

INSECTICIDE RESIDUE LEVELS IN EGGS OF WILD PHEASANTS IN ILLINOIS

ROBERT E. GREENBERG and WILLIAM R. EDWARDS
Illinois Natural History Survey, Urbana

ABSTRACT. — Residues of both aldrin and DDT were found in 119 of 120 eggs from 21 clutches of eggs of wild pheasants (*Phasianus colchicus*) in east-central Illinois in 1966; heptachlor residues were detected in 61 of the 120 eggs. Averages and maximum residue levels observed were, for dieldrin, 0.30 ± 0.08 ppm and 2.82 ppm, and for p,p'-DDE, 0.15 ± 0.02 ppm and 0.61 ppm, as determined for wet weight of entire egg contents. The maximum level of heptachlor epoxide observed was 0.40 ppm. Although average values for individual pesticides appeared within the "safe" range, analysis revealed high variability in residue levels both among eggs and among clutches of eggs. These findings suggest a widespread contamination of the pheasants' environment in this area by persistent residues of chlorinated hydrocarbon insecticides.

Our purpose is to report levels of three common chlorinated hydrocarbon insecticide residues (dieldrin, p, p'-DDE, and heptachlor epoxide) in the eggs of wild pheasants, collected in 1966 in Champaign, Ford, Livingston, and McLean counties, east-central Illinois. The population sampled occurs on an intensively cultivated region dominated by cash-grain farming. In 1966 corn and soybeans occupied 46 and 27 percent, respectively, of the total land area of the counties in which the eggs were collected (Illinois Cooperative Crop Reporting Service 1967:6-8).

From 1964 through 1966, about half of the corn acreage in Illinois was treated with aldrin, most com-

monly in granular form. Aldrin was typically either broadcast on and disked into the soil or applied directly in the row at planting, at a rate of 1 to 1½ lb technical material per acre, to control injurious insects in the larval stage (H. B. Petty, Illinois Natural History Survey, Urbana, unpublished data). The period of aldrin application (May) coincides with the seasonal peak of pheasant egg-laying. Once ingested, aldrin is readily converted into its epoxide, dieldrin, by metabolic activity (Bann et al. 1956:939).

In Illinois, the present trend is toward decreasing use of aldrin in the production of corn. The amounts applied in 1967 and 1968 were, respectively, about 5 percent and 25 percent less than in 1966 (H. B. Petty, Illinois Natural History Survey, Urbana, unpublished data). Aldrin is being replaced by the organo-phosphates because of the rapid spread of strains of corn rootworms (*Diabrotica* spp.) and a complex of other root-feeding insects resistant to aldrin. Nevertheless, we feel it is important to make this report because (1) large quantities of aldrin have been applied to pheasant habitat over much of the pheasant range, and this material will persist in the environment in its epoxide form, dieldrin, for many years, and (2) we anticipate

continued extensive use of aldrin on the basis of its past record and the promotional activities of its manufacturer.

The use of heptachlor paralleled that of aldrin, but heptachlor made up only about 20 percent of the total quantity of chlorinated hydrocarbon insecticides used in Illinois agriculture during the middle 1960's (Stevenson Moore III, Illinois Natural History Survey, Urbana, unpublished data).

The use of DDT in Illinois agriculture was widespread from the late 1940's through the mid 1950's, reaching a peak in 1956 and declining rapidly thereafter as DDT was phased out in favor of other chemicals. DDT has not been used extensively in Illinois agriculture since the late 1950's (Stevenson Moore III, Illinois Natural History Survey, Urbana, unpublished data).

METHODS

Pheasant eggs were collected during June, 1966, from hayfields at the time of mowing. One hundred twenty-two eggs, representing 21 individual clutches, were obtained. The eggs were labeled, placed individually in polyethylene bags, and frozen until they could be analyzed. All eggs were analyzed individually except for 1 clutch of 2 eggs that was pooled.

Although chlorinated hydrocarbon insecticide residues are concentrated in the lipids of the yolk (Lamb et al. 1967:25), our determinations of residue levels were based on the wet weights of the entire contents of whole eggs after their shells and the associated membranes were removed.

This method was followed because the incubation status of the eggs was variable and we had no knowledge of any possible effect of incubation on residues or moisture in the remaining yolk. Our method differs from that of several workers (Baxter et al. 1969:100, as described by R. L. Linder, personal communication, 3/14/69; Atkins and Linder 1967:747; and Lamb et al. 1967:25), who presented data based on the wet weight of the yolk alone, but is the same as that of Labisky and Lutz (1967:16).

