup>®</sup> dosages of 10, 50, and 90 mg/kg (0.83 percent, 4.19 percent, and 7.54 percent of LD<sub>50</sub>, respectively) did not significantly alter bobwhite gross energy intake, metabolized energy, excretory energy, fecal energy, efficiency of metabolism, or body weight.
2. Initial exposure to 0.25, 1.13, and 2.0 mg/kg Furadan<sup>®</sup> significantly decreased bobwhite gross energy intake, and metabolized energy, while 1.13 and 2.0 mg/kg Furadan decreased body weight and increased fecal energy significantly.
3. Furadan treatments did not cause cumulative effects on bobwhite gross energy intake, metabolized energy, excretory energy, fecal energy, or weight. Quail recovered from initial effects of Furadan in 6 days (even when being dosed these 6 days).
4. Corn oil in 0.5 ml doses stimulated bobwhite food intake, metabolized energy, and weight gain. The use of corn oil as the insecticide carrier caused greater energetic variable changes than the insecticide, Sevin.
5. Sublethal levels of Sevin and Furadan used in this study did not cause accumulative or overall brain cholinesterase inhibition in bobwhites.
6. Agricultural utilization of the nonpersistent, carbamate insecticides, Sevin and Furadan, is not detrimental from a bobwhite energetic view point.

## ACKNOWLEDGEMENTS

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## APPENDIX

Table 4. Caloric content of feed and corn oil used  
in the carbaryl and carbofuran studies.

SEVIN STUDY		FURADAN STUDY	
Food samples			
Period	kcal/g	Period	kcal/g
1	4.368	1	4.282
1	4.327	1	4.277
3	4.353	3	4.262
3	4.352	3	4.276
5	4.352	5	4.259
5	4.348	5	4.292
Mean	4.350	Mean	4.275
S.D.	$\pm 0.013$	S.D.	$\pm 0.013$
Corn Oil Samples			
1	9.423	1	9.402
2	9.432	2	9.389
Mean	9.428 or 9.049 kcal/ml	Mean	9.395 or 4.509 kcal/0.5ml

Table 5a. Degrees of freedom and mean squares of two-way analysis of variance for Sevin<sup>R</sup> study.

SEVIN Actual Data									
	df	GROSS ENERGY MS	METABOLIZED ENERGY MS	EFFICIENCY OF METABOLISM MS	BODY WEIGHT MS	EXCRETORY ENERGY MS	FECAL ENERGY MS		
Treatment	3	123.45	69.07	11.28	2179.51*	9.780	.0691		
Bird/treatment	24	45.81	20.53	5.74	641.88	5.042	.0187		
Period	3	443.48**	341.21**	59.27**	6.43**	8.466**	.8591**		
Trt·period interaction	9	27.80*	16.03*	2.89	1.12	1.704	.0082		
Residual	64	10.69	6.12	2.79	1.36	1.001	.0079		
SEVIN Ratio Data									
Treatment	3	.0395	.0482	.0027	.00012	.0347	.0059		
Bird/treatment	24	.0346	.0431	.0021	.00016	.0374	.0039		
Period	2	.2143**	.3093**	.0111**	.00007	.0510**	.0153**		
Trt·period interaction	6	.0095	.0068	.0004	.00002	.0175	.0002		
Residual	42	.0053	.0055	.0005	.00006	.0089	.0006		

\* Mean squares (treatment, period, or trt·period interaction) which yield significant (P<.05) F values.

\*\* Mean squares (treatment, period, or trt·period interaction) which yield highly significant (P<.01) F values.

Table 5b. Degrees of freedom and mean squares of two-way analysis of variance of Furadan<sup>R</sup> study.

<u>FURADAN Actual Data</u>									
	df	GROSS ENERGY MS	METABOLIZED ENERGY MS	EFFICIENCY OF METABOLISM MS	BODY WEIGHT MS	EXCRETORY ENERGY MS	FECAL ENERGY MS		
Treatment	4	59.042	36.187	4.427	357.27	4.230	.0172		
Bird/treatment	27	30.895	18.589	2.749	640.21	2.301	.0055		
Period	3	109.173**	67.892**	4.541**	4.71**	5.110**	.0189**		
Trt*period interaction	12	19.828**	11.552**	0.824	2.49**	1.036**	.0039		
Residual	72	8.034	4.608	0.778	0.82	0.710	.0020		

<u>FURADAN Ratio Data</u>									
	df	GROSS ENERGY MS	METABOLIZED ENERGY MS	EFFICIENCY OF METABOLISM MS	BODY WEIGHT MS	EXCRETORY ENERGY MS	FECAL ENERGY MS		
Treatment	4	.0940*	.1061*	.00125	.000604**	.0711	.00339**		
Bird/treatment	27	.0239	.0253	.00065	.000120	.0323	.00031		
Period	2	.0605**	.0485**	.00114**	.000113	.0221*	.00093*		
Trt*period interaction	8	.0074	.0062	.00004	.000024	.0083	.00009		
Residual	46	.0047	.0048	.00014	.000037	.0063	.00027		

\* Mean squares (treatment, period, or trt\*period interaction) which yield significant ( $P < .05$ ) F ratios.

