(TDR03B) DATA EVALUATION RECORD PAGE 1 OF

CASE GS0109 TERBUFOS PM 04/15/82

CHEM 105001 Terbufos ( S-(((1,1-dimethylethyl)thio)

BRANCH EEB DISC 40 TOPIC 10200042

FORMULATION 00 = ACTIVE INGREDIENT

FICHE/MASTER ID 00085177 CONTENT CAT 01

Fink, R.; Reno, F.E. (1973) Final Report: One-generation Reproduction Study--Bobwhite Quail: Project No. 362-145. (Unpublished study received May 1, 1974 under 4F1496; prepared by Environmental Sciences Corp., submitted by American Cyanamid Co., Princeton, N.J.; CDL:090808-S)

SUBST. CLASS = S.

DIRECT RVW TIME = 8 hrs. (MH) START-DATE 10/4/82 END DATE 12/02/82

REVIEWED BY: James D. Felkel
TITLE: Wildlife Biologist
ORG: Ecological Effects Branch, Hazard Evaluation Division (TS-769)
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SIGNATURE: James D. Felkel DATE: 12/10/82

APPROVED BY:
TITLE:
ORG:
LOC/TEL:

SIGNATURE: DATE:

10 pages

DATA EVALUATION RECORD

1. Chemical: Terbufos (Shaughnessy #105001)

2. Formulation: Technical AC92100 - 89.0%

3. Citation: Fink, R.; Reno, F.E. (1973) Final Report: One-generation Reproduction Study--Bobwhite Quail: Project No. 362-145, (Unpublished study received May 1, 1974 under 4F1496; prepared by Environmental Sciences Corp., submitted by American Cyanamid Co., Princeton, N.J.; CDL:090808-S) MRID#00085177

4. Reviewed by: James D. Felkel, Wildlife Biologist
Ecological Effects Branch
Hazard Evaluation Division (TS-769)

5. Date Reviewed: October 21, 1982

6. Test Type: Avian Reproduction

A. Test Species: Bobwhite quail (Colinus virginianus)

7. Reported Results: No adverse effect on the reproductive success of the quail occurred in this study, with dietary levels of 2 and 20 ppm.

8. Reviewer's Conclusions: This study is scientifically sound but pen-by-pen data are required for statistical evaluation of results. The study may fully meet the intent of proposed guidelines (7/10/78) following submission and evaluation of pen-by-pen data.

METHODS

## EXPERIMENTAL MATERIAL

Identification - Technical AC92100 - 89.0%, Inert Ingredients - 11.0%

Description - A clear liquid

Receipt Date - March 29, 1973

HLI Identification No. - LH 14,856A

## WILDFOWL

Species - Bobwhite Quail (Colinus virginianus)

Supplier - Dennis Quail Farm, Willards, Maryland

Blood Testing - U.S. Pullorum-Typhoid Clean

Age At Initiation - Nine months

## EXPERIMENTAL

Pen-reared bobwhite quail, phenotypically indistinguishable from wild birds, were quarantined for 14 days upon arrival at the Truslow Farms research facility. Each bird was examined for physical injury that may have been encountered in transit. Six males and six females were sent to the Maryland Animal Health Department Laboratory for analysis. When the birds were diagnosed negative for abnormal lesions at necropsy, bacteriology, and serology, one hundred eight quail (36 males and 72 females) were randomly distributed into the following groups:

| Group No. | Dosage Level | No. of Pens | Birds/Pen | |
|---|---|---|---|---|
| | | | Male | Female |
| 1 - Controls | 0 | 12 | 1 | 2 |
| 2 - AC92100 | 2 ppm | 12 | 1 | 2 |
| 3 - AC92100 | 20 ppm | 12 | 1 | 2 |

Diet Preparation

Technical AC92100 was added to edible grade corn oil and premixed with aliquots of basal diet utilizing a mortar and pestle. The concentrates were frozen until utilized for weekly preparation of fresh diets. The final diet consisted of 98 parts by weight of the basal diet and two parts by weight of corn oil. The basal diet was a commercial game bird breeder ration containing 20.9% protein, 5.3% fat, and 4.8% fiber (analysis by Department of Poultry Science, College of Agriculture, University of Maryland).

