EED

079401

MRID 146843

DATA EVALUATION RECORD

71-4

1. Chemical: Endosulfan

2. Test Material: Technical, 97.2 percent

3. Study Type: Avian Reproduction

   Species tested: Mallard Duck
   (Anas platyrhynchos)

4. Study ID: Roberts, N., C. Phillips, A. Anderson, I. Dawe, D. Chanter and S. Cook. (1985) The Effects of Dietary Inclusion of Endosulfan-Technical (Code: HOE002671 OI2D970003) on Reproduction in the Mallard Duck. Performed by Huntingdon Research Centre, England; submitted by American Hoechst Corp., Somerville, NJ. Registration Number 8340-13; Accession No. 256129.

5. Reviewed by: John J. Bascietto
   Wildlife Biologist
   EEB/HED
   Signature: John J. Bascietto
   Date: 13 Sept 185

6. Approved by: Dave Coppage
   Supervisory Biologist
   EEB/HED
   Signature: Dave Coppage
   Date: 13/Sept/85

7. Conclusions:

The study is scientificaly sound and fulfills the requirement of the Pesticide Assessment Guidelines for a reproduction study with a wild waterfowl species.

However, the conclusion of the authors (i.e., that a NOEL = 60 ppm) is rejected because it is inconsistent with the data obtained. EEB's evaluation indicates that statistically significant impairment of egg production occurred at all levels tested (30, 60, 120 ppm) in the biologically significant first half of the experimentally induced laying period. A NOEL was not established. Therefore, we conclude that endosulfan can cause significant impairment of avian reproduction at any dietary level.

8. Recommendations:

Until a "NOEL" for impairment of egg production is established, there is no assessable margin of safety for endosulfan's effects on avian reproduction.

9. Background:

The study was submitted in response to the Registration Standard, but was 2 years late.

10. Discussion of Individual Test:

N/A

11. Materials and Methods:
(Definitive Test)

a. Test Animals - Forty-eight male and 120 female mallards (Anas platyrhynchos) were obtained to serve as test birds, plus 12 males and 12 females for use as replacement birds. Source was Mr. John Coles, County Game Farms, Ashford, Kent, England. They were about 31 weeks old on arrival at facility; 33 weeks old at start of study. Birds were maintained on a 7 hrs light/17 hrs dark photoperiod from time of arrival until week 7 of the study. Birds were individually identified by metal wing tags.

b. Dose - Dietary inclusion of toxicant to treatment groups. Toxicant was dispersed and mixed in basal diets by acetone (to dissolve) and corn oil (for dispersal). Toxicant and dispersants were added to 500 g of basal diet to make a 2 percent premix. This was stirred at 40 °C to evaporate acetone, then more basal diet was added to complete the premix and shaken in polyethene bags for 3 minutes. Aliquots of premix were used to prepare final diets, which were made in 40 kg batches in large blenders. Diet samples were taken for analysis at various times during the study.

Birds were fed control or test diets for 12 weeks prior to the start of egg production, then for an additional 12 weeks during egg production. A 4-week "withdrawal" period was added (weeks 23, 24, 25, and 26) in which birds were fed "clean" basal diets only (untreated).

c. Design - Seven days prior to treatment period the ducks were randomized to study pens. Each study pen, or replicate, consisted of two males and five females. There were 4 treatment groups: a) 0 ppm control; b) 30 ppm; c) 60 ppm; and d) 120 ppm with 6 replicates

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each group --a total of 48 males and 120 female ducks. An additional 12 males and 12 females were used as replacement birds.

Adult birds were maintained in galvanized steel floor pens, 1.2 m x 1.5 m with solid sides and wire mesh floors. Each pen had an automatic cup drinker and food hopper. Floors were covered with plastic padded matting to reduce risk of cracked eggs. Maximum and minimum temperature and relative humidity were recorded once daily.

At the start of week 8 lighting was increased from 7 hours per day to 16 hours per day and maintained at this level until the end of week 14. At week 15, photoperiod was lengthened by 15 minutes per week until the end of the study. (Low light intensity was used during weeks 1 to 6 to prevent birds from coming into egg production too early.)

