

US EPA ARCHIVE DOCUMENT

5-4-87
X

TDMS DATA EVALUATION RECORD

PAGE 1 OF

CASE GS _____

PM ____/____/____

CHEM Diazinon

BRANCH EEB DISC _____

FORMULATION 23% Microencapsulated (Knox Out 2FM)

FICHE/MASTER ID ROODI004

CITATION: Beavers, J.B. (1978a) Acute Oral LD50 - Bobwhite Quail, Knox Out 2FM, Final Report, Submitted by Pennwalt Corp.;
Beavers, J.B. (1978b) Eight-Day Dietary LC50 - Bobwhite Quail Knox-Out 2FM, Submitted by Pennwalt Corp.;
Beavers, J.B. (1978c) Eight-Day Dietary LC50 - Mallard Duck, Knox Out 2FM, Submitted by Pennwalt Corp.;
Pennwalt Corp. (1979) Knox Out 2FM Insecticide -
→ Simulated Avian Field Study, Submitted by Pennwalt Corp.

SUBST. CLASS=

OTHER SUBJECT DESCRIPTORS
PRIM:

DIRECT REVIEW TIME= (MH) START DATE - July 1980 END DATE - August 1980

REVIEWED BY: John Leitzke
TITLE: Ecologist
ORG: EEB
LOC./TEL:

SIGNATURE:

APPROVED BY: Harry Craven
TITLE: Supervisory Biologist
ORG: EEB
LOC./TEL: 557-7600

SIGNATURE: *H.T. Craven*

5/4/87

Bobwhite LD50 supplemental due to use of corn oil; Mallard LC50 core for formulation oral testing; Bobwhite LC50 core for formulation oral testing; pen study unacceptable due to use of inappropriate test species for turf hazard and use of lower application rates than allowed by proposed label.

DATA EVALUATION RECORD

1. Chemical: Diazinon
2. Formulation: Knox Out 2FM (23% Microencapsulated)
Diazinon Emulsifiable
3. Citation: Pennwalt Corp. (1979) Knox Out 2FM insecticide
- Simulated Avian Field Study, Project No.
W1-2-79; Prepared and Submitted by Pennwalt
Corp. (Accession No. 240993).
4. Reviewed by: John S. Leitzke
Ecologist, Section 3
EEB/HED
5. Date Reviewed: August 19, 1980
6. Test Type: Avian Small Pen Field Test
Test Species: Bobwhite Quail (Colinus virginianus)
7. Reported Results:

Knox Out at the highest possible label rate will not be hazardous to avian wildlife when compared with the results from this test at 10.9 lb ai/A.

8. Reviewer's Conclusions:

Diazinon at 32.7 lb ai/A, whether as Knox Out or an emulsifiable formulation, was more adverse on quail in terms of mortality and weight loss than either form at 10.9 lb ai/A. At the higher rate, Knox Out was more hazardous than diazinon emulsifiable in terms of mortality. Knox Out was also more hazardous than the emulsifiable at the lower rate in terms of severity of weight loss. Greater effects were due to greater amounts of diazinon on grass in the Knox Out plots than in the emulsifiable plots. In general, the study was sound and well run. However, bobwhite quail is an inappropriate test species for turf areas (the more sensitive mallard, as a turffeeder, would have been more appropriate), and the application rates on the proposed label are lower than those used in the test. For these reasons this study is unacceptable in meeting the Guidelines requirement for an avian small pen field test on the formulation, Knox Out 2FM.

9. Materials and Methods:

Both Knox Out 2FM (23% microencapsulated) and a diazinon emulsifiable formulation were applied at 10.9 and 32.7 lb ai/A on separate plots with a fifth plot as a control. (For easier discussion, the pens receiving diazinon and Knox Out at 10.9 lb ai/A and at 3-times that rate will be referred to as pens D-1x, KO-1x, D-3x, and KO-3x, respectively.)

Mature bobwhite quail, initially weighing 240 to 320 gm, were housed by pairs in 4 x 5 ft cages. Six pairs (12) of caged, individually tagged birds were acclimated to each of the 50 x 80 ft test plots about 2 1/2 weeks prior to the 2 week test period. All birds that died during the acclimation and most that died during the test period were replaced as soon as mortality was noted. All birds (including replacements) were weighed 3 times, at the beginning of the acclimation and test periods, and at the end of the test (or at death during the test period). The only exceptions were the original 12 birds in pen KO-3x, those exposed to Knox Out at 32.7 lb ai/A, since they all died after a few days. However, their replacements did have the same 3 weighings as the others.

