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Indicator Bird Species for Toxicity Determinations: Is the Technique Usable in Test Method Development?

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ABSTRACT: Pesticide, food, drug, and cosmetic industries have used indicator animal species for many years to provide data on the relative hazards of synthetic or natural chemical products to applicators or users. A considerable amount of toxicological information is available on the relative susceptibility of different mammal species to various chemicals. This information has been used to predict hazards to other mammals, including man. Indicator species often are used for evaluating the efficacy of chemicals or their hazards to wild birds. However, the available toxicological information is generally not sufficient to extrapolate valid relationships between the indicator species and the target species.

In this paper we have presented the state of the art for the use of indicator bird species and have expressed our opinion on the use of these data. Because many current uses for bird indicator species are not well supported by available data, we recommend that further test method development with avian indicator species be terminated until additional data can be gathered on the relevancy of the technique.

KEY WORDS: vertebrate pest control, acute toxicity, birds, chemicals, hazard evaluation, pesticides, review, subacute toxicity, test methods, toxicology.

With the increasing concern about the effects of chemicals in the environment, scientists from many disciplines are working to develop test methods that can predict hazardous effects of these materials. The increasing use of pesticides, particularly during the last three decades, has resulted in a specific need to find accurate methods for predicting hazards to humans, domestic animals, fish, wildlife, and the environment in general. Although decades of experience in the development of human and veterinary drugs have provided a rational, if tenuous, basis for the extrapolation of acute and subacute toxicological data from one

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mammalian species to another [*1*],[3] there is a relative paucity of such data or experience for the extrapolation of data from one bird species to another.

The indicator species concept for predicting the hazards of pesticides to wild birds is being widely practiced by pesticide developers and promoted by governmental regulatory agencies. Current test methods probably do not yield relevant data, and new test methods based on the same assumptions may be equally irrelevant. We will show, however, that through the evaluation of specific data, there is some reason to believe that the indicator species concept may be used in the future. We will limit our discussion to the use of indicator bird species for predicting acute or subacute hazards to wild birds from pesticides. We will identify the state of the art with respect to the use of indicator bird species, and we will show that most of the currently used test methods are not well supported by existing data and thus lead to erroneous conclusions about hazards, real or imagined.

Wild Bird Populations in North America

About 650 species of wild birds, representing about 75 families, live or breed or both on the North American continent (excluding Mexico and Central America). An additional 100 to 150 species are casual or accidental visitors. The maximum postbreeding population of these species has been estimated at 20 billion by Robbins, Brun, and Zim [*2*]. Accurate population levels of only a very few, if any, species are known, but it is possible to estimate approximate levels by using various sources [*3,4*]. We have estimated that 200 million waterfowl (ducks and geese) and 800 million upland game birds (pheasant, quail, etc.) exist at present population levels. These game bird species, representing five separate families, probably constitute no more than 5 percent of the total avifauna of North America. The 27 families of perching birds (order Passeriformes) may constitute more than 50 percent of the total North American postbreeding population. Within this order, the most abundant families are the Sturnidae (starlings), Icteridae (blackbirds and orioles), Ploceidae (old world sparrows), and Fringillidae (new world sparrows, grosbeaks, finches, and longspurs). Icterids themselves may contribute as many as one to three billion birds to the postbreeding populations, or about 15 percent of the estimated total North American bird population [*4,5*].

Historical and Current Use of Indicator Species

The domestic chicken (*Gallus domesticus*) was probably the first avian species routinely used as a test animal in toxicological studies for pesticides because of its availability and the knowledge of its husbandry requirements. It was used prevalently in the 1930's and 1940's when rudimentary LD_{50} tests were performed. In the late 1940's and early 1950's, bobwhite quail (*Colinus virginianus*) and ring-

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[3] The italic numbers in brackets refer to the list of references appended to this paper.

necked pheasants (*Phasianus colchicus*) replaced the chicken as primary test species, placing the emphasis in toxicological determinations closer to the wild game bird species. In the 1960's, coturnix quail (*Coturnix coturnix*), bobwhites, pheasants, and mallards (*Anas platyrhynchos*) were commonly used in tests, culminating in the present U.S. Environmental Protection Agency (EPA) regulations which require bobwhite and mallard data for all pesticides that are to be used outdoors (Federal Insecticide, Fungicide, and Rodenticide Act, as amended, Section 162.82). Mallards and bobwhites were selected for these tests, probably because they can be readily bred and maintained in laboratory conditions, are rather uniform and stable test animals, and represent two of the primary North American game bird species. The literature, however, contains little information on the relationship between the toxicological sensitivity of bobwhites and mallards and that of other bird species.