Summary of Design

Group	Treatment	No. of Replicates	Birds per Replicates M	Birds per Replicates F	Birds per Treatment M	Birds per Treatment F
A	Control	6	2	5	12	30
B	30 ppm	6	2	5	12	30
C	60 ppm	6	2	5	12	30
D	120 ppm	6	2	5	12	30

Chicks were housed in floor pens with concrete floors. Each pen contained two automatic drinkers and one food hopper. Wood shavings were used as bedding. A 300-watt infra-red lamp placed at bird level supplied additional heat. Chicks were fed standard chick diet with no antibiotic or other growth promoter.

d. Statistics - Generally, analysis of variance (ANOVA) was used to statistically examine differences of the following parameters between groups and control.

1. Adult food consumption
2. Adult mortality and body weight
3. Number of eggs laid and proportion damaged
4. Egg weight
5. Egg shell thickness
6. Number of infertilities, embryonic deaths, and hatchings
7. Numbers of 14-day old surviving chicks
8. Chick bodyweights at hatching and 14 days later.

Specific statistical methods are described in the Appendix to this DER.

12. Reported Results:

The authors reported the following:

- At all dietary levels of endosulfan general behavior and health as well as food consumption were not affected. Bodyweights were reduced the first 2 weeks at 60 ppm and markedly so at 120 ppm. After first 2 weeks, bodyweight gains were similar in all groups.

- No treatment-related mortality was observed.

- Autopsies of birds dying on test and those sacrificed after the study ended revealed no treatment-related findings.

- Adult mortality was not significantly different among groups during the prelaying period; however, there was a significant ($p < 0.05$) difference during the egg-laying period at 120 ppm. The majority of deaths occurred in female birds and was thought to be mostly due to "bullying."

- A dietary concentration of 120 ppm impaired egg production, and resulted in a higher proportion of cracked and broken eggs than in the control during the first half of the egg production period.

- "At dietary concentrations of 30 ppm and 60 ppm the egg production was within the range of normal variation and there was no indication of any reproductive impairment as assessed from egg weights, egg shell thickness, number of infertile eggs, early and late embryonic death, hatching, chick health and mortalities, chick bodyweights and number of 14-day survivors." (p.i)

13. Study Authors' Conclusions/QA Measures

The study authors concluded: "Under the conditions of this test, and taking the results as a whole, it was concluded that the dietary concentration of 60 ppm of Endosulfan technical, equivalent to an estimated intake of approximately 10 mg/kg/day, represented the "no toxic effect level" for reproductive impairment in the Mallard duck." (p. i)

14. Reviewer's Discussion and Interpretation of the Study

a. Test Procedures: The test was conducted in close agreement with protocols for avian reproduction studies recommended by the Pesticide Assessment Guidelines, Subdivision E.

However, the dose levels were based on a preliminary feeding study, not on expected environmental concentrations, although the levels tested would be expected in a limited number of uses.

b. Statistical Analysis: EEB accepts the statistical analysis presented by the registrant (Appendix 12 of report - attached).

c. Discussion/Results: Although several mortalities of treatment and control birds occurred during this study, most deaths are attributable to aggression. Enough replacement replicates were available to obtain statistical validity. We emphasize that the mortality observed, while high only in treatment groups, does not invalidate this test because these deaths are clearly attributable to aggression.

Regarding the ANOVA for egg production, the number of eggs produced was not corrected for mortalities during the laying period. The report claims that this is the correct approach because if the mortalities were related to treatments, correction would introduce bias into the analysis. [N.B. - the report also finds that mortalities were not treatment-related, but rather due to "bullying" by males. Two females died at 30 ppm during egg production; one female at 60 ppm; four females at 120 ppm. None were replaced during this period (early in the egg-laying phase - weeks 10, 11, and 12). EEB agrees with noncorrection approach taken to avoid introducing bias.]

When a general linear model was applied to replicate data, it was found that all treatment groups laid significantly fewer eggs than controls during weeks 11 to 16 (first half of laying period). Block analysis showed no significant differences; treatment analysis showed the following P values for differences from controls: 30 ppm, $P < 0.05$; 60 ppm, $P < 0.05$; 120 ppm, $P < 0.01$ (a stronger statistical correlation at 120 ppm could be an artifact because of the smaller number of producing hens in this group). During weeks 17 to 22 (second half of laying period) only the high treatment, 120 ppm, produced significantly fewer eggs than the control ($P < 0.01$).

