AVIAN AND MAMMALIAN WILDLIFE TOXICOLOGY: SECOND CONFERENCE

A symposium sponsored by ASTM Committee E-35 on Pesticides AMERICAN SOCIETY FOR TESTING AND MATERIALS Louisville, Ky. 18 March 1980

ASTM SPECIAL TECHNICAL PUBLICATION 757 D. W. Lamb, Mobay Chemical Corp., and E. E. Kenaga, The Dow Chemical Co., editors

ASTM Publication Code Number (PCN) 04-757000-48



Copyright© by American Society for Testing and Materials 1981 Library of Congress Catalog Card Number: 81-68429

NOTE

The Society is not responsible, as a body, for the statements and opinions advanced in this publication.

Printed in Baltimore, Md. December 1981

Foreword

The second symposium on Avian and Mammalian Wildlife Toxicology was held on 18 March 1980 in Louisville, Ky. The event was sponsored by the American Society for Testing and Materials, through its Committee E-35 on Pesticides and its Subcommittee E35.25 on Wildlife Toxicology and Hazard Assessment. Cochairing the symposium were D. W. Lamb, Mobay Chemical Corp., and E. E. Kenaga, Dow Chemical Co., both of whom also served as editors of this publication.

Related ASTM Publications

- Avian and Mammalian Wildlife Toxicology: First Conference, STP 693 (1979), \$16.25, 04-693000-48
- Ecological Assessments of Effluent Impacts on Communities of Indigenous Aquatic Organisms, STP 730 (1981), \$32.50. 04-730000-16
- Aquatic Toxicology: Third Conference, STP 707 (1980), \$39.50, 04-707000-16
- Aquatic Toxicology: Second Conference, STP 667 (1979), \$37.75, 04-667000-16
- Vertebrate Pest Control and Management Materials, STP 680 (1979), \$31.50, 04-680000-48
- Test Methods for Vertebrate Pest Control and Management Materials, STP 625 (1977), \$26.00, 04-625000-48

A Note of Appreciation to Reviewers

This publication is made possible by the authors and, also, the unheralded efforts of the reviewers. This body of technical experts whose dedication, sacrifice of time and effort, and collective wisdom in reviewing the papers must be acknowledged. The quality level of ASTM publications is a direct function of their respected opinions. On behalf of ASTM we acknowledge with appreciation their contribution.

ASTM Committee on Publications

Editorial Staff

Jane B. Wheeler, Managing Editor
Helen M. Hoersch, Senior Associate Editor
Helen P. Mahy, Senior Assistant Editor
Allan S. Kleinberg, Assistant Editor
Virginia M. Barishek, Assistant Editor

Contents

Introduction	1
Use of Captive Starlings to Determine Effects of Pollutants on Passer-ine Reproduction—C. E. GRUE AND C. L. CHRISTIAN	5
Reproductive Tests of Diazinon on Bobwhite Quail—K. L. STROMBORG	19
Genetic Response of Mallards to Toxic Effects of Methyl Mercuric Chloride—H. H. PRINCE	31
Subacute Toxicity Testing with Young Birds: Response in Relation to Age and Intertest Variability of LC ₅₀ Estimates—E. F. HILL AND M. B. CAMARDESE	41
The Laughing Gull (Larus atricilla) as a Model for Assessment of Secondary Poisoning—R. J. FINK AND M. J. JABER	66
Diagnostic Brain Residues of Dieldrin: Some New Insights G. H. HEINZ AND R. W. JOHNSON	72
Formalin Preservation of Avian Blood for Organochlorine Analysis— C. J. STAFFORD AND W. H. STICKEL	93
Development of an Avian Dietary LC ₅₀ Toxicity Test for Potential Use Under the Toxic Substances Control Act—L. w. Turner	98
Subacute Dietary Toxicities of Dicrotophos and Dieldrin in Time- Replicated Trials with Young Ring-Necked Pheasants and Mallards—E. F. HILL	105
Placental and Mammary Transfer of Polychlorinated and Polybrominated Biphenyls in the Mink and Ferret—M. R. BLEAVINS, R. J. AULERICH, AND R. K. RINGER	121
A Short-Term Dietary Toxicity Test on Small Mammals—	122
J. A. MCCANN, W. TEETERS, D. J. URBAN, AND N. COOK	132