Toxicological Relationships Between Bird Species

Few published studies exist that can be used to indicate subacute toxicological relationships among wild bird species. Two papers [*6,7*] discuss these relationships between four species of young birds (coturnix quail, bobwhites, ring-necked pheasants, and mallards), representing two orders, Anseriformes and Galliformes. Toxicological sensitivity was correlated with the body size of the bird and the basic chemical structure of the pesticide, but not with the species. However, the authors of the two papers just cited note that "... inconsistencies in the relative sensitivity of the four species ... suggests that other species would be much more sensitive to different chemicals." In a third study [*8*], a similar procedure was used to test four pesticides on four bird species representing the orders Galliformes and Passeriformes. In addition to showing that the Passeriformes [blue jay (*Cyanocitta cristata*), house sparrow (*Passer domesticus*), and cardinal (*Richmondena cardinalis*)] were considerably more sensitive to the four pesticides than were bobwhites, the author (Hill, Ref *8*) indicated that increased sensitivity was also a function of decreasing body weight. This latter factor is particulary critical in subacute toxicity feeding studies because avian energy requirements and, hence, food consumption increase with decreasing body weight [*9*]. Although avian sensitivity to individual chemicals varied considerably, according to chemical structure and body weight, it also appeared that there was a difference in toxicological sensitivity between at least two of the three orders tested (Passeriformes had a much greater sensitivity than Galliformes).

Several published reports deal with acute avian toxicological sensitivity to pesticides. Tucker and Haegele [*10*] summarized many of their laboratory studies [*11,12*] designed to determine whether the indicator species concept was valid, and, if so, which species were most sensitive. They presented data on 16 pesticides on one Anseriforme, three Galliformes, one Columbiforme, and one Passeriforme. By using a ranking procedure, they showed that no significant toxicological sensitivity differences existed among the species tested [two species, chukar partridges

(*Alectoris graeca*) and house sparrows were at or near the $P = 0.05$ confidence limits], but there was a large variation among species in their susceptibility to a given compound. They concluded, "...our study indicates that the coturnix is no better as a representative species than any other..." These authors also pointed out that indicator species are often selected because of financial constraints, but that "...the species itself should be used in toxicity determinations to avoid the need for extrapolation," a valid but often impractical suggestion when one considers the 650 species of birds normally present in North America.

In four papers previously written by the authors of this study published here, we attempted to show that one can generalize about the acute toxicological sensitivity of pesticides and soporifics to various orders, families, and species. In 1967 we presented data suggesting that red-winged blackbirds (*Agelaius phoeniceus*) were more sensitive to carbamate soporifics than were starlings (*Sturnus vulgaris*) [*13*]. In 1972, we expanded these data by including one Anseriforme, one Galliforme, one Columbiforme, and two additional Passeriformes, and also included other classes of chemicals. This study, when analyzed by the sum of ranks procedure [*14,15*], indicated that on the average, the ring-necked pheasant was significantly ($P \leq 0.05$) less sensitive and the house finch (*Carpodacus mexicanus*) and red-winged blackbird significantly more sensitive ($P \leq 0.05$) to the chemicals tested than were the other species [*16*]. These data support the findings from subacute studies that Passeriformes may be significantly more sensitive to chemical intoxication than some Galliformes.