Differences in numbers of eggs produced in each period (first weeks 11 to 16; second - weeks 17 to 22) were less for the treatment groups and the control group: low $P < 0.05$, intermediate $P < 0.01$, high $P < 0.01$. In all cases however, actual egg production was greater during weeks 11 to 16 of the study, as would be expected, since this initial period is less physiologically stressful (compared to the overall test). It is interesting to

note that while supressing egg production in all treatment groups, the toxicant also reduced the difference (deficit) in egg production numbers accompanying the induced lengthy production period.

It is our judgment at this time that the impairment of egg production during the important (and perhaps more representative as an actual breeding scenario) initial half of the egg laying phase, i.e., weeks 11 to 16 of the study, represents a biologically significant effect. This effect is adverse, i.e., significantly reduces the potential to reproduce young, occurs at all dietary levels tested, and is directly caused by the toxicant, endosulfan.

A statistical difference between the first and second halves of the laying period is again encountered with significantly ($P < 0.01$) more eggs cracked by the high treatment during weeks 11 to 16 compared to the controls.

Eggs mass was significantly lower in all treatment groups for weeks 11 to 16, and for 120 ppm weeks 17 to 22.

The other reproductive parameters examined did not show statistical differences between treatments and controls either between first and second laying periods, or the overall test.

The study is scientifically sound and fulfills the requirements of the Pesticide Assessment Guidelines, Subdivision E. However, the conclusions of the author (NOEL = 60 ppm) are rejected as they are inconsistent with the data obtained.

The study shows that endosulfan impaired the reproductive capacity of the treated mallards, with statistical significance. This was evident through analysis of variance of egg production in the initial one-half of the induced laying period (considered as biologically significant). This occurred at all levels tested (30, 60, and 120 ppm). A "no-effect" level was not established. Until a NOEL is established, it is assumed that these effects could occur at any level of endosulfan. This means there is no assessable margin of safety.

d. Adequacy of Study:

1. Classification: Core
2. Rationale: Guidelines study
3. Repair: N/A

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15. One-liner Completed:

September 10, 1985

16. CBI Appendix:

Data, results and statistical analyses attached.

Endosulfan OECD 6

Page_____ is not included in this copy.

Pages 8 through 28 are not included.

The material not included contains the following type of information:

____ Identity of product inert ingredients.

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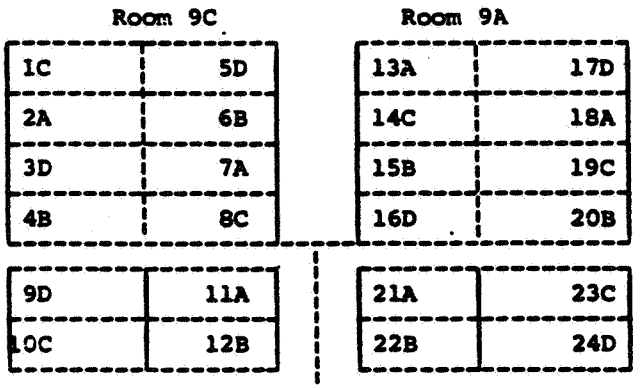
(continued)

TABLE S1

Allocation of treatment to pens



A : Control
B : Endosulfan 30 ppm
C : Endosulfan 60 ppm
D : Endosulfan 120 ppm
--- Block boundaries

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(continued)

TABLE S2

Analysis of variance of total food consumption (weeks 1-22)

Source of variation	Degrees of freedom	Sums of squares	Mean square	Variance ratio
Treatment	3	481310	160437	0.76
Blocks	5	1254982	250996	1.18
Residual	15	3181713	212114	
Total	23	4918005	213826	

TABLE S3

Mean total food consumption of adult birds (g/bird/day)

Treatment group	Pre-laying weeks 1-10	Egg-laying weeks 11-22	Full study weeks 1-22
Control	1574	2135	3708
Low	1577	2255	3831
Intermediate	1778	2322	4099
High	1718	2184	3902
Residual variance	49257	104938	212114

(continued)

TABLE S4

Treatment means (averaged over sex) for rate of change and mean bodyweights

Treatment group	Rate of change in bodyweight (g/fortnight)	Mean bodyweight (g) over 70-day period
Control	31.4	1046
Low	16.7**	1011*
Intermediate	12.5**	989**
High	8.9**	945**
Residual variance	335.7	6083
Treat[1]	12.20***	12.14***
Block[2]	2.59*	1.56
Sex[3]	1.29	17.02***
Int[4]	0.53	0.24

F-ratios: [1] Treatment effect (3,158 d.f.)
[2] Block effect (5,158 d.f.)
[3] Sex effect (1,158 d.f.)
[4] Treatment.sex interaction (3,155 d.f.)