Acute and Subacute Toxicity of Pesticides in Microtine Rodents—	
J. M. CHOLAKIS, M. J. MCKEE, L. C. K. WONG, AND J. D. GILE	143
Summary	155
Index	159

Introduction

This ASTM symposium, which was held 18 Oct. 1980 in Louisville, Ky., was organized as a continuing outlet for papers of current interest in avian and mammalian wildlife toxicology. Further, it served as an informal meeting for exchange of ideas by laboratory, field, and regulatory scientists at a time when a critical need was being felt for interpretation of toxicity data. National decision making regarding the identification of significant effects and hazards of pesticides and other toxic materials on wildlife populations makes use of practical scientific work designed to answer current problems and enable us to avoid serious future problems.

Some of the important wildlife toxicology problems which scientists need to formulate answers to at all levels of decision making are the following:

1. The selection of surrogate species.

Since we cannot ever test all wildlife species, let alone endangered species. we must identify those which are sufficiently representative of large groups of species and also responsive to chemicals of various representative structural groups over a wide range of concentrations. These surrogate species must be useful for answering problems of both acute and chronic toxicity. Some test species must represent large groups of unrelated species for preliminary toxicological investigations. Other species may be used to represent specific target species or closely related species in more extended investigations, which are useful for toxicological testing of chemicals with undesirable properties of toxicity, persistence, or distribution in the environment (for example, DDT, PCBs). These surrogate species should be selected from data based on a sufficient number of experiments with benchmark chemicals for comparative toxicology. Without a sufficient comparative toxicological base the data, if published, can add to the confusion of decision making because of anomalous effects which often occur and which might not be interpretable or, perhaps, not even due to the chemical being studied. For this reason, well-studied species must be selected from those in which the normal daily, seasonal, or yearly variation of life processes affecting them can be compared with those changes identified as resulting from man-made chemical exposures. The preceding comments, which relate to the comparative toxicology data base, apply to the test method as well as to the test species. One cannot be discussed independently of the other.

Knowledge of toxicological responses is limited to a relatively few species of organisms, particularly those used in chronic toxicity tests, which include

life cycles. The choices are few. We need a larger data base. At present it is necessary to pick "surrogates for the surrogates." Another important qualification for at least some of the surrogate species is that they be available for testing both in the laboratory and the field, in order to build a basis of prediction of field toxicological responses from laboratory results. Unlike toxicology studies, in which the rat or some other species is the surrogate for humans, field and laboratory comparisons for many wildlife species can be conducted using the same species.

2. Predicting toxicity.

Scientists must be alert to the conclusions to be developed from the increasing data base on wildlife toxicity and not go blindly on with the same hazard assessment procedures used when the data base was lacking. We now know that certain species and test methods are closely related to others and that effects on these species can be predicted by the use of regression equations within given confidence limits. Where available, these confidence limits are useful for hazard evaluation if the environmental exposure concentrations are below the lowest confidence limits of the predicted toxicological noeffect concentration values. Before being too critical of the predictive values we must realize that the confidence limits of experimental tests are also often large. The question remains—how precise must these toxicological values be?

The most expensive toxicological tests are chronic tests. We have enough data to know that the difference in dosage between acute and chronic effects usually falls between approximately a 10 to 1000-fold (application) factor. If the environmental concentrations are determined to be less than 1000-fold that of the laboratory acute toxicity $LC_{50}s$, then the chances are good that the margin of safety is sufficient and toxicity will not occur in the field. Application factors, experimentally determined from closely related chemical structures, in some cases can be used to predict a smaller application factor to within one order of magnitude. These predictive procedures can obviate the need for a considerable amount of testing if used selectively where they can be most effective.

3. Matching the concentrations of chemicals in various media (for example, water, food) with the concentrations in organisms which cause various toxicological effects or responses.