In 1972 we also presented data [*17*] on the acute toxicity of 369 pesticidal compounds to starlings, red-winged blackbirds, and, where available, rats (*Rattus norvegicus*). These data showed that rats were, on the average, less sensitive ($P \leq 0.10$) to intoxication than the two Passeriformes tested. Testing a large number of compounds, we also showed a difference in toxicity within an order (redwings were more sensitive than starlings). In 1973, we published an additional paper [*18*] that reinforced the idea that it was indeed possible to separate species sensitivity to intoxication within an order by appropriate statistical methods.

The following information provides additional data on using the indicator bird species concept in predicting toxicological sensitivity to chemicals and in preparing test methods.

Methods and Results

The acute oral toxicity of 36 pesticides was determined following the ASTM Recommended Practice for Determining Acute Oral LD_{50} for Testing Vertebrate Control Agents (E555-75) developed by the American Society of Testing and Materials and the method described by Schafer et al [*18*] on each of six species of birds including, in phylogenetic order: the coturnix quail, common pigeon or rock dove (*Columba livia*), starling, common grackle (*Quiscalus quiscula*), red-winged blackbird, and house sparrow.

All the test birds were captured from wild populations in Colorado except the coturnix quail, which were raised in our breeding facilities (descendants of Random Line 926, University of California, Davis). Mixed sexes were used in determining the LD_{50}'s, except as noted. The birds were acclimated to laboratory conditions for at least 14 days before being tested, to stabilize their diet and general condition. The pesticides used were generally of technical grade; the purity and chemical descriptions are listed in Table 1. To determine whether there was a difference in toxicological response by species (Table 2), the authors arbitrarily ranked each compound for its toxicity to all six species, using a scale of 1 to 6 (1 = most toxic; 6 = least toxic; Table 3). To detect any statistical differences in these ranked data, we used the two-way analysis of variance test described by Friedman [*14*]. If there was a significant difference in species response to the tested pesticides with the six species, a further analysis of the data by the "nonparametric multiple comparisons by STP" procedure [*19*] was scheduled.

Initial statistical analysis of the data suggested that there was a difference in the toxicological response of the six species to the 36 chemicals tested ($P < 0.0005$) and that the sensitivity of the individual species could be statistically separated and ranked (Table 4). These data tend to confirm our earlier reports that definite sensitivity relationships can be established among some species of birds, provided that a large number of chemicals is used and that the species are diverse in phylogenetic composition.

Discussion

The preceding review and data suggest that it is possible to rank orders of birds according to their average sensitivity to pesticides, and perhaps also to rank them in their sensitivity to specific groups of pesticides. Within orders and families, species may also be ranked in their sensitivity to pesticides, provided that large samples are used in the evaluations. However, the relationships can be only roughly defined at this time. Sufficient data already exist to indicate that the presently used indicator species (mallards and bobwhites) are poor indicators of acute and subacute chemical avian intoxication since they do not represent the toxicological sensitivities of the majority of the North American bird species. Consequently, the basis for the present use of indicator bird species for estimating or predicting hazards to other species of wild birds is not well established. We believe that a more valid basis for using indicator bird species can be established if sufficient data are collected to estimate the toxicological relationships between many bird species according to the structure of the chemicals, and the body weight, order, family, and species of the birds. The procurement of these data will be expensive and time consuming, but it is absolutely necessary if we are to understand how chemicals are affecting our wild bird populations. Regulatory agencies, private and public producers, and environmental groups should collectively gather data to develop cost-effective methods for evaluating the potential hazards of chemicals to wild bird species in a laboratory setting. Undoubtedly none of the

TABLE 1—*Identification of 36 pesticides used to test comparative avian toxicity.*

