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AN OVERVIEW OF CONTAMINANTS AND BATS, WITH SPECIAL REFERENCE TO INSECTICIDES AND THE INDIANA BAT

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Abstract

Bats accumulate and suffer from adverse effects of environmental contaminants, and this raises concern about potential impacts on endangered species, such as the Indiana bat (*Myotis sodalis*). Organochlorine insecticides are persistent, lipophilic neurotoxins that have been implicated as agents of mortality in populations of other species of bat, including gray bats (*Myotis grisescens*) in Missouri. We provide previously unpublished data, obtained in 1975–1978, on organochlorines in carcasses and brains of 38 Indiana bats and guano from five roosts. These data provide the first evidence for mortality of Indiana bats due to insecticides of the organochlorine pesticide era and provide a benchmark for future studies. Most organochlorines have been eliminated from use in the United States, but they are still found in decreasing amounts in bats and continue to be elevated at "hotspots" of contamination. Organophosphate, carbamate, and pyrethroid insecticides are used most widely in place of organochlorines, but their occurrence and effects in bats are poorly known. Three studies suggest inhibition of cholinesterase in brains of bats caused by exposure to organophosphates or carbamates. Pyrethroid insecticides recently were detected in tissues or guano of some bats, including species of *Myotis* in Missouri. We compare data on insecticide use and agricultural production from counties with records of reproductively active females or juveniles in Missouri and Indiana, searching for differences associated with declines in populations of Indiana bats at hibernacula in Missouri but not in Indiana. Intensity of agricultural production, types of crops requiring insecticides, and general insecticide use and intensity were higher in Indiana. However, pyrethroid insecticides were used more in Missouri. Use of insecticides in southeastern Missouri is very intensive because of applications to cotton, and this area should be investigated for use by Indiana bats and their possible exposure to insecticides.

Key words: agriculture, bats, carbamates, contaminants, Indiana, insecticides, Missouri, *Myotis*, organochlorines, organophosphates, pyrethroids

Introduction

Bats generally produce only one or two young annually and require high annual survival rates to maintain stable populations. Concern about lethal impacts of exposure to environmental contaminants, particularly insecticides, on bat populations originally was expressed ca. 50 years ago (reviewed by Clark and Shore 2001). Although subsequent research showed that specific contaminants contribute to declines in bat populations (Clark et al. 1978, Geluso et al. 1976), much remains to be learned about the importance of contaminants in management of bats. For example, geographic differences in use of insecticides could result in differences in mortality and account, in part, for the different status of populations of Indiana bats (*Myotis sodalis*) in Missouri (declining) and Indiana (stable or increasing—U.S. Fish and Wildlife Service 1999). However, there is little information on the relationship between Indiana bats and environmental contaminants, and recovery efforts for the Indiana bat specifically have called for increasing knowledge on this topic (U.S. Fish and Wildlife Service 1999).

The purpose of our paper is fourfold. First, we summarize the scientific literature about bats and environmental contaminants to provide background knowledge for researchers and managers of Indiana bats. Second, we provide the first assessment of contamination of Indiana bats during the era of organochlorine (OC) pesticides, and third, we make a preliminary assessment of use of insecticides in key

areas utilized by Indiana bats as summer foraging habitat. Finally, we suggest avenues for future research.

Materials and Methods

We provide information about concentrations of OC insecticides in carcasses, brains, and guano of Indiana bats sampled in Indiana, Kentucky, and Missouri, from 1975 to 1978. These samples were gathered during field projects by others and analyzed at the Patuxent Wildlife Research Center, Laurel, Maryland, following standard protocols (Clark et al. 1978, 1980, 1982). We tabulate previously unpublished results of chemical analyses based on individual samples to allow researchers to make a full range of statistical comparisons with data from future surveys (Appendix I). Results are provided for three OCs or metabolites (*p,p'*-DDE, dieldrin, and heptachlor epoxide) that have been implicated in mortality of other species of bat in the United States.

Clark and Shore (2001) recently provided a comprehensive, global summary of literature on contaminants in bats, and therefore, we limit our review to information pertinent to management of the Indiana bat. The proposed recovery plan for the Indiana Bat stresses the need for information on effects of pesticides, particularly insecticides (U.S. Fish and Wildlife Service 1999). We, therefore, summarize data on agricultural production and total use of insecticides for counties with maternity colonies of Indiana bats in Indiana and Missouri, to determine if differences exist in kinds of crops, total use of insecticides, or kinds of insecticides applied. We assume that counties with records of reproductively active female or juvenile Indiana bats (appendix I in U.S. Fish and Wildlife Service 1999) contain maternity colonies.

Sources of data on crops and total use of insecticides are the three most recent censuses of agriculture, conducted in 1987, 1992, and 1997 (U.S. Department of Agriculture 2001), and we sum the data from these sources for each county in the analysis. Areas of each county are provided by the U.S. Bureau of the Census (1994). Information on specific insecticides used in 1992 and 1997 is given by the National Center for Food and Agricultural Policy (Gianessi and Marcelli 2000), which provides summaries only by state and not individual counties.