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(continued)

TABLE S5

Day 154 Bodyweights-treatment means (averaged over sex)

Treatment group	Day 154 Bodyweights (g)
Control	1073
Low	1033*
Intermediate	1027*
High	1001**
Residual variance	6691
Treat[1]	5.06**
Block[2]	2.25*
Sex[3]	36.07***
Int[4]	0.78

F-ratios: [1] Treatment effect (3,147 d.f.)
[2] Block effect (5,147 d.f.)
[3] Sex effect (1,147 d.f.)
[4] Treatment.sex interaction (3,144 d.f.)

(continued)

TABLE S6

Data and analysis of numbers of eggs laid (per pen) in weeks 11-16

	Treatment			
	Control	Low	Intermediate	High
Block 1	140	123	133	22
2	141	113	98	90
3	113	106	95	38
4	152	61	94	20
5	115	93	103	27
6	130	78	57	75
Means :	132	96*	97*	45**

Residual variance : 558.3
Treat[1] 13.61***
Block[2] 1.04

F-ratios: [1] Treatment effect (3,15 d.f.)
[2] Block effect (5,15 d.f.)

(continued)

TABLE S7

Data and analysis of numbers of eggs laid (per pen) in weeks 17-22

	Treatment			
	Control	Low	Intermediate	High
Block 1	28	39	40	0
2	55	52	36	14
3	53	39	76	28
4	41	6	22	0
5	43	31	53	3
6	46	39	52	46
Means :	44	34	46	15**

Residual variance : 147.1
Treat[1] 8.34 **
Block[2] 3.91 *

F-ratios: [1] Treatment effect (3,15 d.f.)
[2] Block effect (5,15 d.f.)

(continued)

TABLE S8

Mean difference in numbers of eggs laid (per pen) between periods

Treatment group	Differences in numbers of eggs laid
Control	-87.5
Low	-61.3*
Intermediate	-50.2**
High	-30.2**
Residual variance	401.0
Treat[1]	8.56**
Block[2]	2.70

F-ratios : [1] Treatment effect (3,15 d.f.)
[2] Block effect (5,15 d.f.)

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(continued)

TABLE S9

Proportion of eggs damaged in weeks 11-16 and estimated probabilities of damage for each treatment

	Treatment			
	Control	Low	Intermediate	High
Block 1	0.02	0.03	0.04	0.05
2	0.02	0.03	0.03	0.08
3	0.04	0.07	0.04	0.08
4	0.08	0.02	0.03	0.05
5	0.01	0.02	0.03	0.37
6	0.02	0.01	0.05	0.05
Estimated probability of damage:	0.03	0.03	0.03	0.10**

(continued)

TABLE S10

Proportion of eggs damaged in weeks 17-22 and estimated probabilities of damage for each treatment

	Treatment			
	Control	Low	Intermediate	High
Block 1	0.04	0.03	0.10	-
2	0.05	0.10	0.03	0.07
3	0.04	0.10	0.16	0.07
4	0.17	0.00	0.05	-
5	0.05	0.06	0.06	0.00
6	0.04	0.05	0.10	0.07
Estimated probability of damage:	0.06	0.07	0.09	0.06

- No eggs laid

(continued)

TABLE S11

Data and analysis of total egg mass (g/pen)
weeks 11-16

	Treatment			
	Control	Low	Intermediate	High
Block 1	8349	7507	7868	1285
2	8441	6905	5668	5052
3	6789	6600	5172	2148
4	9256	3673	5829	1150
5	6920	5628	6189	1735
6	7779	4497	3012	4747
Means :	7922	5802*	5623*	2686**

Residual variance : 2259879
Treat [1] 12.29***
Block [2] 0.84

F-ratios : [1] Treatment effect (3,15 d.f.)
[2] Block effect (5,15 d.f.)