Common or Accepted Name[a]	Chemical Name[b]	Purity, %
	Organophosphates	
Chlorfenvenfos	phosphoric acid, 2-chloro-1-(2,4-dichlorophenyl)vinyl, diethyl ester	91
Chlorpyrifos	phosphorothioic acid, *0,0*-diethyl *0*-(3,5,6-trichloro-2-pyridinyl) ester	94.5
Coumaphos	phosphorothioic acid, *0,0*-diethyl *0*-(3-chloro-4-methyl-2-oxo-2*H*-1-benzopyran-7-yl) ester	98
Demeton	phosphorothioic acid, *0,0*-dimethyl *0* and *S*- 2-(ethylthio)ethyl esters	92
Diazinon	phosphorothioic acid, *0,0*-diethyl *0*-[6-methyl-2-(1-methylethyl)-4-pyrimidinyl] ester	89
Dichlofenthion	phosphorothioic acid, *0,0*-diethyl *0*-(2,4-dichlorophenyl) ester	>50
Dichlorvos	phosphoric acid, 2,2-dichloroethenyl dimethyl ester	96
Dicrotophos	phosphoric acid, 3-(dimethylamino)-1-methyl-3-oxo-1-propenyl dimethyl ester-*N,N*-dimethylcrotonamide	85
EPN	phosphonothioic acid, phenyl-, *0*-ethyl *0*-(*p*-nitrophenyl) ester	91
Fensulfothion	phosphorothioic acid, *0,0*-diethyl *0*-[4-(methylsulfinyl)phenyl] ester	90
Fenthion	phosphorothioic acid, *0,0*-dimethyl *0*-[3-methyl-4-(methylthio)phenyl] ester	99
Fonofos	phosphonodithioic acid, ethyl-, *0*-ethyl *S*-phenyl ester	94.3
Mevinphos	2-butenoate, methyl ester, 3[(dimethyoxyphosphinyl)oxy]	60
Mocap	phosphorodithioic acid, *0*-ethyl *S,S*-dipropyl ester	95.8
Monocrotophos	phosphoric acid, 1-methyl-3-(methylamino)-3-oxo-1-propenyl (*E*)-dimethyl ester	78
Parathion	phosphorothioic acid, *0,0*-diethyl *0*-(4-nitrophenyl) ester	98.8
Phosfolan	phosphoramidic acid, 1,3-dithiolan-2-ylidene-, diethyl ester	85.9
Phosphamidon	phosphoric acid, dimethyl ester, 2-chloro-3-(diethylamino)-1-methyl-3-oxo-1-propenyl	85
Phoxim	benzeneacetonitrile α{[(diethoxyphosphinothioyl)oxy] imino}	89
Thionazin	phosphorothioic acid, *0,0*-diethyl *0*-pyrazinyl ester	95.6
Trichloronat	phosphonothioic acid, ethyl-, *0*-ethyl *0*-(2,4,5-trichlorophenyl) ester	99
	Carbamates	
Aldicarb	propanal, 2-methyl-2-(methylthio)-, *0*-[(methylamino) carbonyl] oxime	95
Aprocarb	carbamic acid, methyl-, 2-(1-methylethoxy)phenyl ester	97
Bufencarb	carbamic acid, methyl-, *m*-(1-methylbutyl) phenyl ester	68.9
Carbanolate	carbamic acid, methyl-, 2-chloro-4,5-dimethylphenyl ester	>50
Carbofuran	carbamic acid, methyl-. 2,3-dihydro-2,2-dimethyl-7-benzofuranyl ester	99
Methiocarb	carbamic acid, methyl-, 3,5-dimethyl-4-(methylthio)phenyl ester	95
Methomyl	ethanimidothioic acid, *N*-{[(methylamino)carbonyl] oxy}-, methyl ester	90
Mexacarbate	carbamic acid, methyl-, 4-dimethylamino-3,5-dimethylphenyl ester	>99
Mobam	benzo[*b*] thiophene-4-ol, methylcarbamate	98
	Clorinated Hydrocarbons	
Aldrin	1,4:5,8-dimethanonaphthalene, 1,2,3,4,10,10-hexachloro-1,4,4a,5,8,8a-hexahydro-*endo, exo*-	95
Dieldrin	1,4:5,8-dimethanonaphthalene, 1,2,3,4,10,10-hexachloro-6,7-epoxy-1,4,4a,5,6,7,8,8a-octahydro-*endo, exo*-	85
Endrin	1,4:5,8-dimethanonaphthalene, 1,2,3,4,10,10-hexachloro-6,7-epoxy-1,4,4a,5,6,7,8,8a-octahydro-*endo, endo*-	97
Isobenzan	4,7-methanoisobenzofuran, 1,3,4,5,6,7,8,8-octachloro-1,3,3a,4,7,7a-hexahydro-	>99
	Others	
4-Aminopyridine	pyridine, 4-amino	98
Ryanodine	alkaloid from *Ryania speciosa*	>50

[a] Based on nomenclature used by the American National Standards Institute, British Standards Institution, or International Standards Organization.