Croplands in the counties of concern make up a large proportion of the total agricultural areas of each state (47% for Indiana and 16% for Missouri in 1997), and with one exception, we assume trends in kinds of insecticides applied to crops over the entire state are representative of counties known to include maternity colonies of Indiana bats. The exception is the intensive application of insecticides to cotton, grown only in a few counties in extreme southeastern Missouri. These data are provided separately. We convert data on individual insecticides to metric measures, organize them by class of insecticide and crops, and present them in Appendices II and III to facilitate future comparisons.

Results and Discussion

Organochlorine Insecticides

Organochlorine insecticides are the most intensively studied class of contaminants in bats. These substances became widely used throughout the United States after World War II. They are neurotoxic, synthetic chemicals that are differentially absorbed by lipids, and many of these compounds or their metabolites are resistant to environmental degradation and to metabolism in mammals, leading to accumulation along the food chain. Insects that are not killed outright by OCs can accumulate them; some populations of insects become resistant to these chemicals, and over time, individuals can contain even larger amounts yet still remain available as prey.

Most bats in the United States are insectivorous and have a high demand for food because of the energetic demands of flight and small body size. Over much of the temperate zone, sufficient insects are not always available to meet these requirements, and bats, therefore, have evolved pronounced annual cycles of fat storage and depletion to support them in hibernation or migration, as well as during reproduction. This fat provides depots for accumulation of lipophilic OCs that later are mobilized and released to the general circulation as body fat declines.

Bats, which have long lifespans over which exposure may occur, can accumulate OCs in amounts that cause mortality at critical phases of the life cycle. For example, milk of bats is rich in lipids (Kunz et al. 1995). As females channel energy stored as fat into their milk, most OCs are passed to the young. Bats also maximize food intake and, thus, exposure to dietary contaminants, during the reproductive season. Young bats may die from poisoning by OCs received from the mother before weaning (Clark et al. 1978) or when utilizing their own fat during migration (Geluso et al. 1976). In addition, adults and young-of-the-year may

die when fat is mobilized during hibernation.

Most OCs were removed from general use in the United States by the 1980s, although some are still used in other parts of the world. Studies of some bat populations, such as gray bats (*Myotis grisescens*) in Missouri and, to a lesser extent, Mexican free-tailed bats (*Tadarida brasiliensis mexicana*) in New Mexico, indicate slow recovery from contamination (Clark and Shore 2001). However, OCs continue to be found in bodies of bats, albeit at increasingly lower concentrations. Potential sublethal effects of chronic exposure to low levels of these compounds that could affect population dynamics of bats have not been studied. In addition to low-level contamination from general use, "hotspots" of OC contamination of bats exist, stemming from past industrial manufacture of OCs (Clark et al. 1988, O'Shea et al. 2001) or from intensive, local application of insecticides (Stansley et al. 2001).

The most extensively documented mortality of bats from agricultural OC insecticides of relevance to Indiana bats is poisoning of gray bats in Missouri. Application of the parent compound aldrin in the mid-1970s to control larval moths in corn resulted in food-chain contamination with the metabolite dieldrin. Dieldrin was transferred to young through lactation and reached concentrations in brains that caused mortality of juveniles (Clark et al. 1978). Metabolites of the OC insecticide heptachlor, subsituted for aldrin in local agricultural applications, were also found at lethal concentrations in gray bats in the late 1970s and early 1980s, as was the highly toxic OC endrin (Clark et al. 1983a, 1983b, Clawson and Clark 1989). All these compounds have been banned from agricultural use in Missouri since the 1970s and early 1980s. Impacts of these OCs, particularly dieldrin, on populations of gray bats in Missouri were likely substantial, but recovery apparently has occurred over the past 20 years, in conjunction with declining concentrations of OCs in insects (Clark and Shore 2001, Clawson 1991).

OC pesticides and Indiana bats in the 1970s— Statistical comparisons of concentrations of OCs in tissues or guano of Indiana bats from Indiana and Missouri were limited because of small numbers of samples, especially when samples were subdivided by factors known to contribute substantial variation, such as location, year, season, and sex. Range of concentrations of dieldrin in 11 female and one male Indiana bat found dead in Wyandotte Cave, Indiana, in autumn 1976, varied from undetectable to 3.5 μg/g, whereas no dieldrin was detected in brains (Appendix I). However, concentrations of dieldrin in guano collected from a roost in Harrison Co., Indiana, in 1976, were similar to those in guano from colonies of gray bats in Missouri that suffered mortality from poisoning by dieldrin (z 0.38 μg/g—Clark et al. 1982). We also report lethal concentrations (see Clark et al. 1982 for definitions) of dieldrin in the brain of one male Indiana bat found dead in spring 1976 in Onyx Cave, Missouri (2.6 μg/g), and in the brain of another male found dead in Bat Cave, Missouri (2.4 μg/g; Appendix I). In addition, three bats from Missouri had elevated concentrations of heptachlor epoxide (2.7, 1.6, and 1.2 μg/g) that were sublethal but may have exacerbated the impact of dieldrin (Clark et al. 1980, 1983b).

