Effect of External Application of Pesticides to the Fertile Egg on Hatching Success and Early Chick Performance 2. Commercial-Herbicide Mixtures of 2,4-D with Picloram or 2,4,5-T Using the Pheasant.

by

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LUTZ and LUTZ-OSTERTAG (1970) encountered serious consequences on game bird eggs by spraying with 2,4-D at recommended field rates. Not only was there extensive embryonic mortality but terata and anomalies of the survivors were noted. In contrast, DUNACHIE and FLETCHER (1967, 1970) did not witness the same degree of mortality nor abnormalities when larger quantities of herbicide were injected directly into the hen's egg. Responding to the results of both groups, SOMERS et al. (1973) investigated the effect of spraying the egg of the hen. Contrary to expectation, contamination with 2,4-D in combination with picloram or 2,4-5-T prior to incubation failed to cause any adverse happenings on both hatching success and early live performance.

One of the more serious inadequacies of using the hen in conveying possible ecological effects to the aves group as a whole lie in its extensive controlled genetic background and superior reproductive performance. Thus, the egg as derived from the domestic chicken may not accurately reflect the consequences on other species deficient in these qualities. The present study should provide an indication as to the significance of specie differences in response to herbicide contamination. The pheasant provided the reproductive unit while all procedures used paralleled those of the aforementioned hen investigation from this laboratory.

Material and Methods

Ring-neck Pheasant eggs (Phasianus colchius sp.) were obtained from a local game farm where natural encounter was the method of fertilization. Egg age was eliminated as a confounding factor through equal distribution across treatments. Both procedure and equipment involved in egg storage and incubation as well as herbicide application and analysis were identical to that employed in the hen experiment of SOMERS et al. (1973). Embryonic development of the pheasant requires 24 days as opposed to 21 for Gallus domesticus; thus, egg transfer from incubator to hatcher presented the only operational difference between studies (19 vs 21 days).

Also identical to that employed in the hen study were the herbicide mixtures, source, production lots, and concentrations sprayed. The 2,4-D:picloram (4:1) and 2,4-D:2,4,5-T (1:1) were used at the suggested rate for the former (2.8 kg/ha) and 10x field recommendations for the latter (11.2 kg/ha). Control eggs were sprayed with water at the volume rate of 746 l/ha. After hatching, random samples of chicks were placed in electrically heated brooder batteries. All birds received the same 28% protein starting ration ad libitum. Inaccuracy of the vent technique in sex determination at hatch necessitated a 4 week delay.

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Results and Discussion

The hatching success of herbicide treated pheasant eggs when compared to controls indicated no adverse effects due to treatment (Table 1). On the contrary, both herbicides resulted in lower proportions of early and late dead germs with ultimately a larger hatch. These data are not statistically significant when each parameter in incubation is taken separately, however, a certain amount of credability for an improvement with herbicide application evolves with the composite of results.

TABLE 1

Incubation Performance of Pheasant Eggs Sprayed with Commercial Herbicide Formulations (% of Viable Germs)[a]

Incubation[b] Parameter	Water Control	2,4-D: Picloram	2,4-D: 2,4,5-T
EDG	16.6	11.3	9.7
LDG	11.6	7.9	6.4
Pip	11.1	13.8	14.9
Hatch	60.8	67.0	68.6

[a] 235 Eggs set/treatment.
[b] Early Dead Germ(1-5 days), Late Dead Germ(post 6 days), Pip or mortality upon breaking shell.

The total hatch for this experiment is lower than would ordinarily be encountered for pheasants (ca. 70-75%). This reduction may be attributed to parent stock which were nearing the end of the breeding season and producing eggs with less viable germs. A gross examination of all late dead germs and pips indicated no differences in either the incidence or type of malformations that could be attributed to treatment. Again herbicide contamination would appear to be the more advantageous circumstance (Table 2).

Growing random samples of chicks that had hatched from each of the treatments to 4 weeks of age elicited a sex effect which was not apparent from their weight upon hatching (Table 3). Males had an improved rate of growth if they were from eggs sprayed with herbicide ($P < 0.078$) while there was no ready response by the female ($P < 0.614$). The sex-combined data on feed utilization and mortality did not support any effects due to treatment.