Testing Phase

All birds in each group received the appropriate diets ad libitum for the 18-week duration of the study.

The birds were housed indoors in Georgia Quail Farm Breeding Pens (Model 206).

The photoperiod for the first eight weeks (April 4, 1973 through May 30, 1973) was seven hours of light per day. The photoperiod was then increased to 17 hours of light per day, and increased by 15 minutes per week for the next 10 weeks. The birds received five footcandles of illumination during the lighting phase of the study.

For the first eight weeks, the temperature in the research facility was maintained at 10°C and 55 percent relative humidity. When the photoperiod was increased to 17 hours of light per day, the temperature was raised to 22°C and maintained at that temperature for the duration of the study. The environmental control system in the quail facility utilized 100 percent makeup air. Outside air entering the facility passed through a particulate filter, an electrostatic filter, a heating-humidifying system, and an ultra-violet filter. The particulate filters were changed twice a week.

Body weights were recorded at initiation of the study, prior to the onset of egg laying, and at termination. Body weights were not recorded during egg laying because of the adverse effect handling may have on egg production. Food consumption was recorded bi-weekly throughout the study.

Eggs were collected daily and stored at 16°C and 55 percent relative humidity. Each egg collected was marked according to the pen from which it was taken. At weekly intervals the eggs were placed in a Chick Master (Model 52E) for incubation. All eggs were candled on Day 0 for eggshell cracks; on Day 11 to measure embryonation; and on Day 19 to measure embryo survival. On Day 19 of incubation, the eggs were placed in a Robins' Incubator (Model 17H) and allowed to hatch. The temperature during incubation was maintained between 37.2°C and 37.4°C.

All eggs were removed on Day 21 of incubation; and the hatchlings were housed according to the appropriate parental grouping and maintained on control diet for 14 days.

At weekly intervals, approximately five percent of the eggs with sound shells from each group were randomly selected to measure eggshell thickness. The eggs were opened at the waist; the contents thoroughly washed out; and the shells dried for one week at 24°C. The thickness of the dried shell plus membrane at the waist was measured using a micrometer calibrated in 0.01 mm units.

Statistical Methods

The individual pen data within each experimental group was utilized for analysis. Body weight, food consumption, eggs laid, eggs cracked, eggs embryonated, live three-week embryos, normal hatchlings, 14-day old survivors, and eggshell thickness data were analyzed by a single classification analysis of variance or F-test. Before completing each F-test, the variances were tested for heterogeneity by the method of Bartlett (1). If homogeneous variances were found, the F-test was completed. When a significant F valve was obtained, significant group differences were determined by Scheffe's method of multiple group comparisons (2). In those instances of heterogeneous variances, a log transform was performed on the data and the test homogeneity was performed again. If the transformed data had homogeneous variances, the analysis described above was performed. If heterogeneous variances still existed, comparisons of individual groups were made by a t-test for unequal variances. Null hypothesis rejections were made at $p \leq 0.05$.

RESULTS

Bobwhite quail were fed dietary levels of technical AC92100 at concentrations of two and 20 ppm for a period of 18 weeks. Data are presented as follows:



Statistical analyses of body weight, food consumption, eggs laid, eggs cracked, eggs embryonated, live three-week embryos, normal hatchlings, 14-day old survivors, and eggshell thickness revealed no differences between the control and test birds.

There was a slight reduction in the number of eggs laid per hen, eggs embryonated of eggs set, 14-day old survivors of normal hatchlings, and 14-day old survivors per hen (Table 1b) in both the two and 20 ppm treatment groups. These differences are well within the normal variation experienced in bobwhite quail reproduction studies, are not statistically significant, and are not considered biologically meaningful.