[b] American Chemical Society nomenclature.

TABLE 2--*Acute oral toxicity of 36 pesticides to six species of birds.*[a]

	LD_{50} in mg/kg					
Chemical	Coturnix Quail	Common Pigeon	Starling	Common Grackle	Red-Winged Blackbird	House Sparrow
Organophosphates						
Chlorfenvenfos	178(100–316)[b]	13.3(NC)[c]	23.7(NC)	17.8(10.0–31.6)	13.3M(NC)	13.3(NC)
Chlorpyrifos	13.3(7.50–23.7)	10.0(5.62–17.8)	75.0(NC)	5.62(3.16–10.0)	13.3M(NC)	10.0(5.62–17.8)
Coumaphos	13.3F(NC)	5.62(3.16–10.0)	75.0(NC)	4.21(NC)	1.78M(1.00–3.16)	10.0(5.62–17.8)
Demeton	13.3F(NC)	13.3(NC)	13.3(NC)	1.78(1.00–3.16)	2.37M(NC)	5.62(3.16–10.0)
Diazinon	4.21F(NC)	3.16(1.78–5.62)	316(100–1000)	7.50(NC)	3.16M(1.78–5.62)	7.50(NC)
Dichlofenthion	316F(NC)	75.0(NC)	2370(465–12100)	75.0(NC)	17.8M(10.0–31.6)	56.2(31.6–100)
Dichlorvos	23.7F(NC)	23.7(13.3–42.1)	42.1(NC)	13.3(NC)	13.3M(NC)	17.8(10.0–31.6)
Dicrotophos	7.50F(NC)	4.21(2.37–7.50)	5.62(3.16–10.0)	1.78(1.00–3.16)	1.0M(0.56–1.78)	4.21(NC)
EPN	10.0F(5.62–17.8)	4.21(NC)	7.50(NC)	4.21(NC)	3.16M(1.78–5.62)	2.37(NC)
Fensulfothion	1.78F(1.00–3.16)	0.56(0.32–1.00)	0.56(0.32–1.00)	0.42(NC)	0.24M(NC)	0.32(0.18–0.56)
Fenthion	17.8F(10.0–31.6)	1.78(1.00–3.16)	17.8(10.0–31.6)	4.21(NC)	1.78M(1.00–3.16)	5.62(3.16–10.0)
Fonofos	31.6F(17.8–56.2)	13.3(7.50–23.7)	42.1(NC)	17.8(10.0–31.6)	10.0M(5.62–17.8)	13.3(NC)
Mevinphos	23.7F(13.3–42.1)	4.21(2.37–7.50)	7.50(NC)	4.21(NC)	1.78M(1.00–3.16)	1.78(1.00–3.16)
Mocap	7.50F(NC)	13.3(NC)	7.50(NC)	10.0(5.62–17.8)	4.21M(NC)	4.21(NC)
Monocrotophos	4.21F(NC)	4.21(2.37–7.50)	5.62(3.16–10.0)	4.21(NC)	1.00M(0.56–1.78)	1.33(NC)
Parathion	4.21F(NC)	1.33(NC)	5.62(3.16–10.0)	5.62(3.16–10.0)	2.37M(NC)	1.33(NC)
Phosfolan	23.7F(NC)	2.37(NC)	5.62(3.16–10.0)	2.37(NC)	2.37M(NC)	2.37(NC)