We know that when 16 ppm of dieldrin occurs in the brain tissues of birds, death ensues. We need much more data with other chemicals to establish these types of relationships. Very few toxicological data have been collected on the distribution of chemicals in body tissues as related to lethality as a major purpose of study. Various approaches can be taken to derive these data from existing field data. For example, multiplication of the field concentrations of chemicals in media, such as the food of organisms, by the appropriate known or calculated bioconcentration factor produces a prediction of the

concentrations occurring in the organisms. Such organism concentrations can be matched with observed acute or chronic toxicological effects, such as mortality, reproduction effects, and so forth, to build up the desired data base.

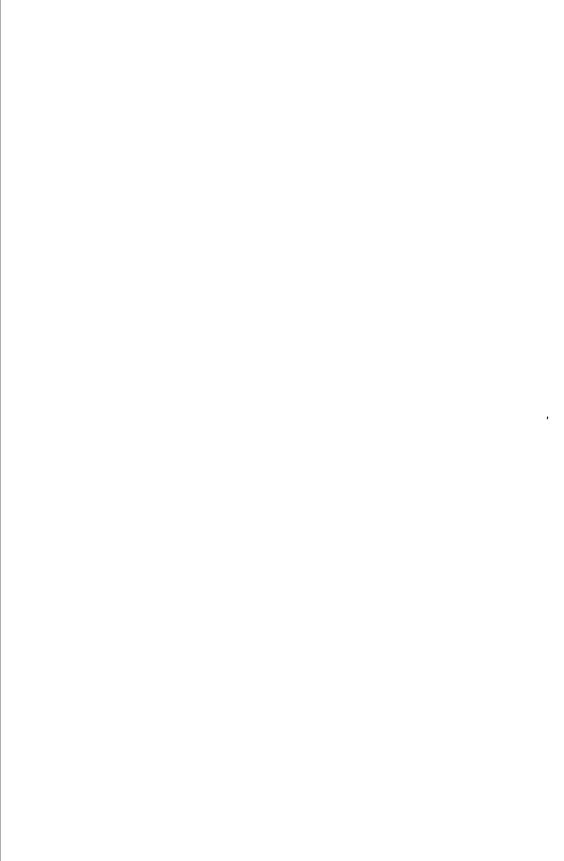
This ASTM symposium provides clues to some of the pressing data needs for assessment of toxicological hazards of chemicals to wild birds and mammals. Obviously, this search for knowledge is a continuing saga of scientific adventure which is exciting in its unfolding revelations and whose end is not yet in sight.

E. E. Kenaga

The Dow Chemical Co., Midland, Mich. 48640; symposium cochairman and coeditor.

D. W. Lamb

Mobay Chemical Corp., Kansas City, Mo. 64120; symposium cochairman and coeditor.



Use of Captive Starlings to Determine Effects of Pollutants on Passerine Reproduction

REFERENCE: Grue, C. E. and Christian, C. L., "Use of Captive Starlings to Determine Effects of Pollutants on Passerine Reproduction," Avian and Mammalian Wildlife Toxicology: Second Conference, ASTM STP 757, D. W. Lamb and E. E. Kenaga, Eds., American Society for Testing and Materials, 1981, pp. 5-18.

ABSTRACT: Three reproductive trials were conducted to develop techniques for propagation of captive starlings (Sturnus vulgaris) which could determine the effects of environmental contaminants on passerine reproduction. Trials were conducted during the spring of 1979 in five adjacent 2.4 by 3 by 12-m outdoor wire pens containing four or ten pairs of starlings, a similar number of nest boxes, perches, water, commercial turkey starter, and alfalfa hay as nesting material. Nestling diets consisted of combinations of Nebraska Brand bird of prey diet, live or frozen mealworms (Tenebrio molitor) and crickets (Acheta domestica), or live earthworms (Pheretima sp.). Starlings reproduced successfully when the number of breeding pairs per pen was reduced from ten to four. The average clutch sizes for each pen (4.3 to 4.9) were similar to those reported for wild starlings. Hatching (60 to 90.4 percent) and fledging (0 to 100 percent) success varied among pens. The fledging success was greatest in the pens which received the most diverse nestling diets: Nebraska Brand diet plus frozen or live mealworms and crickets. Whether the insects were presented alive or frozen appeared to have little effect on the reproductive success. The starlings did not consume or carry earthworms to their young. The body weights of 20-day-old nestlings raised in captivity (X = 73.9 g) were similar to those of starlings in the wild. The use of single pairs per pen may eliminate problems in presentation of nestling diets due to asynchrony in breeding between pairs and excessive interactions among individuals, which may interfere with parental care. The starling appears to be an excellent model for examining the effects of environmental contaminants on the reproduction of songbirds in captivity.