(continued)

TABLE S12

Data and analysis of total egg mass (g/pen)
weeks 17-22

	Treatment			
	Control	Low	Intermediate	High
Block 1	1633	2455	2599	-
2	3323	3303	2153	805
3	3314	2609	4522	1814
4	2517	358	1369	-
5	2718	1912	3163	179
6	2814	2428	2872	3181
Means :	2720	2178	2780	1126*

Residual variance : 700442
Treat [1] 3.56*
Block [2] 2.45

F-ratios : [1] Treatment effect (3,13 d.f.)
[2] Block effect (5,13 d.f.)

- No eggs laid

(continued)

TABLE S13

Mean rates of change in egg weight and mean intercepts for treatments

Treatment group	Rate of change in egg weight (g/egg/week)	Intercept (g/egg)
Control	0.35	62
Low	0.51	62
Intermediate	0.50	60
High	0.78	64
Residual variance	0.1991	142.6
Treat[1]	0.99	2.24
Block[2]	1.22	1.26

F-ratios : [1] Treatment effect (3,15 d.f.)
[2] Block effect (5,15 d.f.)

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(continued)

TABLE S14

Mean shell thickness (mm)

Treatment group	Week 11	13	15	17	19	21
Control	0.31	0.31	0.31	0.34	0.30	0.31
Low	0.32	0.31	0.30	0.35	0.29	0.33
Intermediate	0.29	0.30	0.30	0.31	0.30	0.31
High	0.30	0.31	0.29	0.34	0.28	0.33
Residual variance	4.05 x10^{-4}	3.37 x10^{-4}	9.38 x10^{-4}	1.83 x10^{-4}	2.76 x10^{-4}	-+
Residual d.f.	16	20	20	16	15	
Treat[1]	2.44	0.40	0.39	6.00*	0.37	
Block[2]	0.88	2.07	1.23	0.83	1.19	

F-ratios : [1] Treatment effect (3, Residual d.f.)
[2] Block effect (5, Residual d.f.)

-+ Too few observations for analysis

(continued)

TABLE S15a

Observed proportions infertile of those set weeks 11-16

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. set	Prop.	No. set	Prop.	No. set	Prop.	No. set
Block 1	0.00	124	0.00	103	0.04	116	0.79	19
2	0.12	129	0.00	97	0.07	83	0.01	74
3	0.03	98	0.05	87	0.01	80	0.03	32
4	0.00	127	0.00	55	0.00	81	0.06	17
5	0.03	104	0.00	84	0.04	91	0.00	13
6	0.03	115	0.00	73	0.02	49	0.02	64

(continued)

TABLE S15b

Observed proportions infertile of those set weeks 17-22

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. set	Prop.	No. set	Prop.	No. set	Prop.	No. set
Block 1	0.00	23	0.00	35	0.81	32	-	0
2	0.46	52	0.05	42	0.72	32	0.07	15
3	0.02	49	0.19	37	0.02	61	0.00	24
4	0.00	31	0.00	5	0.56	18	-	0
5	0.03	39	0.12	25	0.69	51	1.00	3
6	0.00	37	0.00	34	0.55	49	0.07	43

(continued)

TABLE S16a

Observed proportion of early embryonic deaths of those fertile weeks 11-16

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. fert.	Prop.	No. fert.	Prop.	No. fert.	Prop.	No. fert.
Block 1	0.01	124	0.02	103	0.00	111	1.00	4
2	0.03	114	0.00	97	0.03	77	0.03	73
3	0.04	95	0.01	83	0.00	79	0.00	31
4	0.00	127	0.00	55	0.02	81	0.00	16
5	0.02	101	0.01	84	0.01	87	0.08	13
6	0.04	111	0.03	73	0.00	48	0.02	63

(continued)

TABLE S16b

Observed proportion of early embryonic deaths of those fertile weeks 17-22

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. fert.	Prop.	No. fert.	Prop.	No. fert.	Prop.	No. fert.
Block 1	0.00	23	0.06	35	0.50	6	*	*
2	0.04	28	0.05	40	0.89	9	0.00	14
3	0.08	48	0.03	30	0.03	60	0.00	24
4	0.00	31	0.00	5	0.25	8	*	*
5	0.00	38	0.00	22	0.19	16	-	0
6	0.00	37	0.06	34	0.18	22	0.02	40

* Excluded from analysis

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(continued)

