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

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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 LD_{50} of a given chemical for one of the six species would predict the LD_{50} of the same chemical for another species. The results indicate that such extrapolations are not reliable. For example, the LD_{50} 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 that 50% of the time the LD_{50} of one species of bird differed from the LD_{50} 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 LD_{50} 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°F ± ½° 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.

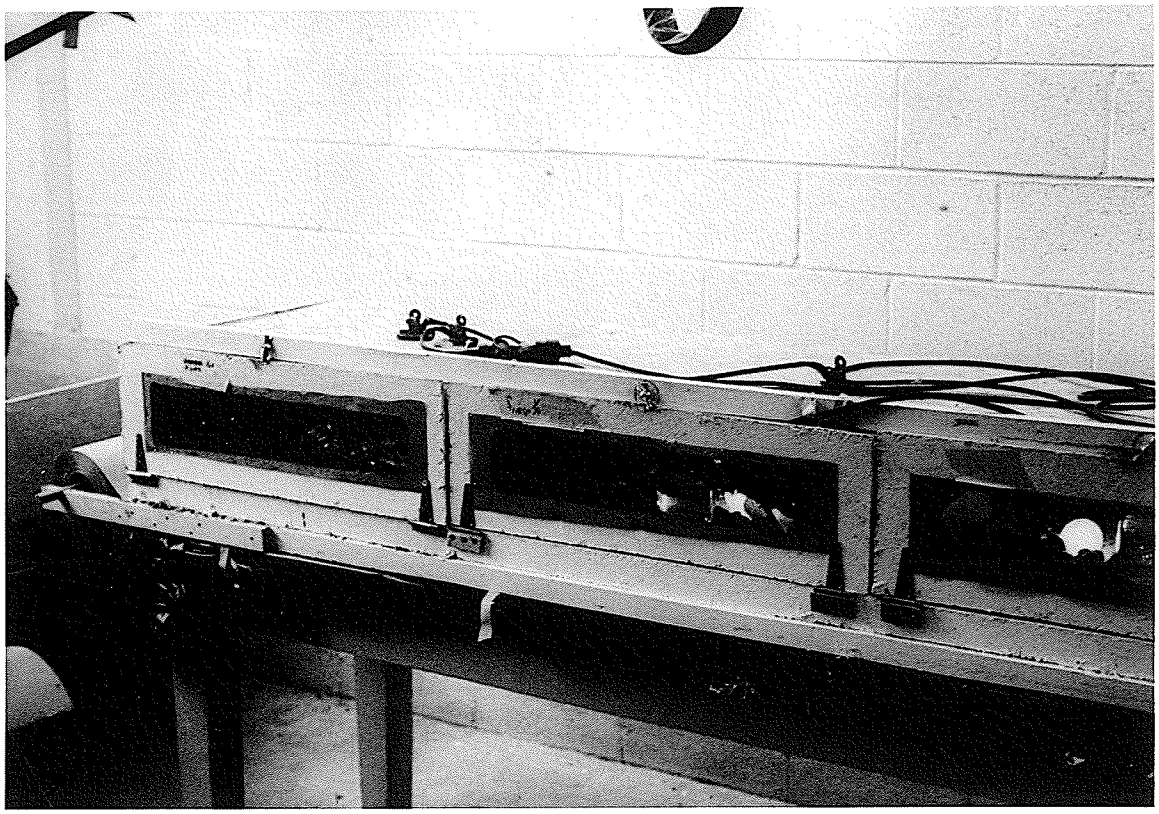

Fig. 2:1--Brooder

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 LD_{50} 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 LD_{50}. The percent mortality in each group was then plotted against the log of dose (Fig. 2:2) and the LD_{50} was calculated to be 69.1 mg/kg.

Similarily, the acute oral LD_{50} 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 LD_{50} 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 LD_{50} 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 LD_{50} dose per day) and an intermediate dose (0.10 of the LD_{50} dose per day).