Concentrations of DDE in Indiana bats and guano from all areas were unremarkable for this era. The highest concentration of DDE in brains of Indiana bats was 0.96 μg/g (Appendix I), whereas minimum lethal concentrations of DDE in brains of little brown bats (*Myotis lucifugus*) were estimated at 540 μg/g (Clark and Stafford 1981). Other OCs detected in Indiana bats, but at trace amounts and not tabulated in this paper, included trans-nonachlor, oxychlordane, components of toxaphene, and polychlorinated biphenyls. Two notable findings were one individual from Onyx Cave with 2.0 μg/g oxychlordane in the brain, and a female from Copper Hollow Sink, Missouri, with 6.5 μg/g *p,p'*-DDT in the carcass. No OCs were detected in guano from two caves in Kentucky.

Concentrations of OCs, indicative of dieldrin mortality in our sample of Indiana bats and guano from Missouri and Indiana, suggested chronic mortality from OC pesticides prior to their discontinuance. Nevertheless, the extent of OC poisoning of Indiana bats during the 1970s possibly was even greater than indicated by our samples. Mortality from poisoning by OCs was most likely to be discovered in bats in late hibernation when energy reserves were depleted, in nursing young metabolizing maternally derived fat, or in adult males that could not eliminate these compounds with reproductive products (milk or fetus). Bats that we examined, however, generally were not in these categories. All of our samples from Indiana were from autumn; most bats from Indiana and Missouri were female (29 of 38), and no suckling young were sampled.

Recent studies related to Indiana bats—One recent study examined two other species of *Myotis*, collected in 1996 and 1997, as surrogates for Indiana bats.

McFarland (1998) determined residues of OCs in carcasses of 27 little brown bats and 15 northern bats (*Myotis septentrionalis*) collected from agricultural areas in northern Missouri in summer, and from 18 little brown bats and 18 northern bats collected at the end of hibernation from caves in southern Missouri. DDE was the OC detected at highest concentrations, typically at less than 1.0 μg/g wet weight of carcass (maximum of 5.1 μg/g), and it was found in nearly all bats. Dieldrin was detected in 90% of bats, also typically at concentrations less than 1.0 μg/g, but with a maximum of 2.7 μg/g. In addition, heptachlor epoxide and oxychlordane were found at lower concentrations and frequencies of detection.

Concentrations of OCs in these recently sampled *Myotis* (McFarland 1998) were generally less than those reported in populations of several species of bat elsewhere in the United States prior to, or just after, restrictions on uses of these compounds (e.g., Clark and Prouty 1976, Reidinger 1976). Data obtained in Missouri in 1996 and 1997 did not suggest potential for lethal exposure when compared with other studies of OCs (Clark and Prouty 1977, 1984, Clark et al. 1978), although knowledge of concentrations on a lipid-weight basis (mass of contaminant/mass of body lipids) were not available to facilitate such comparisons. Dieldrin in carcasses of Indiana bats from Missouri during the 1970s was one-to-two orders of magnitude higher and reached lethal concentrations in brains of some individuals (Appendix I). Heptachlor epoxide also was often an order of magnitude higher in Indiana bats from the 1970s. Concentrations of DDE in carcasses of these recent bats from Missouri, however, encompassed about the same range that we documented for Indiana bats from Missouri 20 years earlier (Appendix I). Schmidt et al. (2002) recently performed additional work on OCs in other species of bat in Missouri.

Sublethal effects—Sublethal effects of OCs have not been studied extensively in bats, although there is a diverse literature on this topic in laboratory mammals. There is, however, evidence that exposure to OCs affects energy metabolism in bats, as shown for shrews fed DDT in the laboratory (Braham and Neal 1974). This effect is intriguing because of the large energy stores required by bats for survival over winter. Sublethal exposure to the insecticide lindane causes increased food intake and 15–23% increases in metabolic rates of European pipistrelles (*Pipistrellus pipistrellus*), and these effects are most pronounced in lean bats under cold conditions (Swanepoel et al. 1999). Little brown bats fed DDT in the laboratory lose weight faster than controls when deprived of food (Clark and Stafford 1981). Exposure to DDE, however, has mixed effects on weight gain in several other species of bat (Clark and Shore 2001). If sublethal exposure to OCs increases energetic requirements in bats, this could have many negative influences on population dynamics. For example, bats would need to compensate by increasing time spent foraging and, thereby, increase susceptibility to predators, and less energy ultimately would be available for reproduction of adults and growth of young. In addition, storage of energy for hibernation or migration could be limited, and expenditure of energy in hibernation could be elevated, resulting in premature depletion of fat and failure to survive winter.

Organophosphates and Carbamates

The major classes of insecticides now in use are organophosphate (OP), carbamate, and pyrethroid compounds. Although highly toxic, OPs and carbamates are not lipophilic, and there is little accumulation in tissues of wildlife. OPs and carbamates act by inhibiting the neurotransmitter cholinesterase (ChE). Bats are similar to other mammals and birds in that these chemicals are quickly metabolized and, if not killed outright, exposed individuals may recover. However, a variety of sublethal effects of OP pesticides occur in other mammals and birds that impact thermoregulation, food consumption, and reproductive behavior (Grue et al. 1997), all of which have implications for bats.

