Data Evaluation Record

1. Chemical: Dylox Technical (Trichlorfon)
   Shaughnessy No.:057901

2. Test Material: Trichlorfon [Dimethyl(2,2,2-trichloro-1-hydroethyl)phosphonate], 99.8%, batch #203-0021, Lot #1542102252 (CAS #52-68-6); a white crystalline solid

3. Study type: Avian Reproduction (71-4)
   Test Species: Northern bobwhite
   Colinus virginianus

4. Study ID: Pedersen, C.A. and S.M. Thompson. 1994. Effects of DYLOX Technical on Bobwhite Quail Reproduction. Performed by Bio-Life Associates, Ltd., Neillsville, WI for Miles Incorporated, Agriculture Division, P.O. Box 4913, Kansas City, MO 64120. Report #106409. **MRID #431195-01.**

5. Reviewed by: Kathryn V. Montague, M.S. Signature:
   Biologist Date: 4/6/94
   EEB/EFED

6. Approved by: Norm Cook Signature: Norman J. Cook
   Head, Section II Date: 04.08.94
   EEB/EFED

7. Conclusions: The study is scientifically sound and is classified as core. The NOEC for hatchling survival was 9 ppm (LOEC = 30 ppm). The NOEC for all other parameters was 30 ppm (LOEC = 85 ppm), except for eggshell thickness, which was not affected at any level tested (NOEC = 85 ppm).

8. Recommendations: N/A

9. Background information: This study was submitted under Section 6(a)(2) as new information, and was reviewed as a submission toward fulfillment of Guideline 71-4 in support of reregistration for trichlorfon.

10. Discussion of Individual Tests: N/A

11. Materials and Methods:
    a. Test animals: Pen-reared bobwhites were obtained from Oak Ridge Game Farm., Gravette, AK. The birds were all from the same hatch and in their first breeding season. They were acclimated to laboratory conditions for 24 days prior to the initiation of the study. They were 18 weeks old at the start of the test.

    b. Test system: The birds were housed in 51 x 25 x 23 cm galvanized steel mesh pens containing one male and 1 female bobwhite each. The birds were given fresh feed weekly and ad

libitum water. The average daily temperature in the study room was 68°F, and the average relative humidity was 65%.

c. Study design: The birds were maintained for 9 weeks on treated feed on a 7-hr light/17-hr dark photoperiod. At the end of the ninth week, the light was increased to 17 hours per day, and this photoperiod was maintained for the rest of the study. The birds were presented with treated feed (Dylox-treated Purina custom gamebird Layena) until the end of week 22. Nominal dosing levels were: control (corn oil and acetone), 10, 30, and 90 ppm a.i. Dylox. Test diets were prepared fresh weekly and stored in a freezer. Stability testing was performed for 1 week under environmental conditions of the animal room during a previous pilot study. Three samples were taken from the top, middle and bottom of the 10 and 90 ppm diets immediately after preparation on week 1 for homogeneity testing. Samples from the control and 30 ppm mixes during week 1 and from all levels during weeks 5, 10, 15 and 20 were taken to verify concentration of Dylox.

Each treatment group consisted of 16 pairs of bobwhite. The adults were observed once a day throughout the study. Body weight was measured biweekly for the first 12 weeks and at test termination. Feed consumption was measured biweekly for weeks 2 - 21. Egg laying began in week 11. Eggs were collected daily; the size of each egg was recorded, the eggs were candled and any cracked, broken, soft-shelled or membranous eggs were noted and discarded. The eggs were held in an egg storage room at 64-67°F, 80% RH and turned once a day for 7 days. At the end of each 7-day collection period (or "hatch"), eggs were moved from the holding room to an incubator, where they were automatically turned every 2 hours. Eggs were candled and misted on day 10 to establish fertility and day 17 to determine embryo survival. On day 21 of incubation, eggs were placed in hatching trays and misted prior to hatch. On incubation days 24-25, hatchlings, unhatched eggs and shells were recovered. Hatchling body weight was recorded and the hatchlings were placed in brooders for 14 days, during which they were fed untreated feed (Purina custom gamebird Startena) and provided water ad libitum. On day 14, hatchling body weight was recorded and the birds were sacrificed.