TABLE 2 Incidence of Pheasant Embryo Malformations from Eggs Sprayed with Herbicides (LDG + Pip)

Treatment	% Malform	Abnormality Distrib., No.				
		Toe	Beak	Leg	Eye	Feather
Water Control	51.1	19	2	0	1	2
2,4-D:Picloram	40.9	16	0	3	1	0
2,4-D:2,4,5-T	37.8	14	1	0	2	0

TABLE 3 Live Performance of Pheasant Chicks Hatched from Herbicide Treated Eggs (0-4 weeks of age)[a]

Treatment	Wt. gain, g[b]		Sex-combined	
	♂	♀	F/G[c]	% Mort.
Water Control	256	217	1.66	6.4
2,4-D: Picloram	263	216	1.63	8.3
2,4-D: 2,4,5-T	280	214	1.65	4.0
$S\bar{x}$ (df)	28(48)	22(39)	-	-

[a] 4 replicate pens of 10 mixed sex chicks/pen/treatment.
[b] Average starting weight was 23 g for all treatments.
[c] Feed consumed/weight gain.

TABLE 4 Residues Resulting from Pre-Incubation Spraying Pheasant Eggs with 2,4-D: 2,4,5-T (ppb wet wt.)[a]

Tissue	Treatment	2,4-D	2,4,5-T	Total
Shell	-	-	-	-
	+	45.21	48.19	93.41
EDG	-	T	T	T
	+	2.21 ± 0.06	1.43 ± 0.15	3.64 ± 0.21
LDG	-	0.01	0.01	0.01
	+	0.15 ± 0.01	0.29 ± 0.03	0.44 ± 0.04
Day old Chick	-	T	T	T
	+	0.12 ± 0.02	0.15 ± 0.03	0.27 ± 0.04

[a] Duplicate analysis have an associated standard deviation, single analysis do not. The % recovery of 2,4-D and 2,4,5-T was 55 and 65, respectively, as based on the extraction procedure of YIP (1971) and electon capture GLC using 6% Carbowax 20 M on 60-80 Mesh Varoport 30.

Residue analysis was performed only with that treatment involving 2,4-D: 2,4,5-T (Table 4). These herbicides were detected in substantial quantity with both the EDG and LDG confirming their penetration through the shell. The recovery of both acids in but trace amounts from the water-sprayed control eggs reduces the likelihood that cross-contamination by volatiles during incubation effected equalization of treatment results.

The present experiment involving eggs from the pheasant completely parallels the results of SOMERS et al. (1973) using the domestic chicken. No adverse effects of herbicide contamination were forthcoming during incubation while an improved post-hatch weight gain was detected only with males. In both species these phenoxyacetic acids entered the egg and decreased in concentration with embryonic age. Because near equal concentrations of 2,4-D and 2,4,5-T were recovered at each stage, it would appear that these avian systems cannot readily distinguish between them. It does not seem likely that the amounts detected in the dead germs were lethal as somewhat comparable quantities of these compounds were also found in chicks that successfully hatched.

The only measurement which indicated that the pheasant had a greater sensitivity than the hen was with regard to EDG residue concentration. Under terms of near equal shell contamination, the EDG of the pheasant was found to have 4 times the amount detected with the hen's egg. This difference was reduced by half at subsequent LDG and hatching stages. Thus, for all intentions of the present study, no outstanding differential effects exist between the fertile egg of the pheasant and hen in its response to the presently used prominent herbicides.

Summary

Aqueous solutions of 2,4-D:picloram and 2,4-D:2,4,5-T equivalent to recommended and 10x field concentrations, respectively, were sprayed on fertile pheasant eggs preceeding incubation. No treatments were found to cause any adverse effect on hatching success, incidence of malformed embryos or subsequent chick mortality relative to water-sprayed control eggs. Herbicide contamination was found to facilitate weight gain of males from 0 to 4 weeks of age while females failed to elicit a response. Residue analysis verified herbicide deposition on the shell and entry into the egg. These results completely paralleled those of an earlier study with the domestic chicken.

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