Determination of exposure to carbamate or OP insecticides is based on bioassays of the activity of ChE in brains of potentially affected animals with activity in brains of unexposed individuals. Such techniques, of course, require sacrificing the animals in question, as well as a reference group. ChE activity in brains less than 2 *SD* below activity in controls commonly is accepted as indicative of exposure, and activities less than 50% of controls are diagnostic of OP or carbamate poisoning as the cause of death (Hill and Fleming 1982, Ludke et al. 1975). Quantitative chemical analysis of contents of the gastrointestinal tract is required to identify the compound responsible for inhibition of ChE, if dietary exposure is suspected.

Observations in the laboratory—There were few laboratory experiments involving bats and OPs or

carbamates to facilitate interpretation of field observations. Hurley and Fenton (1980) noted no apparent impacts on little brown bats sprayed with the OP fenthion at 28 mg/kg. Clark (1986) dosed female little brown bats and big brown bats (*Eptesicus fuscus*) with methyl parathion to determine 24-h median lethal doses (LD_{50}) and doses that impaired coordination but were non-lethal. The LD_{50} in little brown bats was 8.5 times greater (less sensitivity) than in laboratory mice. However, mice either died or appeared behaviorally normal 2–3 h after exposure, whereas many bats lost coordination and were unable to right themselves as long as 24 h later. Time to death was also much longer in some bats than in mice. Big brown bats had an inability to right themselves at about one-third the likely LD_{50}. These findings imply that exposure to non-lethal doses would result in death in the field when bats became debilitated and exposed to predators, harsh sun, inclement weather, or drowning (Clark 1986). Activity of ChE in brains of little brown bats that survived oral dosing ranged from 18.5 to 25.6% of controls. ChE activity in little brown bats that survived dermal application of methyl parathion to the wing was similar to activity in those dosed orally. Thus, dermal exposure to sprays in the field also could result in loss of coordination and susceptibility to other sources of mortality (Clark 1986).

The 24-h LD_{50}, ChE activity, and behavioral responses of little brown bats to oral administration of the OP insecticide acephate also were studied in the laboratory (Clark and Rattner 1987). Little brown bats were much less sensitive to acute lethality from acephate than laboratory mice. As with exposure to methyl parathion, all surviving mice were apparently normal in behavior, whereas about one-third of bats were unable to right themselves. ChE activity in brains decreased with increasing dose of acephate, whereas methyl parathion killed bats too rapidly for major differences in ChE to be apparent (Clark 1986, Clark and Rattner 1987). Serum ChE activity in control bats in the study of acephate was several times higher than in mice or birds, perhaps providing bats additional protection against these chemicals (Clark and Shore 2001).

Observations on animals from the wild—A few recent studies attempted identification of individual OPs or carbamates in guano or gastrointestinal tracts of captured bats. Sandel (1999) examined contents of the gastrointestinal tract of 15 individuals and samples of guano from 24 colonies of Mexican free-tailed bats from agricultural areas of Texas for up to 27 carbamates and OPs. None was detected. Clark et al. (1996) analyzed contents of the gastrointestinal tract of three Mexican free-tailed bats from Texas for 24 OP and carbamate compounds but did not detect their presence, even though one individual had depressed ChE in its brain. McFarland (1998) obtained nine samples from gastrointestinal tracts of little brown bats and northern bats; some samples were from bats caught in areas where indications of depressed ChE activity in brains were found previously, and some were from areas where ChE activity was not depressed. Although analytical procedures allowed identification of five OPs (chlorpyrifos, fonofos, methyl parathion, phorate, and terbufos), none was found. Land (2001) collected guano from maternity roosts of cave bats (*Myotis velifer*) in Texas, and 18 organophosphate compounds were detected at high levels of analytical sensitivity (primarily at ng/g quantities), showing that bats were exposed to and excreted these compounds.

Studies of ChE activities in brains of bats taken from the field were also rare but more indicative of exposure to OP or carbamate insecticides. One Mexican free-tailed bat found dead beneath a roost in Texas in 1993 had ChE activity below 2 *SD* and only 27% of the mean of controls, and the bat may have died from exposure to these insecticides (Clark et al. 1996). ChE activity of European pipistrelles in Spain was depressed 11.2% in juveniles and 16.1% in adults 21 days after agricultural fields were sprayed with the OP insecticide fenitrothion, but there were no known harmful effects (Guillen et al. 1991).

A study of greater relevance to Indiana bats was one that analyzed ChE activity in brains of little brown bats and northern bats in 1996. Bats were collected from riparian areas in northern Missouri in summer and from caves in southern Missouri near the end of hibernation, when exposure to insecticide would not be likely (McFarland 1998). No significant differences in mean ChE activity were evident within either species. However, ChE activity in one little brown bat collected in summer was more than 2 *SD* below the mean of the comparison group. ChE activity in several individuals sampled in summer also was low (depressed to at least 30% of the mean of controls, or outside the lower 95% *CI* of controls). This suggested that some individuals were exposed to ChE-inhibiting insecticides in agricultural areas of Missouri in summer. The same samples of bats also were analyzed for density of

muscarinic cholinergic receptors, another bioassay that indicated exposure to ChE-inhibiting insecticides. No significant differences occurred with this bioassay, although a few depressed outliers were noted among bats collected in summer in agricultural regions (McFarland 1998).