To facilitate comparison of data obtained during this study with data of Lamb et al. (1967), Atkins and Linder (1967:747), and Baxter et al. (1969:100), their data were converted by multiplying with a factor of 0.406. This factor is based on the average composition, by weight, of eggs of ring-necked pheasants, as reported by Romanoff and Romanoff (1949:114).

Individual eggs were prepared for chromatographic analysis by saponification in alcoholic potassium hydroxide and by acetonitrile partitioning (Burchfield et al. 1965). Alkaline digestion does not affect aldrin and dieldrin. DDT is converted to DDE; heptachlor epoxide is stable if not heated (W. N. Bruce, Illinois Natural History Survey, Urbana, personal communication, 8/13/69). Each specimen was analyzed by gas chromatography (Beckman model GC-4), utilizing an electron capture detector. Approximately 12 different stainless steel columns, 3 to 4 feet long, with an inside diameter of 1/16 inch, packed with 1 percent Shell EPON resin 1001 and 0.5 percent DuPont Viton A fluoroelastomer

on a solid support of Varian Aero-graph Chromosorb W, AW-DMCS, 100/120 mesh, were used to make these analyses. Gas flow rates were 40 cc/minute for the carrier helium, 75 cc/minute for the discharge helium, and 2 to 5 cc/minute for the carbon dioxide. The inlet line was operated at 210 C, the column at 180 (to 220) C, the detector line at 237 C, and the detector at 247 C. Retention times with the column at 180 C were felt to be excessive, and were often reduced (at the expense of column life) by increasing the column temperature to 220 C.

Relative retention times of aldrin, heptachlor epoxide, p,p'-DDE, and dieldrin were, respectively, 2.0, 6.1, 8.8, and 11.5, permitting proper identification and quantification of each of these insecticides. Because wide variations occurred in the lower limits of detection of heptachlor epoxide, no attempt was made to estimate the mean value, and incidence must be considered a minimum figure.

As a basis for estimating residues, standards were analyzed with every specimen. Measurements were double- or triple-replicated for each sample; residues were estimated to the nearest 0.01 ppm (micrograms of insecticide per gram of egg contents, wet weight). In addition, several interlaboratory check samples were analyzed and confirmed by Dr. W. N. Bruce and his associates in the Section of Economic Entomology, Illinois Natural History Survey, Urbana. Determinations were not verified by paper or thin-layer chromatography.

Percent recovery tests indicated a range of 80 to 110 percent. Data

presented in this report do not include a correction factor for percent recovery.

RESULTS

Residues of both aldrin/dieldrin and DDT were detected in 119 of 120 eggs analyzed individually and in each of 20 clutches (Tables 1 and 2). In addition, residues of heptachlor were observed in 61 of the 120 eggs and in 15 of 20 clutches (Table 3). These incidence figures represent a minimum accounting because the incidence of heptachlor residues at levels below 0.1 ppm is not adequately represented and because no attempt was made to account for lindane, chlordane, endrin, and for metabolites and isomers of DDT other than p,p'-DDE.

Mean (with 95 percent confidence limits) residue levels observed per egg (120 eggs) were, for dieldrin, 0.30 ± 0.08 ppm, and for p,p'-DDE, 0.15 ± 0.02 ppm, as determined for wet weight of entire egg contents.

Considerable variation in residue levels was evident within individual clutches as well as among clutches of the entire sample of 21 clutches (Tables 1-5). Although our sample was small and contained few complete clutches, the data show, as one might expect, that variability was less within individual clutches than it was over the entire sample (Table 4). In general, for clutches having high average residue levels, all eggs tended to be relatively high, and vice versa. Analysis of variance verified that significant differences ($F_{3,46} = 10.19$) existed in dieldrin levels among the four clutches compared.

TABLE 1.—Levels of dieldrin (ppm wet weight) in contents of eggs of wild pheasants, collected in east-central Illinois in June, 1966.