TABLE S17a

Observed proportions of late embryonic deaths of those set on Day 14 weeks 11-16

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. set	Prop.	No. set	Prop.	No. set	Prop.	No. set
Block 1	0.06	123	0.11	101	0.09	111	-	0
2	0.07	111	0.12	97	0.12	75	0.03	71
3	0.18	91	0.05	82	0.09	79	0.13	31
4	0.04	127	0.11	55	0.18	79	0.19	16
5	0.10	99	0.14	83	0.14	86	0.17	12
6	0.10	107	0.10	71	0.27	48	0.03	62

(continued)

TABLE S17b

Observed proportions of late embryonic deaths of those set on Day 14 weeks 17-22

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. set	Prop.	No. set	Prop.	No. set	Prop.	No. set
Block 1	0.04	23	0.15	33	0.00	3	*	*
2	0.11	27	0.00	38	0.00	1	0.07	14
3	0.25	44	0.17	29	0.12	58	0.08	24
4	0.03	31	0.20	5	0.17	6	*	*
5	0.03	38	0.05	22	0.00	13	-	0
6	0.03	37	0.06	32	0.11	18	0.10	39

* Excluded from analysis

(continued)

TABLE S18a

Observed proportions hatching of those set on Day 21
weeks 11-16

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. set	Prop.	No. set	Prop.	No. set	Prop.	No. set
Block 1	0.63	116	0.74	90	0.80	101	-	0
2	0.70	103	0.80	85	0.58	66	0.58	69
3	0.81	75	0.73	78	0.58	72	0.63	27
4	0.80	122	0.57	49	0.38	65	0.46	13
5	0.70	89	0.55	71	0.59	74	0.90	10
6	0.73	96	0.69	64	0.51	35	0.42	60

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(continued)

TABLE S18b

Observed proportions hatching of those set on Day 21
weeks 17-22

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. set	Prop.	No. set	Prop.	No. set	Prop.	No. set
Block 1	0.73	22	0.71	28	0.67	3	*	*
2	0.88	24	0.79	38	1.00	1	0.69	13
3	0.82	33	0.79	24	0.78	51	0.91	22
4	0.90	30	0.50	4	0.80	5	*	*
5	0.78	37	0.52	21	0.77	13	*	*
6	0.81	36	0.87	30	0.69	16	0.77	35

* Excluded from analysis

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(continued)

TABLE S19a.

Observed proportions hatching of those fertile
weeks 11-16

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. fert.	Prop.	No. fert.	Prop.	No. fert.	Prop.	No. fert.
Block 1	0.59	124	0.65	103	0.73	111	0.00	4
2	0.63	114	0.70	97	0.49	77	0.55	73
3	0.64	95	0.69	83	0.53	79	0.55	31
4	0.76	127	0.51	55	0.31	81	0.38	16
5	0.61	101	0.46	84	0.51	87	0.69	13
6	0.63	111	0.60	73	0.38	48	0.40	63

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(continued)

TABLE S19b

Observed proportions hatching of those fertile weeks 17-22

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. fert.	Prop.	No. fert.	Prop.	No. fert.	Prop.	No. fert.
Block 1	0.70	23	0.57	35	0.33	6	*	*
2	0.75	28	0.75	40	0.11	9	0.64	14
3	0.56	48	0.63	30	0.67	60	0.83	24
4	0.87	31	0.40	5	0.50	8	*	*
5	0.76	38	0.50	22	0.63	16	-	0
6	0.78	37	0.76	34	0.50	22	0.67	40

* Excluded from analysis

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APPENDIX 12

(continued)

TABLE S20

Estimated probabilities of infertility, early and late embryonic death and hatching for each period

	Infertility from those initially set		Early embryonic death from those fertile		Late embryonic death from those set on Day 14		Hatching from those set on Day 21		Hatching from those fertile	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd
Control	0.03	0.08	0.02	0.02	0.09	0.07	0.73	0.82	0.65	0.72
Low	0.01	0.04	0.01	0.03	0.11	0.07	0.69	0.75	0.61	0.66
Intermediate	0.02	0.53	0.01	0.24	0.14	0.06	0.59	0.75	0.51	0.56
High	0.06	0.07	0.04	0.01	0.06	0.06	0.54	0.77	0.50	0.71
Residual d.f.	15	13	15	12	14	12	14	12	14	12
Treat[1]	1.38	6.90**	0.27	5.11*	1.49	0.14	2.43	1.05	2.38	1.23
Block[2]	0.76	2.16	0.17	1.26	0.46	2.15	0.58	1.24	0.73	0.23

1st : weeks 11-15
2nd : weeks 17-22

F-ratios : [1] Treatment effect (3, Residual d.f.)
[2] Block effect (5, Residual d.f.)