There are hundreds of incidents of mortality due to ChE-inhibiting insecticides in other wildlife (Grue et al. 1983). However, only one die-off in bats, involving methyl parathion and free-tailed bats in 1968, tentatively is identified as due to these compounds (Clark 1986). It is likely that lethal poisoning by OPs or carbamates takes place in bats but simply has not been documented. Diagnosis of mortality in bats due to exposure to OP or carbamate insecticides is more difficult than diagnosis of mortality from exposure to OCs. Control groups are necessary for comparison of ChE activity, and confirmation of the presence of these newer compounds in digestive tracts is desirable. Determination of ChE activity in brains can be complicated by postmortem reactivation of ChE, depending on class of insecticide (Hill 1989), and this can hamper the ability to confirm effects of these substances on bats that have died at unknown times prior to collection.

Despite these technical problems and a lack of field examples, results of laboratory experiments suggest possible scenarios. For example, bats in flight may be exposed to aerosols applied in the stillness of evening or early morning (a common practice to avoid drift and killing of honeybees). Exposure during foraging or traveling, therefore, may be pulmonary or dermal, and no residues may be detected in the ingesta. Effects may occur rapidly upon exposure, and bats away from roosts may become uncoordinated and tumble to the ground, making their discovery unlikely. Even if not killed, recovery of normal function may be delayed, and bats might succumb to exposure or predation. Furthermore, bats with lowered ChE activity could experience sublethal effects that contribute to eventual mortality.

Pyrethroids

Pyrethroid insecticides are growing in usage. They are neurotoxins with a mode of action similar to DDT, affecting sodium channels in nervous membranes and resulting in tremors following acute poisoning. These compounds have potential to affect bat populations, particularly those forms that are chlorinated (such as cypermethrin) and likely to persist in the environment (Clark and Shore 2001). There has been, however, little research on the presence of pyrethroids or their effects on bats.

Observations in the laboratory—Median lethal dose (LD_{50}) of permethrin for little brown bats was about 38–45 mg/kg by stomach gavage for a field-grade formulation, which was ca. 25 times more toxic than in laboratory rats (McFarland 1998). Field-grade formulations were designed to increase delivery of active ingredients to target insects through enhanced absorption, and these formulations killed bats more quickly and at lower dosages than analytical-grade permethrin in LD_{50} trials. Ability to fly was impaired in little brown bats at a median effective dose of the analytical grade formulation that was about one-sixth the LD_{50}. Concentrations of permethrin in carcasses of little brown bats dosed in the laboratory (including survivors in LD_{50} trials) were one-to-two orders of magnitude higher than in samples collected in the field in Missouri (McFarland 1998). In England, experimental studies of vespertilionid bats that roosted in structures made from permethrin-treated wood showed no significant mortality, nor was permethrin detectable in tissues (Shore et al. 1991).

Observations on animals from the wild—McFarland (1998) determined concentrations of four pyrethroid insecticides (cyhalothrin, esfenvalerate, permethrin, and tefluthrin) in the same carcasses of bats from Missouri sampled for OCs. Sixteen of the 78 bats had detectable residues of pyrethroids, but 15 of these were collected from hibernacula in southern Missouri, rather than agricultural areas of northern Missouri in summer. Permethrin was detected in little brown bats collected in late hibernation, with concentrations ranging from 0.08 to 0.89 μg/g wet weight of carcass at one cave and reaching a maximum of 2.5 μg/g at another cave. Esfenvalerate was found in two little brown bats at 0.14 and 0.25 μg/g. One northern bat had detectable residues of permethrin (0.05 μg/g), and five had traces of esfenvalerate (0.09 to 0.41 μg/g). All bats with detectable pyrethroid residues were from hibernacula, except one northern bat. These findings suggested that exposure to pyrethroids was from local sources in southern Missouri or that insecticides were acquired prior to migration and retained in the body, with residual amounts detectable at the end of hibernation (McFarland 1998). It was tempting to speculate that these bats foraged in southeastern Missouri, where use of pyrethroids on cotton was extensive, because most

Table 1.—Agricultural statistics for 38 counties in Indiana[a] and 16 counties in Missouri[b] where maternity colonies of Indiana bats occur (U.S. Fish and Wildlife Service 1999). Statistics were compiled from U.S. Department of Agriculture (2001) and U.S. Bureau of the Census (1994).