Clutch Reference Number												
	45A	45B	94	57	100	101	105	123	124	125	126	127
County	Liv.	Liv.	Ford	Liv.	Ford	Cham.	Cham.	McL.	Cham.	McL.	Ford	Ford
No. eggs in clutch.	NK ¹	NK	NK	11	8	17	NK	8	NK	13	11	NK
No. eggs analyzed.	1	3	2	2	1	17	3	2	8	11	11	9
Incubation (days).	0 ¹	14	NK	10	8	20	18	16	19	7	21	15
Mean dieldrin.	0.14	0.44	0.03	2.33	0.04	0.31	0.42	0.14	1.55	0.47	0.05	0.26
Dieldrin in eggs.	0.14 ²	0.52 0.50 0.30	0.03 0.03	Pooled	0.04	0.26 0.24 0.26 0.33 ⁸ 0.38 ⁸ 0.43 0.63 ⁴ 0.25 0.24 0.40 ⁹ 0.21 0.38 0.39 0.42 0.29 0.23 0.14 0.42 ⁹	0.66 0.04 ⁹ 0.57 ⁹	0.19 0.10	1.92 1.75 1.54 2.82 2.20 0.63 ⁴ 1.04 0.49	0.23 ⁷ 0.42 0.24 0.31 0.33 1.53 0.34 0.20 0.35 0.15 ⁷ 1.12	0.03 ³ 0.06 0.05 0.03 0.01 0.07 0.04 0.02 0.02 0.13 0.06	0.15 0.66 0.22 0.26 0.19 0.22 ⁶ 0.35 ⁵ 0.19 ⁶ 0.13

TABLE 1.—(Cont.)

	Clutch Reference Number								
	128	130	134	136	137	153	154	155	158
County.....	Liv.	McL.	McL.	Cham.	Cham.	Liv.	Liv.	Ford	Liv.
No. eggs in clutch.....	11	5 ¹	NK	NK	NK	NK	NK ¹	NK ¹	12
No. eggs analyzed.....	7	5	6	9	5	6	2	1	11
Incubation (days).....	18	0 ¹	8	NK	NK	NK	0 ¹	0 ¹	12
Mean dieldrin.....	0.27	0.04	0.15	0.16	0.13	0.22	0.06	0.02	0.08
Dieldrin in eggs.....	0.17	0.02 ²	0.17	0.07	0.16	0.12	0.07	0.02 ²	0.09
	0.13	0.02	0.16	0.07	0.12	0.12	0.05		0.10
	0.32	0.11	0.11	0.16	0.04	0.68			0.09
	0.16	ND	0.21	0.07	0.16	0.06			0.13
	0.53	0.04	0.16	0.11	0.19	0.18			0.06
	0.52		0.10	0.67		0.15			0.06
	0.09			0.09					0.08
				0.16					0.09
				0.06					0.10
									0.06
									0.06

NK = not known.

ND = not detected (less than 0.004 ppm).

1. Clutch incomplete; hen still laying. 2. Egg in oviduct. 3. Hatching. 4. Embryo dead. 5. Embryo dead and decomposed. 6. Embryo dead at 1 day. 7. Embryo dead at 2 days. 8. Embryo dead at 8 days. 9. Embryo at 14 days.

TABLE 2.—(Cont.)

	Clutch Reference Number								
	128	130	134	136	137	153	154	155	158
County.....	Liv.	McL.	McL.	Cham.	Cham.	Liv.	Liv.	Ford	Liv.
Mean p,p'-DDE.....	0.12	0.04	0.11	0.12	0.09	0.07	0.06	0.22	0.10
Individual eggs.....	0.06 0.05 0.10 0.07 0.21 0.26 0.09	0.02 ² 0.02 0.11 0.01 0.04	0.03 0.22 0.11 0.12 0.11 0.09	0.04 0.07 0.16 0.04 0.04 0.50 0.08 0.12 0.07	0.08 0.12 0.04 0.12 0.08	0.02 0.12 0.15 0.02 0.10 0.03	0.06 0.06 0.05	0.22 ²	0.17 0.14 0.08 0.13 0.09 0.06 0.13 0.05 0.10 0.08 0.09

Trace = detected at level below 0.01 ppm.
 ND = not detected (less than 0.08 ppm).
 See Table 1 for explanation of superscripts.

TABLE 3.—(Cont.)

Clutch Reference Number									
	128	130	134	136	137	153	154	155	158
County.....	Liv.	McL.	McL.	Cham.	Cham.	Liv.	Liv.	Ford	Liv.
Mean heptachlor epoxide.....	0.10	Trace	ND	0.04	Trace	Trace	Trace	ND	0.03
Individual eggs.....	0.06 0.03 0.13 0.07 0.12 0.26 0.04	ND ² ND 0.01 ND ND ND	ND ND ND ND ND ND	0.04 0.04 0.02 0.08 0.04 ND 0.05 0.01 0.04	ND ND 0.01 ND ND	Trace Trace ND Trace Trace 0.01	Trace Trace Trace	ND ²	0.03 0.01 0.01 0.01 0.04 ND 0.01 0.01 0.10 0.07 ND

ND = not detected (less than 0.5 ppm).
 Trace = detected at level below 0.01 ppm.
 See Table 1 for explanation of superscripts.