The perennial rye grass in the pens was cut to 1 1/2 to 2 inches height. One-half the cages in each pen = test plot were provided feed and water immediately after insecticide application, and one-half were withheld feed and water for 12 hours. Thereafter, all cages were supplied daily. Each cage was moved every 2 days to a fresh area of the test plot. Amprolium was added to the water since several birds had died earlier due to acute intestinal coccidiosis.

Soil samples (0 to 3 inches) and grass samples were taken from each plot on test days 1, 4, 8, and 14.

- a. Statistical Analysis: All statistical analyses were redone on the EEB computer terminal using standard SAS and BMDP programs. Mortality data were analyzed on an expanded 2 x 5 Contingency Table program for all test plots and on the 2 x 2 Contingency Table program for pens KO-3x, the two test pens receiving 32.7 lb ai/A, one by Knox Out 2FM and one by diazinon emulsifiable (attached). Body weights, percent differences in body weight between different weighing times and their arcsin transformations were analyzed by 2-Way and 1-Way ANOVA programs with Duncan's Multiple Rate Tests (attached).

- b. Discussion/Results: All dead birds were necropsied to determine cause of death. While the cause of only a few deaths could not be determined, most deaths were attributed to pesticide poisoning, some possibly to disease, and a few as definitely to disease or harassment. Of the latter group, one occurred in the control pen, none in pen D-1x, one in pen KO-1x, one in pen D-3x, and none in KO-3x. Most of the deaths occurred in pen KO-3x and a number in pen D-3x as well. All 12 of the original birds in pen KO-3x died within the first few days and were replaced. Several of these replacements died as well.

Virtually all birds showed some reduction in weight between the beginning of acclimation and the actual test period. Additional reduction occurred during the test period, the severity depending on the formulation and rate of application the birds were exposed to in that particular pen.

Greater residues of diazinon were on the grass (and at first in the soil) in Knox Out pens than in the diazinon emulsifiable pens. This is discussed in more details below.

10. Reviewer's Evaluation

- a. Test Procedures: In general the experimental design and protocol and the day-to-day operation of this small pen field test are quite sound. However, two major problems prevent it from being adequate to use in a hazard assessment for the proposed registration on turf at 5.5 lb ai/A. The first problem is the use of bobwhite quail as the test species. Not only are bobwhite quail an inappropriate test species for turf areas and a waterfowl such as mallard much more appropriate, the mallard is also considerably more sensitive than the bobwhite quail to Knox Out in dietary tests; the LC₅₀'s are 149 and 345 ppm ai, respectively. The second problem is that the proposed label rate for Knox Out on turf is 5.5 ai/A, while the field test was conducted at 10.9 and 32.7 lb ai/A.
- b. Statistical Analysis: Mortality data from all pens were first analyzed on an expanded 2 x 5 Contingency Table in several ways. First, mortality in all birds (including replacements) was tested against all deaths, only those deaths due to pesticide poisoning (determined from the necropsies), and all nondisease and harassment deaths. Then mortality only in the original birds

(excluding replacements) was tested the same three ways. In all cases there was a statistically significant nonrandom distribution of deaths among the five pens (attached).

Mortality data were then analyzed on a 2 x 2 Contingency Table using pens KO-3x and D-3x to test for comparative hazard of Knox Out to diazinon emulsifiable in the same six ways as above. Again, there was a statistically significant nonrandom distribution of deaths (attached).

Changes in body weight were analyzed in several ways by 2-Way and 1-Way ANOVA's with Duncan's Multiple Range Tests. Data on weights came from all birds that survived the entire length of the test, from replacements of those that died and from those that died but were not replaced. This differed from the experimenters' statistical analyses in that they excluded this latter group. Another difference is that they excluded pen KO-3x since all original birds died within a few days. However, because some of the replacement birds in pen KO-3x died as well and the survivors exhibited toxic symptoms, these birds were included in the present analyses. Body weights of dead birds that weren't replaced were included because not all dead birds had severely reduced weights and some surviving birds did.

Note: Only the results of the Duncan's tests are given (attached). Raw body weight data were analyzed with a 2-Way ANOVA for all pens against all three sampling dates (beginning of acclimation, beginning of actual test and end of test or death). This test showed significant differences across pens as well as across sample times.

Next, a 2-Way ANOVA was run using the percent change in body weights from the first weighing (beginning of acclimation) to the second (beginning of actual test) and the first to the third (end of test or death) for all pens against the percent differences of sample times 1 to 2 and 1 to 3. This was done to duplicate analyses of the experimenters, who ran separate 1-Way ANOVA's. However, since percents (and percent differences) are not normally distributed and can't be used in an ANOVA calculation, the data underwent arcsin transformation, and another 2-Way ANOVA was run on the transformed percent differences. Both analyses showed significant differences across pens as well as the two sets of percent differences.