	Missouri			Indiana		
Category	1987	1992	1997	1987	1992	1997
Area of total cropland (ha x 10^6)	1.23	1.21	1.21	2.58	2.57	2.44
Proportion of area in crops (%)	48.0	47.0	47.2	64.4	64.2	61.0
Area to which insecticides were applied (ha x 10^3)	98.3	93.7	74.5	466.9	432.6	408.4
Percent of total area to which insecticides were applied (%)[c]	3.8	3.6	2.9	11.7	10.8	10.2
Area of major crops (ha x 10^3)						
Soybeans	360 (29.3)[d]	302 (24.9)	317 (26.2)	886 (34.3)	951 (37.0)	992 (40.7)
Alfalfa/Hay	187 (15.2)	200 (16.5)	208 (17.2)	289 (11.2)	231 (9.0)	94 (3.9)
Corn	165 (13.4)	186 (15.4)	181 (15.0)	967 (37.5)	1,174 (45.7)	1,091 (44.7)
Wheat	27 (2.2)	62 (5.1)	44 (3.6)	101 (3.9)	91 (3.5)	84 (3.4)
Sorghum	20 (1.6)	24 (2.0)	11 (0.9)	NR	NR	NR
Oats	NR[e]	NR	NR	13 (0.5)	9 (0.4)	5 (0.2)

[a] Counties in Indiana are Blackford, Boone, Clinton, Delaware, Fountain, Fulton, Hendricks, Henry, Howard, Huntington, Jasper, Jay, Jefferson, Jennings, Knox, Kosciusko, Lagrange, La Porte, Marion, Martin, Montgomery, Parke, Pulaski, Putnam, Randolph, Ripley, Rush, St. Joseph, Shelby, Starke, Steuben, Tippecanoe, Vermillion, Vigo, Wabash, Warren, Wayne, and Wells.

[b] Counties in Missouri are Chariton, Clinton, Daviess, Iron, Jefferson, Knox, Linn, Macon, Mercer, Monroe, Nodaway, Pike, Pulaski, Scotland, Sullivan, and Washington.

[c] Percentage of total area in county, whether it is cropland or not.

[d] Percentage of total cropland in parentheses.

[e] NR = not reported as a major crop.

use of esfenvalerate (60%) in Missouri in 1997 was on cotton (Appendix II). However, esfenvalerate also was used elsewhere on corn, and permethrin was not used on cotton.

In addition to the study in Missouri, guano from Mexican free-tailed bats from Texas was analyzed for cypermethrin, fenvalerate, and *cis*-and *trans*-permethrin (Sandel 1999). Two pyrethroids were found in guano from colonies in seven counties; *cis*-permethrin ranged from 0.02 to 0.10 μg/g, and *trans*-permethrin was 0.02–0.08 μg/g. Although this finding indicated dietary exposure, pyrethroids were not found in six carcasses or three milk samples of Mexican free-tailed bats that also were analyzed (Sandel 1999).

Use of Insecticides in Missouri and Indiana

Although hibernating populations of Indiana bats in Missouri are declining and those in Indiana are increasing, there were no drastic differences in crops planted or general usage of agricultural insecticides between states. Amount and proportions of croplands and amounts and proportions of areas treated with insecticides were consistently higher in 38 counties in Indiana with presumed maternity colonies of Indiana bats than in 16 corresponding counties in Missouri (Table 1). Corn received the greatest treatment with insecticides of all crops grown in either state (Appendix III). In the three censuses from 1987–1997, corn grown in counties known to include maternity colonies of Indiana bats in Missouri averaged 13.4–15.4% of total area of cropland in the state. This percentage was up to threefold higher for the counties in Indiana (Table 1). In terms of area, soybeans were the most important crop in Missouri and second most important in Indiana (Table 1), but this crop received low amounts of insecticides in both states (Appendix III). Alfalfa ranked second in Missouri and third in Indiana, and it received the second greatest treatment with insecticides in both states. Greater applications of insecticides to alfalfa occurred in 1992 compared with 1997 in both states.

Noteworthy differences emerged in use of specific

insecticides and insecticide classes on a statewide basis (Appendices II–III). Chief among these were greater use of pyrethroids in Missouri and a major increase in the proportion of area of corn treated with pyrethroids in Missouri from 1992 to 1997. Permethrin use in Indiana remained at 2% of the area planted to corn between the two periods, but increased from 9 to 25% in Missouri; λ-cyhalothrin also increased from 0 to 10% in Missouri, and there was a general increase in use of pyrethroids on all crops. In Missouri, amount of permethrin used on all crops more than doubled, from 14,829 to 29,834 kg, but use of permethrin decreased from 11,986 to 8,265 kg in Indiana.

Use of other pyrethroids was also greater in Missouri than in Indiana and increased between the two censuses. Missouri used 5,449 kg of other pyrethroids in 1992 and Indiana used 533 kg. Much of this was applied to cotton, although permethrin was not used on cotton. In 1992, eight pyrethroid compounds were used in Missouri, and just two in Indiana; four compounds were used exclusively on cotton. This pattern occurred again in 1997. Use of pyrethroids increased in Indiana, as well as in Missouri, from two compounds totaling 12,519 kg in 1992 to six compounds totaling 26,802 kg in 1997 (Appendix II). Much of this was due to new applications of tefluthrin to corn in 1997 (Appendix III).

The proportion of corn treated with individual carbamate or OP insecticides was roughly equivalent for Indiana and Missouri in 1992, except greater use of terbufos in Indiana. In 1997, greater proportions of corn were treated with OPs in Indiana, particularly tebupirimphos and terbufos (Appendix III). Methyl parathion was used exclusively in Missouri in 1997. Use of carbamates and OPs on alfalfa was generally much higher in Missouri than in Indiana in 1992 but not in 1997, when use of these compounds on alfalfa had declined and were comparable in both states. Application of pyrethroids to alfalfa did not occur in Missouri in 1992 but was evident in 1997, as was an increase in use of pyrethroids on alfalfa in Indiana between the two censuses. Use of OPs and carbamates on sorghum in Missouri also declined substantially between the censuses, whereas pyrethroid applications on sorghum became evident in 1997.