TABLE 4.—Variation in dieldrin residues of clutches of eggs of wild pheasants from east-central Illinois, 1966.

	Clutch Reference Number				Sample of 20 Clutches
	126	158	101	125	
No. eggs in clutch.....	11	12	17	13	Not applicable 120
No. eggs examined.....	11	11	17	11	
Range (ppm).....	0.01-0.13	0.06-0.13	0.14-0.42	0.15-1.53	0.0-2.82
Mean (ppm).....	0.05	0.08	0.31	0.47	0.30
Standard deviation.....	0.03	0.09	0.09	0.44	0.44
Coefficient of variation.....	0.60	1.13	0.29	0.94	1.46

TABLE 5.—Variation in p,p'-DDE residues of clutches of eggs of wild pheasants from east-central Illinois, 1966.

	Clutch Reference Number				Sample of 20 Clutches
	126	158	101	125	
No. eggs in clutch.....	11	12	17	13	Not applicable 50
No. eggs examined.....	11	11	17	11	
Range (ppm).....	0.03-0.22	0.05-0.17	ND-0.15	0.27-0.61	ND-0.61
Mean (ppm).....	0.09	0.10	0.08	0.41	0.15
Standard deviation.....	0.05	0.04	0.03	0.09	0.13
Coefficient of variation.....	0.57	0.37	0.38	0.22	0.90

ND=not detected (less than 0.08 ppm).

DISCUSSION

It appears that in 1966 virtually every pheasant egg laid in the area of our collections contained residues of at least aldrin/dieldrin and DDT. In addition, about half the eggs apparently contained heptachlor residues. Considering the history of widespread use of insecticides in Illinois agriculture (H. B. Petty and Stevenson Moore III, Illinois Natural History Survey, Urbana, unpublished data), this condition undoubtedly extended well beyond the area sampled.

These data provide a limited picture of the occurrence of three chlorinated hydrocarbon residues in the eggs of wild pheasants, but they do not define the role of such insecticides as a factor in the ecology of that population. We have no knowledge of what residue levels in the ranges observed implies in terms of physiological status of the wild hens that laid them or in the chicks they might produce. If residue levels do reflect a problem or a potential problem, as we suspect, we do not yet know its parameters or the

trends in its magnitude. The incidences of aldrin/dieldrin, DDT, and heptachlor residues, however, leave little doubt that the pheasants' environment in east-central Illinois in 1966 had been extensively contaminated by these persistent insecticides.

The average levels of contamination for our sample were approximately 10 times the mean level of 0.03 ppm (after conversion) for dieldrin and 15 times the mean level of 0.01 ppm (after conversion) for DDE found in a sample of wild pheasant eggs from southeastern South Dakota in 1967 (Baxter et al. 1969:100). Although no error terms which would facilitate comparisons were given for the South Dakota samples, it appears that the eggs in our sample contained more dieldrin and DDE than did the eggs of wild pheasants from South Dakota in 1967.

The highest residue levels observed in eggs in our Illinois sample were 2.82 ppm dieldrin, 0.61 ppm DDE, and 0.40 ppm heptachlor epoxide. These levels were, respectively, about 5, 20, and 4 times the maximum levels observed by Baxter et al. (1969:100) in eggs from the South Dakota sample (0.59 ppm dieldrin, 0.03 ppm DDE, and 0.10 ppm heptachlor epoxide, after conversion).

The dieldrin levels we observed were lower than those reported by Labisky and Lutz (1967:16) who found 4.60, 4.96, and 7.91 ppm dieldrin in three samples each comprised of two eggs from nests of wild pheasants in an area near Donovan, Illinois, that was subjected to an aerial, solid block application of aldrin at a rate of 2 lb technical material per acre in April, 1960, to contain an

infestation of Japanese beetles (*Popillia japonica*). It was their conclusion (1967:23) that, after treatment of the area, pheasants suffered "a substantial mortality from acute poisoning and a lowered reproductive efficiency from chronic poisoning." The high concentrations of dieldrin that they observed, in comparison with our findings, are believed to reflect the more extensive nature of the treatment, the lack of immediate incorporation of the toxicant into the soil, the timing of the application (April rather than May), or some combination of these factors.