\*\* Mean squares (treatment, period, or trt\*period interaction) which yield highly significant ( $P < .01$ ) F ratios.

Table 6. Chi square analysis of the three weight classes of quail during the Sevin and Furadan studies.

SEVIN <sup>1</sup>				FURADAN <sup>1</sup>							
		Weight-loss		Weight-loss		Weight-loss					
		Quail <sup>2</sup>	Quail <sup>3</sup>	Quail <sup>2</sup>	Quail <sup>3</sup>	Quail <sup>2</sup>	Quail <sup>3</sup>				
		Weight-stable		Weight-stable		Weight-stable					
		Quail <sup>4</sup>	Quail <sup>4</sup>	Quail <sup>4</sup>	Quail <sup>4</sup>	Quail <sup>4</sup>	Quail <sup>4</sup>				
Oil Control	2 (2.71) <sup>5</sup>	2 (1.06)	12 (12.24)	16	Water Control	0 (1.14) <sup>5</sup>	3 (1.94)	13 (12.91)	16		
10 mg/kg	5 (6.76)	4 (2.65)	31 (30.59)	40	Oil Control	1 (1.14)	3 (1.94)	12 (12.91)	16		
50 mg/kg	5 (6.76)	2 (2.65)	33 (30.59)	40	0.25 mg/kg	3 (2.57)	8 (4.37)	25 (29.06)	36		
90 mg/kg	11 (6.76)	1 (2.65)	28 (30.59)	40	1.13 mg/kg	2 (2.57)	1 (4.37)	33 (29.06)	36		
	23	9	104	136	2.00 mg/kg	4 (2.57)	2 (4.37)	30 (29.06)	36		
					10	17	113	140			
Chi square = 6.915		df = 6		P>.25		Chi square = 10.263		df = 8		P>.25	
Period 1	0 (6.36) <sup>5</sup>	3 (3.32)	49 (42.32)	52	Period 1	0 (4.07) <sup>5</sup>	14 (5.74)	30 (34.20)	44		
2	19 (6.36)	1 (3.32)	32 (42.32)	52	2	9 (4.07)	6 (5.74)	29 (34.20)	44		
3	1 (4.89)	1 (2.55)	38 (32.55)	40	3	4 (4.07)	2 (5.74)	38 (34.20)	44		
4	2 (3.43)	5 (1.79)	21 (21.79)	28	4	1 (2.96)	2 (2.96)	29 (24.87)	32		
5	1 (1.96)	2 (1.02)	13 (13.02)	16	5	3 (1.85)	0 (1.85)	17 (15.54)	20		
	23	12	153	188		17	24	143	184		
Chi square = 49.557 <sup>6</sup>		df = 8		P<.005		Chi square = 32.682		df = 8		P<.005	

<sup>1</sup> The first chi square analysis does not include period 1 quail.

<sup>2</sup> Lost greater than 1.0 percent body weight.

<sup>3</sup> Gained greater than 1.0 percent body weight.

<sup>4</sup> Less than 1.0 percent body weight change.

<sup>5</sup> Expected value (column total X row total ÷ grand total).

<sup>6</sup> See Figure 3.

Table 7. Correlations (r) of period 1 minus period 2 ( $\Delta$ ) energetic variable changes for bobwhites treated with Sevin or Furadan.

SEVIN	$\Delta$ Gross Energy	$\Delta$ Met. Energy	$\Delta$ Efficiency of Met.	$\Delta$ Weight	$\Delta$ Excretory Energy	$\Delta$ Fecal Energy
$\Delta$ Metabolized Energy	0.9810**					
$\Delta$ Efficiency of Met.	0.2320	0.4084*				
$\Delta$ Weight	0.6543**	0.6099**	0.0399			
$\Delta$ Excretory Energy	0.8384**	0.7169**	-0.3139	0.6388**		
$\Delta$ Fecal Energy	-0.0461	-0.1662	-0.7200**	-0.1077	0.3016	
FURADAN						
$\Delta$ Gross Energy		0.9872**	0.1606	0.7769**	0.9199**	-0.1099
$\Delta$ Metabolized Energy			-0.0062	0.7537**	0.8459**	-0.2229
$\Delta$ Efficiency of Met.				-0.2377	-0.5225**	-0.6648**
$\Delta$ Weight					0.7487**	-0.1547
$\Delta$ Excretory Energy						0.1835

\* Significant,  $P < 0.05$

\*\* Highly significant,  $P < 0.01$

Table 8. Energetic variable changes, in relation to the corn oil control, of bobwhites dosed with Sevin<sup>R</sup>.