To test for changes occurring during the actual test period, two 1-Way ANOVA's were run for all pens against the percent difference of sample times 2 to 3, one using the percent differences and the second using arcsin transformed data. Again, there were significant differences across the pens.

- c. Discussion/Results: From the 2 x 2 Contingency Tables and ANOVA/Duncan's Tests it is apparent that 1) the most adverse effects took place in pen KO-3x and D-3x and that 2) the effects were significantly more adverse in pen KO-3x than in pen D-3x. In other words, the greater the exposure the greater the hazard, and Knox Out is more hazardous than diazinon emulsifiable at the higher rate. In addition, the same pattern holds true at the lower rate in that there is a significant difference in raw body weight shown between pens D-1x and KO-1x in the first 2-Way ANOVA/Duncan's Test.

The reason for the greater adverse effects in the Knox Out pens is due to the greater residues of diazinon on the grass (and at first in the soil) in the Knox Out pens. These data are tabulated below:

Diazinon Residues on Grass (ppm)

<u>Pen</u>	<u>Day 1</u>	<u>Day 4</u>	<u>Day 8</u>	<u>Day 14</u>	<u>Fraction of Day 1</u>
Control	0.01	--	--	--	
D-1x	314	122	25	13	0.04
KO-1x	582	613	187	90	0.15
D-3x	1491	601	189	115	0.08
KO-3x	1461	2141	700	553	0.22

Two birds apparently died at KO-1x, neither due to pesticide, based on necropsies.

Diazinon Residues on Soil (ppm)

<u>Pen</u>	<u>Day 1</u>	<u>Day 4</u>	<u>Day 8</u>	<u>Day 14</u>
Control	N.D. (<0.01)	--	--	--
D-1x	6.1	4.4	1.6	0.45
KO-1x	12.1	5.2	2.7	0.30
D-3x	21.7	12.0	8.2	1.16
KO-3x	39.0	10.5	11.4	0.29

Analytical recoveries of diazinon for known samples of Knox Out and diazinon emulsifiable were 96 and 90%, respectively.

d. Validation: SUPPLEMENTAL - Not Repairable.

(Use of bobwhite quail on turf is inappropriate, whereas the more sensitive mallard (lower dietary LC₅₀) as a turf-feeder would have been more appropriate; application rates on the proposed label are lower than those used in the test).

CELL FREQUENCY COUNTS

All Deaths
All Birds
(incl. replments)

		EXPOSURE (VAR 1)					TOTAL
		CONTROL	D-1x T1	KO-1x T2	KO-3x T3	D-3x T4	
		1.00	2.00	3.00	4.00	5.00	
RESPONSE ALIVE	1.00	I 12	12	12	8	11 I	55
(VAR 2) DEAD	2.00	I 1	0	2	16	5 I	24
TOTAL		I 13	12	14	24	16 I	79

MINIMUM NONZERO EXPECTED VALUE IS 3.646

----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC	VALUE	D.F.	PROB.
PEARSON CHISQUARE	25.062	4	0.0000

CELL FREQUENCY COUNTS

"Pestic" Deaths
All Birds
(incl replments)

		EXPOSURE (VAR 1)					TOTAL
		CONTROL	D-1x T1	KO-1x T2	KO-3x T3	D-3x T4	
		1.00	2.00	3.00	4.00	5.00	
RESPONSE ALIVE	1.00	I 12	12	12	8	11 I	55
(VAR 2) DEAD	2.00	I 0	0	0	13	3 I	16
TOTAL		I 12	12	12	21	14 I	71

MINIMUM NONZERO EXPECTED VALUE IS 2.704

----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC	VALUE	D.F.	PROB.
PEARSON CHISQUARE	29.128	4	0.0000

CELL FREQUENCY COUNTS

Non-Disease/Harassement Deaths
All Birds (incl replments)

		EXPOSURE (VAR 1)					TOTAL
		CONTROL	D-1x T1	KO-1x T2	KO-3x T3	D-3x T4	
		1.00	2.00	3.00	4.00	5.00	
RESPONSE ALIVE	1.00	I 12	12	12	8	11 I	55
(VAR 2) DEAD	2.00	I 0	0	1	16	4 I	21
TOTAL		I 12	12	13	24	15 I	76

MINIMUM NONZERO EXPECTED VALUE IS 3.316

----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC	VALUE	D.F.	PROB.
PEARSON CHISQUARE	30.043	4	0.0000

CELL FREQUENCY COUNTS

All Deaths
Original Birds
(exclude replacements)