Responses reported for caged pheasants fed standardized dosages of dieldrin offer a clue to the possible responses of wild pheasants to that poison. However, interpretation must be undertaken with caution, as other factors may have influenced the respective responses. Atkins and Linder (1967:752) reported that average levels of dieldrin residues in the eggs of pheasants fed 2, 4, and 6 mg of dieldrin per week for 12 weeks were (after our conversion of their data): 3.2, 7.5, and 13.6 ppm, respectively. These averages were considerably higher than those detected in our sample but, except for the 6 mg group, they were in the same general range as those reported by Labisky and Lutz (1967:16). Atkins and Linder (1967:752) concluded that parameters of reproduction were not adversely affected in hens in the 2 and 4 mg treatment classes but that condition (body weight) was lowered and egg production reduced in the 6 mg class. The highest level of dieldrin residues observed in any egg of our sample was also lower than the mean content

of dieldrin in eggs from hens in the 2 mg class.

CONCLUSION

Thus, at first glance, it appears that the average level of dieldrin residues in our sample was within the "safe" range. But average values for individual toxicants are poor yardsticks because they fail to recognize the degree of variation from the norm expressed by individuals. In our sample, the more contaminated eggs contained 5 to 10 times the average level of dieldrin. Consideration of average levels of individual insecticides tends to obscure the total degree of contamination and the fact that several toxicants in combination can be more lethal than one alone (Chemical and Engineering News 1966:19). Also of potential significance is the finding of Baxter et al. (1969:96) that reproductive parameters were significantly reduced in groups of penned pheasants when second-generation hens had received dieldrin via the egg.

It appears that, in Illinois, the use of the long-persistent chlorinated hydrocarbons, particularly aldrin, DDT, and heptachlor will continue to be reduced in favor of more toxic but less stable compounds. We are hopeful that the long-term effect of this change will be potentially less severe on wildlife than the possible effects of continued application of high levels of the chlorinated hydrocarbons.

ACKNOWLEDGMENTS

Acknowledgment is extended the following personnel of the Illinois Natural

History Survey: to Dr. Glen C. Sanderson for administrative and technical supervision; to Dr. Richard R. Graber and David A. Casteel for collecting the eggs; to Ronald E. Duzan and Mary Ann Johnson for analyzing the eggs; and to Dr. Ronald F. Labisky, William L. Anderson, Stanley L. Etter, Dr. Glen C. Sanderson, and Helen C. Schultz for technical and editorial suggestions.

LITERATURE CITED

- ATKINS, T. D., and R. L. LINDER. 1967. Effects of dieldrin on reproduction of penned hen pheasants. *J. Wildl. Mgmt.* 31(4):746-753.
- BANN, J. M., T. J. DECINO, N. W. EARLE, and Y.-P. SUN. 1956. The fate of aldrin and dieldrin in the animal body. *J. Agr. and Food Chem.* 4(11):937-941.
- BAXTER, W. L., R. L. LINDER, and R. B. DAHLGREN. 1969. Dieldrin effects in two generations of penned hen pheasants. *J. Wildl. Mgmt.* 33(1):96-102.
- BURCHFIELD, H. P., D. E. JOHNSON, and ELEANOR E. STORRS. 1965. Guide to the analysis of pesticide residues. Vol. 1. U.S. Dept. Health, Ed. and Welfare, Public Health Service, Office of Pesticides, Washington, D.C. Sections II D. 1. a. (1 and 2) and II B. 1. a. (1 and 2).
- Chemical and Engineering News. 1966. Combinations raise insecticide toxicity. *Chem. and Eng. News* 44(5):28.
- Illinois Cooperative Crop Reporting Service. 1967. Illinois agricultural statistics. Assessors' annual farm census 1967. Illinois Cooperative Crop Reporting Service, Springfield. S.F.C.-30. 29 pp.
- LABISKY, R. F., and R. W. LUTZ. 1967. Responses of wild pheasants to solid-block applications of aldrin. *J. Wildl. Mgmt.* 31(1):13-24.
- LAMB, D. W., R. L. LINDER, and YVONNE A. GREICHUS. 1967. Dieldrin residues in eggs and fat of penned pheasant hens. *J. Wildl. Mgmt.* 31(1):24-27.
- ROMANOFF, A. L., and ANASTASIA J. ROMANOFF. 1949. *The avian egg*. John Wiley & Sons, Inc., New York. 918 pp.

Manuscript received November 24, 1969.