Other Contaminants and Bats

Occurrence of polychlorinated biphenyls (PCBs) was documented in six species of bat in the United States (Clark and Shore 2001). These compounds were used in a variety of commercial and industrial applications, but their manufacture was discontinued in the United States in 1977 because of their persistence, lipophilicity, and recognition as widespread contaminants in certain foods, wildlife, and humans. They tended to be found in highest concentrations in bats near urban or industrial areas (Clark and Shore 2001). A limited amount of information was available on uptake and loss of dietary PCBs from laboratory studies of big brown and little brown bats (summarized in Clark and Shore 2001). Past research quantified PCBs in relation to commercial mixtures, but recent studies of bats reported concentrations of major congeners. Other than recent work in Missouri (A. Schmidt *pers. comm.*), these studies of individual PCB congeners were based on European bats (Clark and Shore 2001).

Although PCBs caused reproductive failure in several species of mammals (Shore and Rattner 2001), effects on reproduction in pregnant little brown bats and big brown bats experimentally fed PCB mixtures were not apparent (Clark 1978, Clark and Krynitsky 1978, Clark and Lamont 1976a, 1976b). Female bats transfer a large proportion of their PCBs to offspring through lactation and, secondarily, through gestation, and these chemicals could affect the young. Several PCBs also were documented as endocrine disrupters, but evidence for such effects has not been sought in bats (Clark and Shore 2001). PCBs have not been implicated in mortality of bats.

Toxic elements, such as metals, were determined in surveys of ca. 10 species of bat or their guano in the United States (Clark and Shore 2001). Although bats could be exposed to and accumulate toxic elements (Clark 1979, Clark et al. 1986, O'Shea et al. 2001), there were no experimental studies of potential effects of toxic elements in bats and no strong field observations linking contamination with effects on bat populations (Clark and Shore 2001). One study suggested a possible link between elevated concentrations of lead and stillbirths in big brown bats and little brown bats in suburban Maryland (Clark 1979). Another report postulated a relationship between mild kidney lesions in gray bats in northern Alabama and high concentrations of cadmium in their guano (Clark et al. 1988). There were no published studies of radionuclide contamination in bats.

Bats die by asphyxiation after drinking solutions containing cyanide from open holding ponds of gold-mining operations that employ cyanide-extraction methods (Clark and Hothem 1991). Death occurs

rapidly upon exposure, with sublethal doses quickly detoxified and eliminated. Cyanide is not persistent and does not biomagnify. However, the extent of the problem may be underestimated. Laboratory experiments in which little brown bats receive sodium cyanide show that death in some bats occurs several hours after exposure, when they potentially are far from the source of poisoning (Clark et al. 1991). Although most attention is directed to large mining operations in the West, cyanide-extraction methods also are used in the southeastern United States. Straightforward management measures include covering ponds, excluding bats, or eliminating cyanide from exposed water. Finally, mortality of bats also occurs from oil pollution when individuals attempting to drink become mired in oil-covered ponds and sludge pits (Finley et al. 1983).

Conclusions and Suggestions for Research

Several points are evident from comparison of agricultural production and use of insecticides. Patterns at the scale investigated do not suggest major differences in insecticide use on summer ranges in Missouri and Indiana that might be associated with declines in populations of Indiana bats at hibernacula in Missouri. Application of pyrethroids, however, has been greater in Missouri and is increasing in both states, and these compounds recently were found in tissues or guano of bats in Missouri and Texas. The possible persistence of some chlorinated pyrethroids (Clark and Shore 2001), their newly discovered presence in tissues of bats, and their increasing use in agriculture in the United States calls for more detailed surveys of their occurrence and experiments on their effects in bats.

The most intensive use of insecticides in Missouri is in the southeast, the only area in either state where cotton is grown. This limited area accounts for all or much use of at least two carbamates (oxamyl and thiodicarb), six OPs (dicrotophos, dimethoate, disulfoton, malathion, methyl parathion, and profenofos), and five pyrethroids (bifenthrin, cyfluthrin, cypermethrin, esfenvalerate, and λ-cyhalothrin), depending on year. Counties with Priority One hibernacula (U.S. Fish and Wildlife Service 1999) are ca. 100 km from cotton-growing areas in Missouri, and it is conceivable that some autumn or spring foraging by Indiana bats occurs in this region. Determination of the amount of foraging by Indiana bats in the cotton-growing region of southeastern Missouri would help understand the likely maximum degree of exposure to insecticides endured by this species.

There is a need to increase passive collection of data and samples of Indiana bats for analysis of contaminants or their effects, and this can be done incidental to other projects. Ecological studies of bats at maternity colonies should include provisions to collect guano for chemical analysis, particularly in agricultural areas. For detection of modern insecticides, guano should be as fresh as possible and preferably stored frozen, ideally at very cold temperatures (-60°C—Clark and Shore 2001). Insects at known foraging areas also should be collected and sampled for contaminants.

