

## Mercury Residues in Tissues of Dead and Surviving Birds Fed Methylmercury

Mack T. Finley<sup>1</sup>, William H. Stickel<sup>2</sup>, and R. E. Christensen<sup>3</sup>

<sup>2</sup>U.S. Fish and Wildlife Service, Patuxent Wildlife Research Center, Laurel, Md. 20811,

<sup>3</sup>WARF Institute, Inc., Madison, Wis. 53701

Organic mercury compounds, particularly methylmercury, became widely distributed in the environment and caused human illnesses before recognition of the problem and the imposition of bans. Prevalence of mercury residues in wild birds has suggested the possibility of problems with these species also (BORG et al. 1969, MARTIN 1972, DUSTMAN et al. 1972, BASKETT 1975). The identification of critical organs, critical concentrations, and critical effects of mercury in man has received intensive study (NORDBERG 1976). Some attention has been given to these key factors as they relate to birds, but results have been inconclusive (FIMREITE 1971, GARDINER 1972, HOUGH and ZABIK 1972).

The objective of the present study was to determine whether a critical organ and critical concentration for lethality could be identified for birds fed high dosages of methylmercury.

### MATERIALS AND METHODS

Test animals consisted of four species of birds that were trapped during late summer near Laurel, Maryland. First-year starlings (*Sturnus vulgaris*), grackles (*Quiscalus quiscula*), red-winged blackbirds (*Agelaius phoeniceus*), and adult cowbirds (*Molothrus ater*) were held in large outdoor wire pens equipped with running water and shelter. For each species, groups of 14 males were fed ad libitum turkey starter crumbles containing 40 ppm methylmercury (as 1.5% methylmercury dicyandiamide in Morsodren). Additional groups of 14 birds of each species served as controls and received untreated feed. Feed was prepared by mixing a solution of Morsodren in propylene glycol into the crumbles; the carrier was equivalent to 2% (w/w) of the total diet. The control diet contained an equivalent amount of propylene glycol. Birds were maintained on mercury diets until 5 of the 14 birds of a treatment group were dead or dying. Following the death of the fifth bird, 5 apparently healthy survivors of the same group were sacrificed with chloroform.

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<sup>1</sup>Present address: Columbia National Fishery Research Laboratory, Columbia, Mo. 65201

Birds were weighed, necropsied, and prepared for analysis by removing the skin, beaks, feet, wings, and digestive tracts. Brains, livers, kidneys, breast muscles, and carcasses were weighed fresh and preserved in 10% formalin. Samples were shipped to WARF Institute, Inc., Madison, Wisconsin for mercury analysis by modified atomic absorption procedures (Analytical Methods Committee, 1961). Tissues were drained and weighed and the formalin from each sample was measured and saved. Samples of whole brains, kidneys, livers, and 5 g of breast muscle were analyzed. Remaining carcasses were ground and 5 g samples analyzed. Samples were homogenized and digested by refluxing with sulfuric and nitric acid. A mixture of hydroxylamine hydrochloride, stannous chloride, and sulfuric acid was added to the digest to reduce ionic mercury to elemental mercury. Since preliminary analysis revealed that low levels of mercury leached from soft tissues into the formalin, 10 g aliquots of formalin were analyzed from all samples. Residues are reported as ppm based on fresh wet weight with limits of sensitivity of 0.05 ppm. Residues were corrected for loss of mercury from tissues into formalin. Mercury that leached into the formalin averaged only 0.9 to 2.5% for tissues of dosed birds, but was considerably higher (22 to 46%) for the controls where the concentrations were low. Average mercury recovery for the analytical method was  $93.4\% \pm 2.4$  (S.E.) based on 5 spiked samples. Data were not corrected for recovery.