		EXPOSURE (VAR 1)					TOTAL
		CONTROL	D-1x T1	KO-1x T2	KO-3x T3	D-3x T4	
		1.00	2.00	3.00	4.00	5.00	
RESPONSE ALIVE	1.00	I 11	12	10	0	7 I	40
(VAR 2) DEAD	2.00	I 1	0	2	12	5 I	20
TOTAL		I 12	12	12	12	12 I	60

MINIMUM NONZERO EXPECTED VALUE IS 4.000
 ----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES
 EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC VALUE D.F. PROB.
 PEARSON CHISQUARE 35.250 4 0.0000

CELL FREQUENCY COUNTS

"Pestic" Deaths
Original Birds
(exclude replacements)

		EXPOSURE (VAR 1)					TOTAL
		CONTROL	D-1x T1	KO-1x T2	KO-3x T3	D-3x T4	
		1.00	2.00	3.00	4.00	5.00	
RESPONSE ALIVE	1.00	I 11	12	10	0	7 I	40
(VAR 2) DEAD	2.00	I 0	0	0	11	3 I	14
TOTAL		I 11	12	10	11	10 I	54

MINIMUM NONZERO EXPECTED VALUE IS 2.593
 ----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES
 EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC VALUE D.F. PROB.
 PEARSON CHISQUARE 43.065 4 0.0000

CELL FREQUENCY COUNTS

Non-Disease/Harassment Deaths
All Birds (exclude replacements)

		EXPOSURE (VAR 1)					TOTAL
		CONTROL	D-1x T1	KO-1x T2	KO-3x T3	D-3x T4	
		1.00	2.00	3.00	4.00	5.00	
RESPONSE ALIVE	1.00	I 11	12	10	0	7 I	40
(VAR 2) DEAD	2.00	I 0	0	1	12	4 I	17
TOTAL		I 11	12	11	12	11 I	57

MINIMUM NONZERO EXPECTED VALUE IS 3.281
 ----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES
 EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC VALUE D.F. PROB.
 PEARSON CHISQUARE 40.494 4 0.0000

CELL FREQUENCY COUNTS

All Deaths

All Birds (incl replments)

RESPONSE (VAR 2)

		ALIVE 1.00	DEAD 2.00	TOTAL	
EXPOSURE CONTROL	1.00	I 8	16 I	24	Knox Out- 3x
(VAR 1) CHEMICAL	2.00	I 11	5 I	16	Diazinon- 3x
TOTAL		I 19	21 I	40	

MINIMUM NONZERO EXPECTED VALUE IS 4.000

----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(1-TAIL)			0.0299
PEARSON CHISQUARE	4.829	1	0.0280

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(2-TAIL)			0.0515
YATES° CORRECTED CHISQUARE	3.513	1	0.0609

CELL FREQUENCY COUNTS

All Deaths

Original Birds
(excl replmnts)

RESPONSE (VAR 2)

		ALIVE 1.00	DEAD 2.00	TOTAL	
EXPOSURE CONTROL	1.00	I 0	12 I	12	Knox Out- 3x
(VAR 1) CHEMICAL	2.00	I 7	5 I	12	Diazinon- 3x
TOTAL		I 7	17 I	24	

MINIMUM NONZERO EXPECTED VALUE IS 3.500

----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(1-TAIL)			0.0023
PEARSON CHISQUARE	9.882	1	0.0017

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(2-TAIL)			0.0046
YATES° CORRECTED CHISQUARE	7.261	1	0.0070

CELL FREQUENCY COUNTS

<u>All Deaths</u> All Birds (incl replmnts)		RESPONSE(VAR 2)			
		ALIVE 1.00	DEAD 2.00	TOTAL	
EXPOSURE CONTROL	1.00	I 8	13 I	21	Knox Out- 3x
(VAR 1) CHEMICAL	2.00	I 11	3 I	14	Diazinon- 3x
TOTAL		I 19	16 I	35	

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MINIMUM NONZERO EXPECTED VALUE IS 6.400
 ----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES
 EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(1-TAIL)			0.0210
PEARSON CHISQUARE	5.546	1	0.0185

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(2-TAIL)			0.0364
YATES° CORRECTED CHISQUARE	4.034	1	0.0446

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CELL FREQUENCY COUNTS

<u>"Pestic" Deaths</u> Original Birds (excl replmnts)		RESPONSE(VAR 2)			
		ALIVE 1.00	DEAD 2.00	TOTAL	
EXPOSURE CONTROL	1.00	I 0	11 I	11	Knox Out- 3x
(VAR 1) CHEMICAL	2.00	I 7	3 I	10	Diazinon- 3x
TOTAL		I 7	14 I	21	

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MINIMUM NONZERO EXPECTED VALUE IS 3.333
 ----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES
 EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(1-TAIL)			0.0010
PEARSON CHISQUARE	11.550	1	0.0007