Period:	2	3	4	Mean <sup>1</sup>	2	3	4	Mean <sup>1</sup>
	GROSS ENERGY INTAKE (kcal)				EFFICIENCY OF METABOLISM (%)			
Oil Control	0.000	0.000	0.000	0.000 a	0.00	0.00	0.00	0.00 a
10 mg/kg	+4.275	+0.968	-1.936	+1.089 a	-1.35	+0.15	-2.18	-1.13 a
50 mg/kg	-1.210	-1.049	-1.895	-1.371 a	-1.43	+0.98	-1.20	-0.53 a
90 mg/kg	-3.428	-2.016	-3.428	-2.944 a	-2.18	-1.66	-2.93	-2.26 a
	METABOLIZED ENERGY (kcal)				FECAL ENERGY (kcal/g)			
Oil Control	0.000	0.000	0.000	0.000 a	0.000	0.000	0.000	0.000 a
10 mg/kg	+2.212	+0.970	-2.242	+0.303 a	+0.136	+0.126	+0.194	+0.152 a b
50 mg/kg	-1.121	-0.273	-1.757	-1.060 a	+0.185	+0.146	+0.175	+0.168 b
90 mg/kg	-3.000	-2.091	-3.757	-2.969 a	+0.139	+0.159	+0.172	+0.155 a b
	EXCRETORY ENERGY (kcal)							
Oil Control	0.000	0.000	0.000	0.000 a				
10 mg/kg	+1.597	+0.160	+0.399	+0.718 a				
50 mg/kg	-0.080	-0.689	+0.020	-0.249 a				
90 mg/kg	-0.349	+0.130	+0.399	+0.060 a				

<sup>1</sup> Means sharing a common letter are not significantly ( $P>0.05$ ) different.

Table 9. Correlation coefficients (r) of the Furadan treated quails' energetic variables.

CORRELATIONS <sup>1</sup>		Oil Control	0.25 mg/kg	1.13 mg/kg	2.0 mg/kg
Gross Energy	- Metabolized E.	0.9951**	0.9960**	0.8894**	0.9910**
Intake	- Efficiency	-0.1597	0.0484	0.0580	0.3216
	- Weight	0.5134	-0.0142	0.2789	0.2367
	- Excretory E.	0.9635**	0.9647**	0.8308**	0.9119**
	- Fecal E.	0.3373	0.0475	-0.2177	-0.2627
Metabolized Energy	- Efficiency	-0.0619	0.1363	0.4460	0.4445*
	- Weight	0.4945	0.0073	0.4418	0.2634
	- Excretory E.	0.9323**	0.9374**	0.6089**	0.8493**
	- Fecal E.	0.2717	0.0234	-0.3188	-0.3667*
Efficiency of Met.	- Weight	-0.1992	0.1896	0.3900*	0.2701
	- Excretory E.	-0.4166	-0.2138	-0.4349*	-0.0928
	- Fecal E.	-0.6882**	-0.2922	-0.5152**	-0.8285**
Weight	- Excretory E.	0.5389*	-0.0772	0.1067	0.1312
	- Fecal E.	0.4253	0.2895	0.3213	-0.2869
Excretory Energy	- Fecal E.	0.4983	0.1167	0.1130	0.0880

\* Significant, P<.05      \*\* Highly significant, P<.01

<sup>1</sup> For each treatment group, all data (periods 1-4) were combined and analyzed.



Table 10. Energetic variable changes, in relation to the corn oil control, of bobwhites dosed with Furadan<sup>R</sup>.