KEY WORDS: captive, passerine, pesticides, pollutants, reproduction, starling, *Sturnus vulgaris*, toxicity testing, toxicology, wildlife

Studies have shown that pollutants, including organochlorine, organophosphate, and carbamate pesticides (for review, see Ref 1),² polychlorinated

¹Biologist and biological aid, respectively, U.S. Fish and Wildlife Service, Patuxent Wildlife Research Center, Laurel, Md. 20708.

²The italic numbers in brackets refer to the list of references appended to this paper.

biphenyls (PCBs) (for review, see Ref 2), and heavy metals (for example, see Refs 3 and 4), can affect behavioral and physiological parameters essential for reproductive success in birds. Pesticide applications often coincide with avian breeding seasons when insects and birds are most abundant. However, relatively little is known about the effects of environmental contaminants on the reproduction of songbirds, though approximately 60 percent of all living species of birds are Passeriformes [5]. Past research on the effects of contaminants on passerine reproduction has concentrated on the effects of dichlorodiphenyltrichloroethane (DDT) and its metabolites [6-16]. Relatively few studies [17-25] have examined the effects of the newer pesticides, organophosphates and carbamates, and only five of these have examined reproductive effects in some detail. The effects of PCBs and heavy metals on passerine reproduction have not been investigated, with the exception of one study [26] which examined the effects of automotive lead on barn swallows (Hirundo rustica).

Studies on the effects of a wide variety of contaminants on songbird reproduction will be difficult in the field because of limited availability of study areas that can provide suitable acreage, treatment, accessibility, and sample size. Development of a methodology for the captive reproduction of a North American songbird would facilitate determination of the effects of pollutants on passerine reproduction. Research possibilities would be numerous without many of the problems and costs associated with field studies or colony maintenance because birds would have to be housed in test pens only during the breeding season. A similar approach [12] was used successfully in studying the effects of DDT and dichlorodiphenyldichloroethylene (DDE) on reproduction of the Bengalese finch (Lonchura striata).

In the present study, we selected the starling because of its abundance, adaptability, and the excellent success Risser [27] and Schafer et al [28] have had in inducing this species to lay eggs and hatch young in captivity. Because of their research objectives, however, neither Risser nor Schafer and his coworkers allowed the adults to raise their young. Previous attempts [29,30] to breed starlings in captivity have failed. However, both Risser [27] and Schafer et al [28] suggested that reproduction in captivity may be possible if adults are provided with a diet that can be taken to their young. It was the objective of our study to develop this portion of the methodology.

Methods

Trial I

Ten nest boxes of the dimensions described by Kessel [31] were placed within each of five adjacent pens 3 m wide, 2.4 m high, and 12 m long (Fig. 1). Among the existing pens at the Patuxent Wildlife Research Center, Laurel, Md., these were the most similar to those used by Risser and by

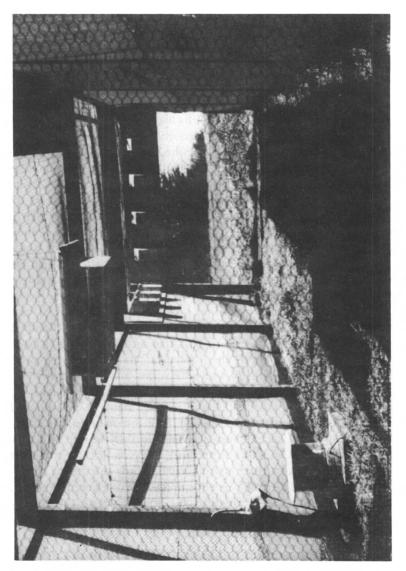


FIG. 1-View of one of the five adjacent pens used during Trial I.