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(2-TAIL)			0.0010
YATES° CORRECTED CHISQUARE	8.615	1	0.0033

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CELL FREQUENCY COUNTS

Nondisease/Harassment Deaths
All Birds (incl. replmnts)

RESPONSE(VAR 2)

		ALIVE		DEAD		TOTAL	
		1.00		2.00			
EXPOSURE CONTROL	1.00	I	8		16	I	24
(VAR 1) CHEMICAL	2.00	I	11		4	I	15
TOTAL		I	19		20	I	39

Knox Out- 3x
Diazinon- 3x

MINIMUM NONZERO EXPECTED VALUE IS 7.308

----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(1-TAIL)			0.0171
PEARSON CHISQUARE	5.912	1	0.0150

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(2-TAIL)			0.0225
YATES° CORRECTED CHISQUARE	4.419	1	0.0355

CELL FREQUENCY COUNTS

Nondisease/Harrassment Deaths
Original Birds
(excl replmnts)

RESPONSE(VAR 2)

		ALIVE		DEAD		TOTAL	
		1.00		2.00			
EXPOSURE CONTROL	1.00	I	0		12	I	12
(VAR 1) CHEMICAL	2.00	I	7		4	I	11
TOTAL		I	7		16	I	23

Knox Out- 3x
Diazinon- 3x

MINIMUM NONZERO EXPECTED VALUE IS 3.348

----- ALL STATISTICS ARE COMPUTED USING THE ORIGINAL CELL FREQUENCIES EXCLUDING ROWS AND/OR COLUMNS WHICH ARE ZERO.

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(1-TAIL)			0.0013
PEARSON CHISQUARE	10.977	1	0.0009

STATISTIC	VALUE	D.F.	PROB.
FISHER EXACT TEST(2-TAIL)			0.0013
YATES° CORRECTED CHISQUARE	8.177	1	0.0042

11:13 MONDAY, AUGUST 11

2-WAY ANOVA/RAW BODY WT

ANALYSIS OF VARIANCE PROCEDURE

DUNCAN'S MULTIPLE RANGE TEST FOR VARIABLE Y

MEANS WITH THE SAME LETTER ARE NOT SIGNIFICANTLY DIFFERENT

ALPHA LEVEL=.05 DF=165 MS=675.48

GROUPING	MEAN	N	TRT	<u>Pens</u>
A	240.972222	36	B	- <u>Diazinon-1x</u>
A				
A	233.611111	36	A	- <u>Control</u>
B				
B	226.944444	36	C	- <u>Knox Out-1x</u>
B				
B	222.916667	36	D	- <u>Knox Out-3x</u>
B				
B	222.777778	36	E	- <u>Diazinon-3x</u>

B

11:13 MONDAY, AUGUST 11

2-WAY ANOVA/RAW BODY WT

DUNCAN'S MULTIPLE RANGE TEST FOR VARIABLE Y

MEANS WITH THE SAME LETTER ARE NOT SIGNIFICANTLY DIFFERENT

ALPHA LEVEL=.05 DF=165 MS=675.48

GROUPING	MEAN	N	REP	<u>Sample Time</u>
A	264.000000	60	1	
B	225.666667	60	2	
C	198.666667	60	3	

13

8:27 TUESDAY, AUGUST 12

2-WAY ANOVA/PERC DIFF 2 of Sample Times
1 to 2 & 1 to 3

ANALYSIS OF VARIANCE PROCEDURE

DUNCAN'S MULTIPLE RANGE TEST FOR VARIABLE Y

MEANS WITH THE SAME LETTER ARE NOT SIGNIFICANTLY DIFFERENT

ALPHA LEVEL=.05 DF=110 MS=69.0507

GROUPING	MEAN	N	TRT	<u>Pens</u>
A	22.968333	24	D	- <u>KO-3x</u>
A				
B A	21.563333	24	E	- <u>D-3x</u>
B A				
B A	19.712500	24	C	- <u>KO-1x</u>
B A				
B	17.190000	24	B	- <u>D-1x</u>
B				
B	16.492500	24	A	- <u>Control</u>

8:27 TUESDAY, AUGUST 12

2-WAY ANOVA/PERC DIFF 2

DUNCAN'S MULTIPLE RANGE TEST FOR VARIABLE Y

MEANS WITH THE SAME LETTER ARE NOT SIGNIFICANTLY DIFFERENT

ALPHA LEVEL=.05 DF=110 MS=69.0507

GROUPING	MEAN	N	REP	
A	24.757167	60	2	Perc Diff of Sample Times
B	14.413500	60	1	1 to 2 & 1 to 3

14

8:33 TUESDAY, AUGUST 13

2-WAY ANOVA/ARCSIN 2 (of Perc Diff 2)