There were no observable symptoms of toxicity or behavioral abnormalities observed during the study. All birds appeared normal throughout the study; and there was no mortality.

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(1) Ostle, Bernard, Statistics in Research, Iowa State College Press, Ames, Iowa, 1956.

(2) Ibid.

Reviewer's Evaluation

Methods reported were generally consistent with proposed guidelines (7/10/78). There were a few minor differences from recommended temperatures and/or humidity levels. Body weights were recorded at initiation, before egg laying and at termination rather than at weeks 0, 2, 4, 6, 8, and termination. Eggshell thickness measurements were not done on all newly laid eggs every two weeks, but rather weekly on 5% of the eggs with sound shells. None of the differences found would prevent the study from meeting the intent of the guidelines. However, results cannot be critically reviewed until the pen-by-pen data is submitted to permit statistical evaluation.

An initial screen was performed on the summary data submitted, using the "SUPER" (chi-square) program available at EEB. Impairment in treatment relative to controls ($p < 0.05$) was detected overall (eggs to 14-day old survivors) and in viable embryos of eggs set at the 2 ppm test level. At 20 ppm, impairment ($p < 0.05$) was detected overall, in eggs set of eggs laid, and in viable embryos of eggs set. However, these results must be confirmed or rejected by ARSIN analysis before any conclusions can be drawn. Pen-by-pen data is required for this.

CONCLUSIONS:

1. Category: Supplemental.

2. Rationale: Methods were generally consistent with proposed guidelines (7/10/78). However, pen-by-pen data are required for statistical evaluation of results.

3. Repairability: Yes. Study may be upgraded following submission and evaluation of pen-by-pen data.

TABLE 1a

Reproductive Data - Bobwhite Quail

| | Controls | Technical AC 92100 | |
|---|---|---|---|
| | | 2 ppm | 20 ppm |
| Eggs Laid | 815 | 719 | 715 |
| Eggs Cracked | 25 | 34 | 39 |
| Eggs Set* | 676 | 563 | 626 |
| Eggs Embryonated | 614 | 537 | 543 |
| Live Three-Week Embryos | 603 | 525 | 533 |
| Normal Hatchlings | 538 | 493 | 487 |
| Fourteen-Day-Old Survivors | 468 | 424 | 414 |

* Excluces those cracked and those removed for eggshell thickness analysis. The above differences were not significant at the 95 per cent level of confidence.

TABLE 1b

Reproductive Success - Bobwhite Quail

| | Controls | Technical AC 92100 | |
|---|---|---|---|
| | | 2 ppm | 20 ppm |
| Eggs Laid Per Hen In Eight Weeks | 34.0 | 30.0 | 29.7 |
| Eggs Cracked Of Eggs Laid (%) | 3.1 | 4.7 | 5.5 |
| Eggs Embryonated Of Eggs Set (%) | 90.8 | 95.1 | 86.7 |
| Live Three-Week Embryos Of Embryonated Eggs (%) | 98.2 | 97.8 | 98.2 |
| Normal Hatchlings Of Live Three-Week Embryos (%) | 89.2 | 93.9 | 91.5 |
| Fourteen-Day Survivors Of Normal Hatchlings (%) | 87.0 | 86.0 | 85.1 |
| Fourteen-Day Survivors Per Hen | 19.5 | 17.7 | 17.3 |

* Differences were not significant at the 95 per cent level of confidence.

TABLE 2

Eggshell Thickness - Bobwhite Quail

| | Controls | Technical AC 92100 | |
|---|---|---|---|
| | | 2 ppm | 20 ppm |
| No. Of Eggs Analyzed | 48 | 48 | 48 |
| Mean Shell Thickness (mm) | 0.206 | 0.200 | 0.203 |

The above differences were not significant at the 95 per cent level of confidence.