(continued)

TABLE S21a

Observed proportions of 14-day old surviving chicks and estimated probabilities of surviving weeks 11-16

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. hat.	Prop.	No. hat.	Prop.	No. hat.	Prop.	No. hat.
Block 1	0.89	73	0.85	67	0.89	81	*	*
2	0.88	72	0.85	68	0.58	38	0.85	40
3	0.84	61	0.89	57	0.83	42	0.88	17
4	0.87	97	0.79	28	0.68	25	0.83	6
5	0.76	62	0.90	39	0.84	44	0.78	9
6	0.90	70	0.93	44	0.94	18	0.76	25
Estimated probability	0.86		0.87		0.80		0.83	

F-ratios : Treatment effect (3,14 d.f.) - 1.07
Block effect (5,14 d.f.) - 1.01

* Excluded from analysis

(continued)

TABLE S21b

Observed proportions of 14-day old surviving chicks and estimated probabilities of surviving weeks 17-22

	Treatment							
	Control		Low		Intermediate		High	
	Prop.	No. hat.	Prop.	No. hat.	Prop.	No. hat.	Prop.	No. hat.
Block 1	1.00	16	0.95	20	1.00	2	*	*
2	0.95	21	1.00	30	1.00	1	1.00	9
3	0.89	27	0.95	19	0.95	40	1.00	20
4	1.00	27	1.00	2	1.00	4	*	*
5	0.83	29	1.00	11	1.00	10	*	*
6	0.97	29	1.00	26	1.00	11	1.00	27
Estimated probability	0.98		1.00		1.00		1.00	

F-ratios : Treatment effect (3,12 d.f.) - 5.46*
Block effect (5,12 d.f.) - 4.09*

* Excluded from analysis

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APPENDIX 12

(continued)

TABLE S22

Mean initial chick bodyweights (g) for treatments

Treatment group	Week 11	12	13	14	15	16	17	18	19	20	21	22
Control	37	36	37	36	36	35	38	36	38	36	42	41
Low	37	37	36	37	36	35	40	37	37	33	37	41
Intermediate	35	37	35	35	34	33	35	32	38	37	35	35
High	39	36	35	33*	35	37	38	37	40	38	38	34
Residual variance	24.6	40.1	37.2	48.1	37.5	45.9	12.0	40.4	14.1	4.10	-+	-+
Residual d.f.	12	11	14	13	11	10	10	9	5	5		
Treat[1]	1.85	0.67	1.77	2.61	0.69	1.22	6.62**	2.50	1.13	5.04		
Block[2]	0.23	0.60	0.97	0.59	0.17	0.08	2.31	0.04	1.12	4.93		

F-ratios : [1] Treatment effect (3, Residual d.f.)
[2] Block effect (5, Residual d.f.)

-+ Too few observations for analysis

APPENDIX 12

(continued)

TABLE S23

Mean chick bodyweights (g) at Day 14 for treatments

Treatment group	Week 11	12	13	14	15	16	17	18	19	20	21	22
Control	165	223	221	186	214	172	186	157	193	200	243	150
Low	196	210	235	203	225	186	190	164	186	192	270	229
Intermediate	217	188*	199	181	196	176	169	143	207	206	218	152
High	159	170*	220	157*	180*	185	195	157	228*	246	208	110
Residual variance	3462	2703	988	1272	1564	5965	1598	1707	469	1535	-+	-+
Residual d.f.	12	10	14	12	10	7	10	9	5	5		
Treat[1]	5.68**	4.63*	9.75***	8.07**	6.22*	0.21	1.55	0.90	6.62*	2.59		
Block[2]	0.30	2.78	6.62**	1.11	0.92	0.27	0.88	0.46	2.93	2.06		

F-ratios : [1] Treatment effect (3, Residual d.f.)
[2] Block effect (5, Residual d.f.)

-+ Too few observations for analysis