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[1] Active ingredient

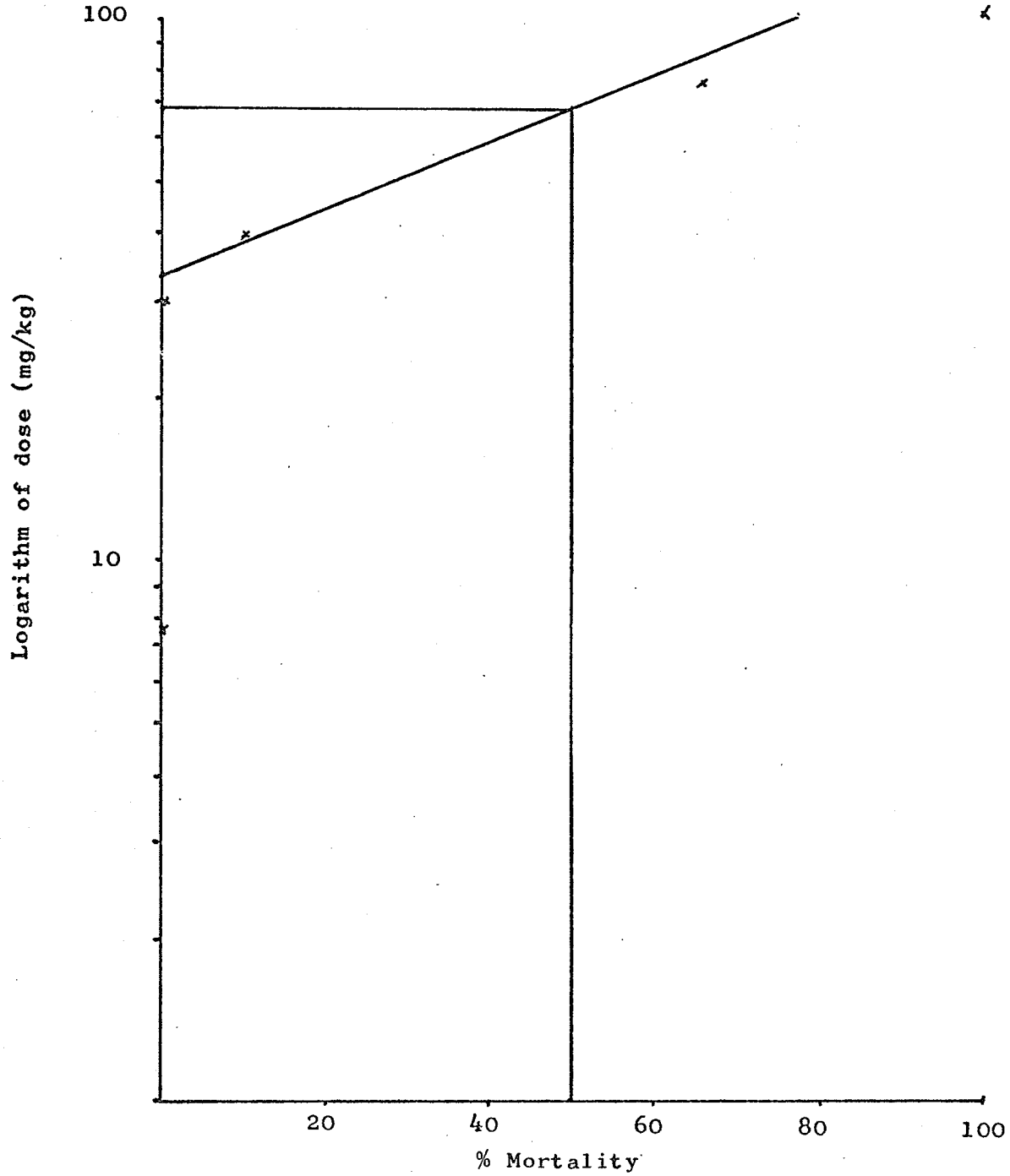

Fig. 2:2.--Acute oral LD_{50} for Japanese quail fed Abate