## RESULTS

### Condition of Birds

Grackles were the most susceptible to mercury poisoning; the times to death of the fifth bird of each species were: 6 days for grackles, 8 for starlings, 10 for cowbirds, and 11 for redwings. Grackles that died lost only an average of 4% of their body weight. Cowbirds, redwings, and starlings lost both flesh and fat, and an average of 21, 22, and 28% of their body weight, respectively. Presumably the grackles would have lost more weight had they not been so susceptible to mercury poisoning.

Starlings that were sacrificed lost an average of 27% of their body weight. Redwings, grackles, and cowbirds, however, weighed an average of 1, 4, and 5% more at sacrifice than they did before treatment. All starlings were near death when sacrificed. No control birds died and necropsy showed that all were in good condition. Signs of mercury poisoning in treated birds were weakness, ataxia, and drowsy dullness. Necropsy revealed numerous abnormalities in mercury-killed birds including vascular suffusion of the brain, pale or mottled kidney, extensive hemorrhage in livers, and yellowish-green fluid deposits under the skin and in the body cavity. However, these abnormalities did not occur in a consistent enough pattern to characterize mercury poisoning.

## Mercury Residues

Levels of mercury in pre-treatment birds (TABLE 1) were higher than anticipated and may reflect a low-level chronic exposure; they are, however, trivial in relation to levels in the dosed birds. In all four species, birds that died from mercury

TABLE 1

Mercury residues (ppm) in pre-treatment samples of birds. @

Tissue	Species			
	Cowbird	Redwing	Starling	Grackle
Brain	0.91	0.28	0.30	0.18
Liver	1.13	0.94	0.18	0.10
Kidney	1.39	1.79	0.36	0.76
Muscle	0.55	0.24	0.09	0.07
Carcass	0.07	0.08	0.14	0.09
Whole body	0.69	0.22	0.14	0.11

@ One bird of each species was randomly selected for mercury analysis.

poisoning contained levels of mercury similar to those in sacrificed birds (TABLE 2). Residue levels were highest in the livers, followed by kidneys and brains. Levels of mercury were similar in breast muscle, carcass, and whole body. Residue levels were higher in most tissues of birds that died, but concentrations did not differ significantly ( $P < 0.05$ ) except in carcasses of redwings. Carcasses of redwings that died contained an average of 48.2 ppm mercury compared with 41.7 ppm in the carcasses of birds that were sacrificed. Mercury concentrations in brains and livers were slightly higher in all groups of birds that died compared with sacrificed groups. Residue levels in kidneys were slightly higher in birds that were sacrificed. Since few surviving starlings were in good condition following death of the fifth bird, tissues of sacrificed birds were not analyzed.

Comparison of the ratios of mercury in liver-kidney failed to show significant differences between dead and sacrificed birds. Ratios of mercury in kidney-whole body were higher in sacrificed birds, but the differences were significant ( $P < 0.05$ ) only for redwings. Liver-whole body ratios were higher in birds that died, but differences were not significant.

Mercury levels among species differed consistently in both dead and sacrificed birds (TABLE 2). Mercury levels usually were highest in redwings, lowest in grackles, and intermediate in starlings and cowbirds. Differences in levels of mercury accumulated in tissues of grackles and redwings reflect the duration of mercury ingestion necessary to cause death.