Phosphamidon	7.50F(NC)	4.21(NC)	5.62(3.16–10.0)	5.62(2.37–13.3)	1.78M(1.00–3.16)	3.16(1.78–5.62)
Phoxim	23.7F(NC)	23.7(NC)	23.7(NC)	75.0(42.1–133)	10.0M(5.62–17.8)	5.62(3.16–10.0)
Thionazin	3.16M(1.78–5.62)	2.37(NC)	7.50(4.21–13.3)	3.16(1.78–5.62)	2.42M(NC)	4.21(NC)
Trichloronat	23.7F(NC)	13.3(NC)	1000(NC)	5.62(3.16–10.0)	4.21M(NC)	5.62(3.16–10.0)
Carbamates						
Aldicarb	4.21F(2.37–7.50)	3.16(1.78–5.62)	4.21(NC)	0.75(0.33–1.69)	1.78M(1.00–3.16)	0.75(0.33–1.69)
Aprocarb	42.1F(23.7–75.0)	7.50(NC)	13.3(NC)	13.3(7.50–23.7)	3.83M(2.46–5.16)	13.3(NC)
Bufencarb	42.1F(NC)	23.7(NC)	75.0(NC)	42.1(NC)	4.21M(NC)	23.7(NC)
Carbanolate	7.50F(NC)	4.21(NC)	11.5(8.47–15.5)	1.78(1.00–3.16)	3.16M(1.78–5.62)	4.21(NC)
Carbofuran	3.16F(1.78–5.62)	1.33(NC)	5.62(3.16–10.0)	1.33(NC)	0.42M(NC)	1.33(NC)
Methiocarb	8.84F(7.24–11.0)	13.3(NC)	23.7(NC)	10.0(5.62–17.8)	4.67M(2.73–6.94)	17.8(10–31.6)
Methomyl	23.7F(NC)	10.0(5.62–17.8)	31.6(17.8–56.2)	23.7(NC)	10.0M(5.62–17.8)	13.3(NC)
Mexacarbate	2.37F(NC)	5.62(3.16–10.0)	23.7(NC)	7.50(NC)	10.0M(3.68–27.1)	7.50(NC)
Mobam	42.1M(NC)	56.2(31.6–100)	750(NC)	17.8(10.0–31.6)	17.8M(10.0–31.6)	23.7(NC)
Chlorinated Hydrocarbons						
Aldrin	42.1F(23.7–75.0)	56.2(31.6–100)	23.7(NC)	7.50(NC)	23.7M(NC)	13.3(7.50–23.7)
Dieldrin	56.2F(31.6–100)	23.7(13.3–42.1)	237(NC)	42.1(23.7–75.0)	17.8M(7.50–42.1)	13.3(3.75–66.8)
Endrin	4.21M(NC)	5.62(3.16–10.0)	2.37(NC)	0.32(0.18–0.56)	2.37M(NC)	1.78(1.00–3.16)
Isobenzan	4.21F(NC)	10.0(5.62–17.8)	2.37(NC)	1.33(NC)	3.16M(1.78–5.62)	1.00(0.56–1.78)
Others						
4-Aminopyridine	7.65M(6.59–8.89)	7.50(NC)	4.90(3.63–6.61)	2.37(NC)	2.37M(NC)	7.50(NC)
Ryanodine	13.3(NC)	2.31(NC)	3.16(1.78–5.62)	1.78(1.00–3.16)	1.78M(1.00–3.16)	2.37(NC)
Overall $\bar{X}$	29.3	13.0	146	12.3	6.07	8.46