ANALYSIS OF VARIANCE PROCEDURE

DUNCAN'S MULTIPLE RANGE TEST FOR VARIABLE Y

MEANS WITH THE SAME LETTER ARE NOT SIGNIFICANTLY DIFFERENT

ALPHA LEVEL=.05 DF=110 MS=33.2391

GROUPING	MEAN	N	TRT	<u>Pens</u>
A	27.894583	24	D	- <u>KO-3x</u>
A				
B	26.682917	24	E	- <u>D-3x</u>
B				
B	25.377917	24	C	- <u>KO-1x</u>
B				
B	24.195000	24	B	- <u>D-1x</u>
B				
B	23.884583	24	A	- <u>Control</u>

8:33 TUESDAY, AUGUST

2-WAY ANOVA/ARCSIN 2

ANALYSIS OF VARIANCE PROCEDURE

DUNCAN'S MULTIPLE RANGE TEST FOR VARIABLE Y

MEANS WITH THE SAME LETTER ARE NOT SIGNIFICANTLY DIFFERENT

ALPHA LEVEL=.05 DF=110 MS=33.2391

GROUPING	MEAN	N	REP	<u>Arcsin Trans- formation of Pere Diff 2</u>
A	29.377667	60	2	
B	21.836333	60	1	

15

8:35 TUESDAY, AUGUST 1

1-WAY ANOVA/PERC DIFF 1 of Sample Times
1 to 2 & 1 to 3

ANALYSIS OF VARIANCE PROCEDURE

DUNCAN'S MULTIPLE RANGE TEST FOR VARIABLE Y

MEANS WITH THE SAME LETTER ARE NOT SIGNIFICANTLY DIFFERENT

ALPHA LEVEL=.05 DF=55 MS=100.878

GROUPING		MEAN	N	TRT	<u>Pens</u>
	A	20.355000	12	D	- <u>KO-3x</u>
	A				
	A	19.825000	12	E	- <u>D-3x</u>
	A				
B	A	13.010833	12	C	- <u>KO-1x</u>
B					
B	C	7.944167	12	B	- <u>D-1x</u>
	C				
	C	3.011667	12	A	- <u>Control</u>

8:38 TUESDAY, AUGUST 1

1-WAY ANOVA/ARCSIN 1 (of Perc Diff 1)

DUNCAN'S MULTIPLE RANGE TEST FOR VARIABLE Y

MEANS WITH THE SAME LETTER ARE NOT SIGNIFICANTLY DIFFERENT

ALPHA LEVEL=.05 DF=55 MS=62.4267

GROUPING		MEAN	N	TRT	<u>Pens</u>
	A	25.484167	12	D	<u>KO-3x</u>
	A				
	A	24.685000	12	E	<u>D-3x</u>
	A				
B	A	20.359167	12	C	<u>KO-1x</u>
B					
B		16.149167	12	B	<u>D-1x</u>
	C	9.370833	12	A	<u>Control</u>

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DATA EVALUATION RECORD

1. Chemical: Diazinon
2. Formulation: Knox Out 2FM (23% Microencapsulated)
3. Citation: Beavers, J.B. (1978a) Acute Oral LD₅₀ - Bobwhite Quail, Knox Out 2FM, Final Report, Project No. 110-120; Prepared by Wildlife Intl. Ltd.; Submitted Pennwalt Corp. (Accession No. 240993).
4. Reviewed by: John S. Leitzke
Ecologist, Section 3
EEB/HED
5. Date Reviewed: July 22, 1980
6. Test Type: Avian Acute Oral LD₅₀
Test Species: Bobwhite Quail (Colinus virginianus)
7. Reported Results:

LD₅₀ = 472 (380 to 587) mg/kg total test material (23% diazinon).
8. Reviewer's Conclusions:

In terms of active ingredient (ai), the LD₅₀ equals 108.5 (87.5 to 135) ai mg/kg when using corn oil as the carrier for the microencapsulated test material. Although the study is sound in most respects, it is unacceptable in meeting the Guidelines minimum data requirement for an avian acute oral LD₅₀ on the formulation, Knox Out 2FM, because of the use of corn oil as the carrier for the microencapsulated test material and a significant difference (P>0.01) between treatment and control groups by average initial body weight.

9. Materials and Methods:

The test material is the formulated product Knox Out 2FM (23% diazinon) since this test using the formulation is required for registration.

Bobwhite quail at 20 weeks of age were assigned 10 to a group (5 of both sexes), housed in Beacon Battery Brooders, exposed to a 14L:10D lighting regime and fasted before treatment. Corn oil was used as the carrier.