Eggs collected on the first day of 5 collection intervals were tested for eggshell thickness at the equatorial circumference, 3 times per egg. They were broken, washed and allowed to air-dry for 48-hours at room temperature. An Ames pocket thickness measure was used to measure shells to the nearest 0.01 mm.

d. Statistics: The following parameters were analyzed: Adult body weight; adult feed consumption;, eggs laid, per hen; eggs cracked or broken, as a percentage of total laid and per hen;

eggs set, per hen; viable day 10 embryos, as a percentage of eggs set per hen; viable day 17 embryos, as percentage of number viable day 10 embryos per hen; normal hatchlings, per hen and as a percentage of 3-week embryos; hatchling body weight; number of 14-day survivors, as percent of hatchlings per hen; survivor body weight; and eggshell thickness. All statistical analyses were performed by Sebaugh's Information Services as a consultant to Bio-Life Associates. The SAS system was used to perform the statistical analyses.

12. Reported Results: The mean measured concentrations of Dylox in test feed, determined from sampling, were 0.0 (control), 9, 30 and 85 ppm a.i. The diets were determined to be homogeneous and stable for 7 days under conditions of the animal room.

There were 10 adult mortalities during the test period, 1 in the control group, 1 at the 9 ppm level, and 8 at the 85 ppm level. Post-mortem examinations revealed that 6 of the 8 birds that died in the 85 ppm group were emaciated, with no feed in the crop or gizzard. One mortality at this level was emaciated with no feed in the gizzard or upper intestine. The rest of the mortality birds did not have any consistent post-mortem findings, and their pen mates (sacrificed at the time the mortalities were found) were normal. Fifty percent of the surviving birds at each test level were also necropsied. One of these birds at the 85 ppm level had no feed in the crop or gizzard. Friable livers were discovered in 1 bird at 9 ppm, 1 bird at 30 ppm and 4 at 85 ppm, but this effect was not considered treatment related.

No significant differences were noted in mean adult body weight. Feed consumption was decreased at the 85 ppm level for weeks 14, 16, 18, 20 and 22, but not overall.

Clinical signs of toxicity were noted in 6 females at 85 ppm. These signs included: lethargy, stumbling, walking slowly, falling on side, weakness, inactivity and death.

There was a significant decrease in the number of eggs laid per hen and number of viable embryos at 85 ppm. A significant decrease in the number of hatchlings as a percentage of viable embryos and as a percentage of day 17 live embryos was noted at 30 ppm, but was not seen at 85 ppm; therefore, this effect was not considered treatment-related. There was a significant decrease in day 14 survivor body weight at 30 ppm, but this was not seen at 85 ppm and was therefore not considered treatment-related. There was also a significant decrease in the number of day 14 survivors as a percentage of number hatched at 9 ppm, but this was not considered treatment-related as it was not seen at the higher levels tested. There were no significant effects on eggshell thickness, and no gross abnormalities were seen during post-mortem examinations of the hatchlings.

13. Study Author's Conclusions/Quality Assurance Report: Based on the above results, the NOEC was 30 ppm, and the LOEC was 85 ppm, except for eggshell thickness, which was not affected at any level tested.

Quality Assurance and Good Laboratory Practice statements were included in the report.

14. Reviewer's Discussion and Interpretation of the Results:
a. Test Procedure: The test design and procedure were scientifically sound and in accordance with Guidelines recommendations.

b. Statistical Analysis: All statistics were verified using the SAS system and EPA's TOXSTAT computer program. For certain parameters, the assumptions for ANOVA were not met, so William's test (a non-parametric method) was used to assess effects in these cases. When this test was used, a significant decrease in the number of hatchlings as a percentage of eggs set was seen at 85 ppm, as well as a significant decrease in the number of 14-day survivors as a percentage of both number hatched (9 and 30 ppm) and eggs set (30 and 85 ppm). A significant decrease in the number of hatchlings as a percentage of eggs laid was seen at 85 ppm. Adult body weight change was also analyzed; a significant difference in overall male body weight was seen at 85 ppm (using ANOVA with Duncans and Bonferroni tests), as well as significant differences in male body weight change for weeks 0-2 and 2-4 at 85 ppm (using William's test since data did not fit assumptions of ANOVA). A significant difference in female body weight change was noted for weeks 0-2 (William's test) and 6-8 (Duncan's and Bonferroni) at 85 ppm. These results, with the exception of 14-day survival and body weight change, were in agreement with the reported results.

c. Discussion/Results: Based on the reviewer's statistical verification, the LOEC for 14-day hatchling survival is 30 ppm (NOEC = 9 ppm). The LOEC for all other parameters is 85 ppm (NOEC = 30 ppm), with the exception of eggshell thickness, which was not affected at any level tested (NOEC = 85 ppm). The study is scientifically sound and is classified as core.

d. Adequacy of the study:
(1) Classification: Core.
(2) Rationale: N/A
(3) Repairability: N/A