TABLE 3

Body Weight and Food Consumption - Bobwhite Quail

| | Controls | | Technical AC 92100 2 ppm | | Technical AC 92100 20 ppm | |
|---|---|---|---|---|---|---|
| WEEK | B.W. | F.C. | B.W. | F.C. | B.W. | F.C. |
| | g | g | g | g | g | g |
| 0 | 188 | - | 193 | - | 180 | - |
| 2 | | 18.9 | | 17.6 | | 19.4 |
| 4 | | 15.8 | | 17.0 | | 17.0 |
| 6 | | 17.3 | | 16.9 | | 17.6 |
| 8 | 208 | 18.4 | 213 | 18.1 | 208 | 17.6 |
| 10 | | 18.2 | | 20.3 | | 18.6 |
| 12 | | 25.6 | | 24.6 | | 24.6 |
| 14 | | 25.6 | | 24.6 | | 24.6 |
| 16 | | 25.1 | | 26.1 | | 25.1 |
| 18 | 214 | 25.2 | 211 | 26.8 | 213 | 26.4 |

The body weight data are presented as a group mean.
The food consumption data are presented as the group mean feed consumed per bird per day.

The above differences were not significant at the 95 per cent level of confidence.

B.W. = Body Weight
F.C. = Food Consumption

ENTER IN ORDER:
EGGS LAID,EGGS CRACKED,EGGS SET,VIABLE EMBRYOS
3-WEEK LIVE EMBRYOS,NORMAL HATCHLINGS,AND
14-DAY SURVIVORS FOR CONTROLS

815 25 676 614 603 538 468

ENTER IN ORDER:
EGGS LAID,EGGS CRACKED,EGGS SET,VIABLE EMBRYOS
14-DAY SURVIVORS FOR A TREATMENT GROUP

20 ppm

713 39 626 543 535 487 414

TOTAL CHISQUARE= 4.4470690

CONTROLS

| LOST | ALIVE | TOTAL |
|---|---|---|
| 25.00 | 790.00 | 815.00 |
| 62.00 | 614.00 | 676.00 |
| 11.00 | 603.00 | 614.00 |
| 65.00 | 538.00 | 603.00 |
| 70.00 | 468.00 | 538.00 |

TREATMENT GROUP

| LOST | ALIVE | TOTAL | CH. |
|---|---|---|---|
| 39.00 | 674.00 | 713.00 | 6.08 |
| 83.00 | 543.00 | 626.00 | 5.90 |
| 8.00 | 535.00 | 543.00 | 0.04 |
| 48.00 | 487.00 | 535.00 | 0.84 |
| 73.00 | 414.00 | 487.00 | 1.01 |

CHI 1DF=3.84 (P<0.05)

Bobwhite quail

ENTER IN ORDER:
EGGS LAID,EGGS CRACKED,EGGS SET,VIABLE EMBRYOS
3-WEEK LIVE EMBRYOS,NORMAL HATCHLINGS,AND
14-DAY SURVIVORS FOR CONTROLS
815 25 676 614 603 538 468

ENTER IN ORDER:
EGGS LAID,EGGS CRACKED,EGGS SET,VIABLE EMBRYOS
14-DAY SURVIVORS FOR A TREATMENT GROUP

2 ppm

719 34 653 537 525 493 424

TOTAL CHISQUARE= 6.1421130

CONTROLS

| LOST | ALIVE | TOTAL |
|---|---|---|
| 25.00 | 790.00 | 815.00 |
| 62.00 | 614.00 | 676.00 |
| 11.00 | 603.00 | 614.00 |
| 65.00 | 538.00 | 603.00 |
| 70.00 | 468.00 | 538.00 |

TREATMENT GROUP

| LOST | ALIVE | TOTAL | CHI |
|---|---|---|---|
| 34.00 | 685.00 | 719.00 | 3.32 |
| 116.00 | 537.00 | 653.00 | 21.87 |
| 12.00 | 525.00 | 537.00 | 0.56 |
| 32.00 | 493.00 | 525.00 | 7.24 |
| 69.00 | 424.00 | 493.00 | 0.31 |

CHI 1DF=3.84 (P<0.05)