- a. Statistical Analysis: The reported dose-response data were analyzed using the Finney Probit Program, individual initial body weights of birds within treatment groups using the 1-Way ANOVA program and average initial body weights of birds between treatment and control groups using the 2-Sample T-Test on EEB's TI-59 calculator (attached).
- b. Discussion/Results: There was no mortality in any of the 5 control groups. Mortalities in the treatment groups occurred within the first several days of the 14-day observation period decreased body weight gain occurred in as low as the 398 mg/kg test material group and reduced feed consumption in the 631 mg/kg group. In the lowest group (251 mg/kg; 1/10 mortality) lethargy, loss of coordination and lower limb weakness were observed for the first 2 days. In the 398 mg/kg group (4/10 mortality) loss of coordination and ataxia were observed on the first day with lethargy continuing through the first 4 days. In the 631 mg/kg group (6/10 mortality) similar symptoms occurred through the first 5 to 6 days.

10. Reviewer's Evaluation

- a. Test Procedure: The test procedure generally complies with recommended protocol. However, the use of corn oil is considered an inappropriate carrier for the microencapsulated test material.
- b. Statistical Analysis: The Chisquare statistic from the Finney Probit Program indicates a homogeneous dose-response relationship within the test groups.

The F statistic from the 1-Way ANOVA program indicates a homogeneous distribution of birds to test groups by body weight.

The 2 - Sample T-Test, however, showed a significant difference ($p > 0.01$) between treatment and control groups by average body weight.

c. Discussion/Results: The reported LD₅₀ is lower than the recalculated results and will be used in all future considerations based on this test using corn oil as the carrier.

d. Validation:

1. Supplemental
2. Not Repairable

(Use of corn oil as a carrier for the microencapsulated test material; significant difference ($p > 0.01$) between treatment and control groups by average initial body weight).

Diazinon-Knox Out

Bobw Q - LD50

Wildl Intl 78

	----0.	PGM
2-Sample T		
Aveg Init	559.49	PRT
Bwt	192.	
	200.	
<u>Treatment</u>	195.	
	198.	
	196.	<u>Control</u>
	206.	
	196.	
	200.	
	198.	
	202.	

-3.466162967
8.-df
p >.01

Diazinon- Knox Out

Bbw Q

Wildl Intl 78

	200.		199.
ANOVA	179.		196.
Initial	192.		215.
Bwt	201.		217.
	185.	251	203.
	202.		188.
	184.		201.
	191.		172.
	190.		183.
	199.		182.

192.3 195.6
58.01 188.84

Diazinon - Knox Out

Bobwhite Q - LD50

Wildl Intl

78

	----	CLR
	251.	
	1.	
	10.	
	398.	
	4.	
	10.	
	631.	
	6.	
	10.	

	185.		212.
	187.		180.
	216.		204.
	174.	398	201.
	204.		202.
	193.		203.
	186.		192.
	196.		211.
	207.		181.
	203.		197.

195.1 198.3
142.09 110.01

1000. 182. .2852266275 NS
10. 172. 4.
10. 184. 45.

4.898 M
8.105 YINT
df=2 1.600 LW M
1.538 CHI²

	204.	631	
	217.		7221.1
	207.		183.08
	177.		7404.18
	211.		
	206.		
	198.		

NS
473.884 LD50
379.407 LOCL
591.888 UPCL

195.8
223.16

259.367 LD10
176.165 LOCL
381.864 UPCL

865.826 LD90
596.975 LOCL

DATA EVALUATION RECORD

1. Chemical: Diazinon
2. Formulation: Knox Out 2FM (23% Microencapsulated)
3. Citation: Beavers, J.B. (1978c) Eight-Day Dietary LC₅₀ - Mallard Duck Knox Out 2FM, Final Report, Project No. 110-120; Prepared by Wildlife Intl. Ltd.; Submitted Pennwalt Corp. (Accession No. 240993).
4. Reviewed by: John S. Leitzke
Ecologist, Section 3
EEB/HED
5. Date Reviewed: July 17, 1980
6. Test Type: Avian 5(+3)-Day Dietary LC₅₀
Test Species: Mallards (Anas platyrhynchos)
7. Reported Results:
LC₅₀ = 649 (464-908) ppm of test material (23% diazinon).
8. Reviewer's Conclusions:

In terms of active ingredient (ai), the LC₅₀ equals 149 (107 to 209) ppm ai, indicating a high toxicity to avian wildlife such as water fowl in their diet. The study is scientifically sound and is acceptable in meeting the Guidelines minimum data requirement for an avian 5(+3)-day dietary LC₅₀ using a waterfowl on the formulation, Knox Out 2FM.

9. Materials and Methods:

The test material is the formulated product Knox Out 2FM (23% diazinon) since this test using the formulation is required for registration.