[a] The birds are of mixed sexes unless specifically indicated. F = female; M = male.

[b] Numbers in parentheses indicate confidence limits with $P \leq 0.05$.

[c] NC = 95 percent confidence limits not calculable using the statistical method employed.

TABLE 3—*Ranking of relative sensitivity of six species of birds to each of 36 pesticides.*

Chemical	Coturnix Quail	Common Pigeon	Starling	Common Grackle	Red-Winged Blackbird	House Sparrow
			Organophosphates			
Chlorfenvenfos	5.0	2.0	6.0	4.0	2.0	2.0
Chlorpyrifos	5.0	2.0	6.0	1.0	4.0	3.0
Coumaphos	5.0	3.0	6.0	2.0	1.0	4.0
Demeton	5.0	5.0	5.0	1.0	2.0	3.0
Diazinon	3.0	1.5	6.0	4.5	1.5	4.5
Dichlofenthion	5.0	3.5	6.0	3.5	1.0	2.0
Dichlorvos	6.0	4.5	1.5	4.5	1.5	3.0
Dicrotophos	6.0	3.5	5.0	2.0	1.0	3.5
EPN	6.0	3.5	5.0	3.5	2.0	1.0
Fensulfothion	6.0	4.5	4.5	3.0	1.0	2.0
Fenthion	5.5	1.5	5.5	3.0	1.5	4.0
Fonofos	5.0	2.5	6.0	4.0	1.0	2.5
Mevinphos	6.0	3.5	5.0	3.5	1.5	1.5
Mocap	3.5	6.0	3.5	5.0	1.5	1.5
Monocrotophos	4.0	4.0	6.0	4.0	1.0	2.0
Parathion	4.0	1.5	5.5	5.5	3.0	1.5
Phosfolan	6.0	2.5	5.0	2.5	2.5	2.5
Phosphamidon	6.0	3.0	4.5	4.5	1.0	2.0
Phoxim	4.0	4.0	4.0	6.0	2.0	1.0
Thionazin	3.5	1.5	6.0	3.5	1.5	5.0
Trichloronat	5.0	4.0	6.0	2.5	1.0	2.5
			Carbamates			
Aldicarb	5.5	4.0	5.5	1.5	3.0	1.5
Aprocarb	6.0	2.0	4.0	4.0	1.0	4.0
Bufencarb	4.5	2.5	6.0	4.5	1.0	2.5
Carbanulate	5.0	3.5	6.0	1.0	2.0	3.5
Carbofuran	5.0	3.0	6.0	3.0	1.0	3.0
Methiocarb	2.0	4.0	6.0	3.0	1.0	5.0
Methomyl	4.5	1.5	6.0	4.5	1.5	3.0
Mexacarbate	1.0	2.0	6.0	3.5	5.0	3.5
Mobam	4.0	5.0	6.0	1.5	1.5	3.0
			Chlorinated Hydrocarbons			
Aldrin	5.0	6.0	3.5	1.0	3.5	2.0
Dieldrin	4.0	5.5	5.5	3.0	2.0	1.0
Endrin	5.0	6.0	3.5	1.0	3.5	2.0
Isobenzan	5.0	6.0	3.0	2.0	4.0	1.0
			Others			
4-Aminopyridine	6.0	4.5	3.0	1.5	1.5	4.5
Ryanodine	6.0	3.5	5.0	2.0	1.0	3.5
Overall $\bar{x}$	4.81	3.51	5.10	3.07	1.88	2.72

TABLE 4—*Levels of significance for differences in LD_{50}'s determined with 36 pesticides.*

Species	LD_{50} $\bar{x}$ (mg/kg)	Significance, *P* 0.01	Significance, *P* 0.05
Starling	146.38	a[1]	a
Coturnix quail	29.32	ab	ab
Common pigeon	12.99	abc	abc
Common grackle	12.31	abc	bc
House sparrow	8.46	bc	c
Red-winged blackbird	6.07	c	c

[1] LD_{50}'s followed by the same letter are not significantly different at the indicated level of significance.

methods developed will be perfect in the predictions they generate; however, the methods should be based on facts, not assumptions, and should address the following questions.

1. What types of tests are necessary to establish a scientific basis for predicting the potential hazards of chemicals to wild birds?
2. What are the most cost-effective methods for conducting these tests while retaining scientific validity?
3. Which species and how many species should be used in the evaluations?
4. To how many other species can these data be extrapolated while retaining a reasonable degree of confidence in the extrapolation?

Since any change in test methods will involve testing laboratories and pesticide developers, it is imperative that the transition be accomplished in a smooth and orderly manner. New methods should not only use new data, but also allow for extrapolations from existing data, provided that sound background data for such extrapolations are available. This effort should produce a more meaningful evaluation of the potential hazards of pesticides to wild birds, and also result in more cost-effective methods that incorporate the state of the art.

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