Period:	2	3	4	Mean <sup>1</sup>	2	3	4	Mean <sup>1</sup>
	GROSS ENERGY INTAKE (kcal)				EFFICIENCY OF METABOLISM (%)			
Oil Control	0.000	0.000	0.000	0.000 a	0.00	0.00	0.00	0.00 a b
0.25 mg/kg	-7.018	-5.152	-2.272	-4.787 a b	+0.37	+0.52	+0.45	+0.45 a
1.13 mg/kg	-6.856	-4.341	-4.909	-5.355 b	-0.74	-1.04	-0.30	-0.74 b
2.00 mg/kg	-10.913	-10.750	-6.329	-9.331 c	-1.19	-1.04	-0.97	-1.12 b
	NETABOLIZED ENERGY (kcal)				FECAL ENERGY (kcal/g)			
Oil Control	0.000	0.000	0.000	0.000 a	0.000	0.000	0.000	0.000 a
0.25 mg/kg	-5.045	-3.565	-1.420	-3.353 a b	+0.065	+0.013	+0.019	+0.036 a
1.13 mg/kg	-5.317	-4.139	-3.746	-4.380 b	+0.104	+0.055	+0.052	+0.071 b
2.00 mg/kg	-8.429	-8.338	-5.105	-7.281 c	+0.149	+0.087	+0.094	+0.110 c
	EXCRETORY ENERGY (kcal)				WEIGHT (g)			
Oil Control	0.000	0.000	0.000	0.000 a	0.0	0.0	0.0	0.0 a
0.25 mg/kg	-2.001	-1.617	-0.850	-1.493 a b	-1.8	-1.8	-1.1	-1.5 a b
1.13 mg/kg	-1.576	-0.974	-1.161	-1.244 a b	-2.7	-2.4	-1.6	-2.2 b
2.00 mg/kg	-2.519	-2.488	-1.275	-2.104 b	-3.5	-4.0	-2.6	-3.3 c

<sup>1</sup> Means sharing a common letter are not significantly (P .05) different.

THE EFFECTS OF TWO CARBAMATE INSECTICIDES  
ON BOBWHITE (COLINUS VIRGINIANUS) BIOENERGETICS

by

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AN ABSTRACT OF A MASTER'S THESIS

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The purpose of the energetic studies was to determine if two carbamate insecticides, Sevin® and Furadan®, would alter bobwhite (Colinus virginianus) gross energy intake, metabolized energy, excretory energy, efficiency of metabolism, or fecal energy. Brain cholinesterase levels were also measured to determine if Sevin or Furadan exhibited cumulative anticholinesterase activity change, and if the overall treatment levels could be correlated with cholinesterase activity.

An LD<sub>50</sub> study was conducted with 36 quail to aid determination of the sublethal dosage levels for the Sevin-energetics study. The insecticide carrier was corn oil, and the observation period was 4 days. The LD<sub>50</sub> was 1193 mg/kg body weight. Parasympathetic symptoms of diarrhea, lachrimation, salivation, and tremors were recorded.

For the Sevin-energetics study, 52 male bobwhites were divided into four treatment groups (corn oil control, 10 mg/kg, 50 mg/kg, and 90 mg/kg Sevin), and dosed orally every 2 days, for 8 days. For the Furadan-energetics study, 56 male quail were divided into five treatment groups (water control, corn oil control, 0.25 mg/kg, 1.13 mg/kg, and 2.0 mg/kg Furadan), and were dosed orally every 2 days for 8 days. The Sevin and Furadan treatment groups experienced the greatest energetic variable changes when first dosed. With the corn oil control also showing initial energetic changes, a method was devised which removed the corn oil effects and eliminated pretreatment differences among treatment groups. Therefore the insecticide effects were examined in relation to the corn oil control.

During the studies, some quail gained or lost weight. Since the weight-gain quail could not be used in data analysis, a two-way analysis of variance was used with unequal subclasses. The energetic variables for weight-loss quail could be corrected.

The Sevin-energetics data showed no significant ( $P>.05$ ) treatment effect or treatment-period interaction. The highly significant ( $P<.01$ ) period effect indicated that the injection of corn oil was the major cause of energetic variable change. A water control was added to the Furadan-energetics study to determine the actual energetic effects of corn oil.

The Furadan-energetics data showed significant decrease in bobwhite (1.13 and 2.0 mg/kg doses) gross energy intake (5.355 and 0.331 kcal, respectively), metabolized energy (4.380 and 7.281 kcal, respectively), and weight (2.2 and 3.3 grams, respectively), and a significant increase in fecal energy (0.071 and 0.110 kcal/g, respectively). No cumulative effects were noted over the 8 days of treatment. All Furadan treatment groups returned to pretreatment energetic values 6 days after the initial dosing. The 0.5 ml corn oil doses significantly increased bobwhite gross energy intake (7.951 kcal), metabolized energy (5.951 kcal), and excretory energy (2.042 kcal).

Four quail were sacrificed from each insecticide treatment group 48 hours after each dose. The brain cholinesterase data showed no significant treatment effects. Only those quail dying of Sevin poisoning during the LD<sub>50</sub> study showed cholinesterase levels below that of the controls.

As the Environmental Protection Agency bans the use of certain organo-chlorines, carbamates become more popular as substitutes. The agricultural use of the nonpersistent carbamates, Sevin and Furadan, is not harmful from a bobwhite energetic view point.