Mallard ducklings at 14 days of age were 10 to a group and exposed to a 14L: 10D lighting regime. Examination of each groups average initial body weights indicated a random, nonheterogeneous assignment of birds to test and control groups. Test birds were exposed to treated feed for 5-days followed by 3-days observation on clean feed.

- a. Statistical Analysis: The reported dose-response data were analyzed on EEB's TI-59 calculator using the Finney Probit Program (attached).
- b. Discussion/Results: There was no mortality in any of the five control groups. Decreases in body weight gain and feed consumption were noted all test groups the lowest being 23 ppm ai. Depression, reduced reaction to external stimuli and loss of coordination were some of the major symptoms noted. Not all deaths occurred in the first several days; several occurred in the last part of the test.

10. Reviewer's Evaluation:

- a. Test Procedures: The test procedure generally complies with recommended protocol.
- b. Statistical Analysis: The Chisquare statistic indicated a homogeneous dose-response relationship within the test groups.
- c. Discussion/Results: The reported LC₅₀ is less than the recalculated value and will be used in all hazard evaluations.
- d. Validation: Core.

Diazinon Knox Out

Mallard - LC50

Wildl Int'l 100.
78 0.
10.

178.
2.
10.

316.
2.
10.

562.
3.
10.

1000.
6.
10.

1780.
9.
10.

3160.
10.
10.

df=5 2.497 M
-2.032 YINT
2.515 LW M
3.836 CHI²

NS
655.498 LD50
464.597 LOCL
924.839 UPCL

200.952 LD10
115.219 LOCL
350.480 UPCL

2138.208 LD90
1199.116 LOCL
3812.754 UPCL

DATA EVALUATION RECORD

Core for formulated product

1. Chemical: Diazinon
2. Formulation: Knox Out 2FM (23% Microencapsulated)
3. Citation: Beavers, J.B. (1978b) Eight-Day Dietary LC₅₀ - Bobwhite Quail 2FM, Final Report, Project No. 110-121; Prepared by Wildlife Intl. Ltd.; Submitted Pennwalt Corp. (Accession No. 240993).
4. Reviewed by: John S. Leitzke
Ecologist, Section 3
EEB/HED
5. Date Reviewed: July 17, 1980
6. Test Type: Avian 5(+3)-Day Dietary LC₅₀
Test Species: Bobwhite Quail (Colinus virginianus)
7. Reported Results:

LD₅₀ = 1515 (1147-2002) ppm of test material (23% diazinon).

8. Reviewer's Conclusions:

In terms of active ingredient (ai), the LC₅₀ equals 345 (259-461) ppm ai, indicating a high toxicity to avian wildlife such as upland gamebirds in their diet. The study is scientifically sound and is acceptable in meeting the Guidelines minimum data requirement for an avian 5(+3)-day dietary LC₅₀ using an upland gamebird on the formulation, Knox Out 2FM.

9. Materials and Methods:

The test material is the formulated product Knox Out 2FM (23% diazinon) since this test using the formulation is required for registration.

Bobwhite quail chicks at 14 days of age were assigned 10 to a group and exposed to a 14L:10D lighting regime. Examination of each group's average initial body weights indicated a random, nonheterogeneous assignment of birds to test and control groups. Test birds were exposed to treated feed for 5 days followed by 3-days observation on clean feed.

- a. Statistical Analysis: The reported dose-response data were analyzed on EEB's TI-59 calculator using the Finney, Probit Program (attached).
- b. Discussion/Results: There was only 1 mortality in the 5 control groups. Decreases in body weight gain and feed consumption were noted in the 129 ppm ai and higher groups. Depression, reduced reaction to external stimuli, weakness and loss of coordination were some of the major symptoms noted. Not all deaths occurred in the first several days; several occurred in the last part of the test.

10. Reviewer's Evaluation:

- a. Test Procedure: The test procedure generally complied with recommended protocol.
- b. Statistical Analysis: The Chi-square statistic indicated a homogeneous dose-response relationship within the test groups.
- c. Discussion/Results: The LC₅₀ was recalculated from the given dose-response data as 1503 (1127-2005) ppm total test material.
- d. Validation: Core.

Diazinon Knox Out

Bobw O - LC50

Wildl Int'l 562.

78 0.

10.

1000.

4.

10.

1780.

6.

10.

3160.

8.

10.

5620.

10.

10.

3.563.

M

-6.319

YINT

1.908

LW M

2.356

CHI²

1503.350

LD50

1127.314

LOCL

2004.819

UPCL

656.469

LD10

404.091

LOCL

1066.471

UPCL

3442.754

LD90

2176.540

LOCL

5445.593

UPCL