TABLE 2  
Mercury residues (ppm) in tissues of birds. Means  $\pm$  S.E., N=5@

Species		Died		Sacrificed
Brain				
Redwing	a	45.0 $\pm$ 2.7	a	40.4 $\pm$ 0.9
Starling	a	44.9 $\pm$ 3.4		#
Cowbird	b	30.9 $\pm$ 4.5	b	29.1 $\pm$ 3.3
Grackle	c	21.0 $\pm$ 1.5	c	20.3 $\pm$ 1.7
Liver				
Redwing	a	126.5 $\pm$ 17.9	a	92.8 $\pm$ 8.8
Starling	ab	103.6 $\pm$ 15.7		#
Cowbird	bc	78.4 $\pm$ 16.1	b	66.6 $\pm$ 6.3
Grackle	c	54.5 $\pm$ 6.4	b	53.1 $\pm$ 4.7
Kidney				
Redwing	a	74.3 $\pm$ 8.6	a	83.6 $\pm$ 6.1
Starling	a	86.4 $\pm$ 5.5		#
Cowbird	b	48.9 $\pm$ 6.3	b	60.8 $\pm$ 7.9
Grackle	b	40.4 $\pm$ 5.5	c	44.0 $\pm$ 3.6
Muscle				
Redwing	a	57.1 $\pm$ 7.2	a	44.4 $\pm$ 4.9
Starling	b	40.7 $\pm$ 3.0		#
Cowbird	bc	31.5 $\pm$ 4.0	b	29.0 $\pm$ 2.0
Grackle	c	30.0 $\pm$ 2.6	b	30.3 $\pm$ 2.2
Carcass				
Redwing	a	48.2 $\pm$ 2.5*	a	41.7 $\pm$ 1.2
Starling	b	36.4 $\pm$ 4.0		#
Cowbird	b	33.9 $\pm$ 4.1	b	30.2 $\pm$ 3.6
Grackle	c	26.1 $\pm$ 2.9	b	26.3 $\pm$ 2.1
Whole body				
Redwing d/	a	54.2 $\pm$ 3.6	a	45.8 $\pm$ 1.8
Starling	b	42.8 $\pm$ 3.9		#
Cowbird	bc	37.2 $\pm$ 4.6	b	32.7 $\pm$ 3.0
Grackle	c	29.2 $\pm$ 3.0	b	29.2 $\pm$ 1.8

@ The "Died" column shows residues in the first 5 birds that died from ingesting mercury; "Sacrificed" birds consist of 5 apparently healthy birds selected at random from treated groups after the first 5 had died. Any two means in a column preceded by a common letter are not significantly different, Scheffe's test,  $P \leq 0.05$ .

# Surviving starlings were visibly affected and therefore were not analyzed for mercury.

\* Significantly different from mean of survivors that were sacrificed, Student's  $t$ -test,  $P \leq 0.05$ .

d/ Mercury in whole bodies is based on the total micrograms in all tissues after carcass preparation.

## DISCUSSION

In the present study, there were no clearcut differences between concentrations of mercury in tissues of birds that died and in those that were sacrificed while in apparently good condition. Levels tended to be higher in the birds that died, but differences were not statistically significant. Concentrations of mercury in the tissues of these birds were similar to those in other species known to have died of mercury poisoning. For example, FIMREITE and KARSTAD (1971) reported that more than 20 ppm of mercury in livers of hawks could be lethal; and KOEMAN et al. (1971) reported averages of 83, 74, 36, and 27 ppm of mercury in livers, kidneys, breast muscles, and brains, respectively, of kestrels that died as a result of eating mice containing an average of 13.3 ppm of mercury.

Our results with mercury closely parallel those of LONGCORE et al. (1974) with lead, both showing very clear differences between these heavy metals and the lipid-borne organochlorine compounds (STICKEL 1975). In our study, mercury treatment was severe enough to kill over one-third of the birds in a very short time. Sacrificed birds also may have been critically exposed even though they appeared normal. These results should not be generalized to depict the residues or effects of prolonged exposure to low levels of mercury. However, it can be concluded that concentrations of mercury in excess of 20 ppm in soft tissues should be considered extremely hazardous.

## SUMMARY

Concentrations of mercury in passerine birds fed diets containing 40 ppm methylmercury were similar in tissues of birds that died from mercury poisoning and in those that were sacrificed after half the group had died. Residues were higher in tissues of birds that died, but the differences were not statistically significant. Residue levels were highest in livers, followed by kidneys and brains. Levels of mercury were similar in breast muscle, carcass, and whole body. Mercury levels were highest in redwinged blackbirds, lowest in grackles, and intermediate in starlings and cowbirds. Mercury concentrations exceeded 20 ppm in all tissues of all species and were similar to levels reported in wild birds known to have died of mercury poisoning.

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