Schafer and his co-workers. Boxes were mounted along the sides and back walls of each pen with entrance holes 0.6 m apart and 2 m above the ground. The distance between the nest boxes exceeded the reported diameter of the territory of male starlings around a cavity entrance [31,32]. Each pen contained a large hanging feeder (0.3 by 0.6 m), a water pot placed on a heater, and two laterally placed 7-m perches. Burlap was placed on the side walls to provide visual isolation. Diets before egg laying consisted of commercial turkey starter [Turkey Starter AP (medicated) Crumpels,³ Beacon Milling Co., Inc., Cayuga, N.Y.]. A quarter bale of green alfalfa hay was provided as nesting material.

On 30 and 31 Jan. 1979, 20 adult starlings, 10 of each sex, were weighed, banded, and placed in each pen. These birds had been housed in large outdoor pens since March 1978 and were, therefore, at least second-year birds. Starlings were sexed using the methods described by Kessel [33]. Nest boxes were checked every other day at 3 P.M. The adult starlings were not handled while they were within their nest boxes. When the boxes were opened for inspection, most of the birds vacated them. As part of another study, the authors also monitored the reproductive activity of wild starlings within 75 nest boxes located elsewhere on the Patuxent Wildlife Research Center. These data were then compared with those observed within the pens.

Commercial turkey starter and high-protein animal matter, which we believed would be suitable as nestling food, were made available to adults 5 days before the hatching of the first clutch in each pen. The nestling diets consisted of (1) Nebraska Brand bird of prey diet (Central Nebraska Packing Co., North Platte, Neb.), (2) live mealworms (Tenebrio molitor, Rainbow Mealworms, Compton, Calif.), (3) live mealworms and live crickets (Acheta domestica, Ghann's Cricket Farm, Inc., Augusta, Ga.), (4) frozen mealworms and frozen crickets, and (5) live mealworms and Nebraska Brand diet. The selection of diets was based on their availability and practicality for use in experiments with contaminants (the primary route of poisoning of birds by most pesticides appears to be through the ingestion of dead or struggling insects following application of the pesticide [34]). Diets were randomly assigned to the pens. The Nebraska Brand diet was ground using a Hobart blender in an effort to make it more attractive to the birds and similar in consistency to the diet after a toxicant was added. The crickets were full grown. Two samples of each diet were analyzed for organochlorines and heavy metals to determine the baseline levels of contaminants.

Trial II

On 6 April 1979 the number of pairs within each of the five pens was reduced to four. All but four nest boxes were removed; the remaining boxes were evenly spaced (Fig. 2). The existing nests, eggs, and young were

³Use of trade names and names of suppliers is for identification purposes only and does not constitute endorsement by the federal government.

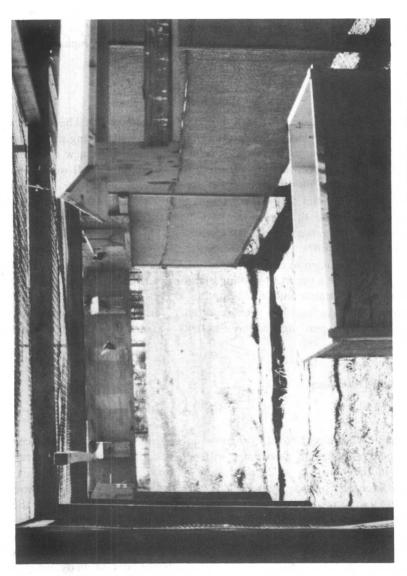


FIG. 2—Spatial distribution of the nest boxes in each of the adjacent pens used in Trials II and III. A feeder containing live crickets is shown in the foreground.