Brains of bats found dead and frozen prior to significant decomposition are useful for determination of diagnostic concentrations of OCs and activity of ChE, and public health agencies are a potential source of this material from specimens submitted for rabies determination. Relationships should be developed with these authorities to ensure that submissions are properly identified to species, and carcasses of Indiana bats fully utilized for analysis of contaminants. Diagnosis of exposure and possible poisoning of bats by both OC and ChE-inhibiting insecticides recently were made through such cooperation (Clark et al. 1996, Stansley et al. 2001). In addition, studies of small samples of skin from museum specimens (Clark 2001) of Indiana bats and surrogate species within their range may provide useful indications of past contamination by OCs.

Diagnosis of mortality in birds and mammals due to OC poisoning is based on threshold concentrations of these compounds in brains, and relationships between concentrations in brains and in lipids or guano have been established for some species of bat (Clark et al. 1982). This allows inferences about the likelihood of mortality when brains of bats found dead are not available. Diagnostic, lethal concentrations of OCs in brains of bats are tabulated in Clark (1988) and Clark and Shore (2001). Concentrations of OCs that are associated with population declines, die-offs, or documented OC-induced mortality in bats of the United States also are available. In addition, concentrations based on lipid content of carcasses, which are predictive of diagnostic residue concentrations in brains, appear in the literature (see Clark 1981, 1988, Clark and Shore 2001). These data provide benchmarks for assessing the significance of some OC residues in guano or carcasses of Indiana bats found dead and submitted for analysis.

More detailed tabulations and analysis of patterns of insecticide use throughout the range of the Indiana

bat would help guide future research. Records of contaminant-related die-offs of other wildlife, particularly insectivorous or aquatic birds, should be assembled to determine if overlap occurs with areas of high use or declines in Indiana bats. Additionally, "hotspots" of contamination by all classes of potentially harmful contaminants within the range of Indiana bats should be identified, and follow-up surveys undertaken to determine if these areas are used by Indiana bats and to estimate their likely degree of exposure.

The most reliable approach for determining effects and occurrence of contaminants in Indiana bats, however, is to carry out well-designed feeding experiments in the laboratory and to follow rigorous sampling designs for collecting specimens in the field for surveys of contaminant residues and biomarkers of exposure. This is particularly necessary for the OP, carbamate, and pyrethroid insecticides, because deaths may occur away from roosts, making fresh carcasses difficult to discover, and because there is so little information available on the presence and effects of these compounds on bats. Determining the impact of sublethal exposure to OCs and OPs on metabolic rates and thermoregulation also is important, when one considers the critical importance of efficient energy metabolism for winter survival and effects of temperature changes in hibernacula in Missouri (Tuttle and Kennedy 2002).

Performing such studies on other species used as surrogates has merit, particularly as a first step. However, differences among species in sensitivity to contaminants or bioassays, such as inhibition of ChE (Hill 1988), make use of surrogates less than definitive. Numbers of hibernating Indiana bats in Missouri have dropped by perhaps 269,000 in 20 years (over 13,000 per year—Clawson 2002). The added mortality from bats sacrificed for well-designed studies of contaminants would be trivial in comparison, particularly if results provide greater insights for management.

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Appendix I

Concentrations of principal organochlorine insecticides or metabolites found in carcasses, brains, and guano of adult Indiana bats (Myotis sodalis), 1975-1978

Original data are presented to facilitate comparisons with future samples and other studies. Concentrations are in μg/g wet weight (carcasses and brains) or dry weight (guano). Samples represent bats that were found dead at roosts or died accidentally during handling. ND = not detected.

Sample	Origin	Date of collection	Sex	Type of sample	Lipid (%)	Dieldrin	*p,p'*-DDE	Heptachlor epoxide
Indiana								
Crawford Co. (Wyandotte Cave)								
76B362/363	Found dead	29 October 1976	Female	Carcass	29.8	0.13	0.78	1.3
				Brain	—	ND	ND	ND
76B360/361	Found dead	29 October 1976	Female	Carcass	31.3	ND	0.59	ND
				Brain	—	ND	0.85	ND
76B358/359	Found dead	29 October 1976	Female	Carcass	31.0	0.86	0.37	0.41
				Brain	—	ND	0.53	ND
76B356/357	Found dead	29 October 1976	Female	Carcass	32.5	2.9	1.0	0.58
				Brain	—	ND	ND	ND
76B354/355	Found dead	29 October 1976	Female	Carcass	2.2	1.2	1.4	0.66
				Brain	—	ND	ND	ND
76B352/353	Found dead	29 October 1976	Female	Carcass	37.5	1.5	0.77	0.43
				Brain	—	ND	ND	ND
76B350/351	Found dead	29 October 1976	Female	Carcass	13.9	2.1	0.80	0.95
				Brain	—	ND	1.2	ND
76B370/371	Found dead	29 October 1976	Female	Carcass	32.8	0.96	0.59	0.41
				Brain	—	ND	1.7	ND
76B368/369	Found dead	29 October 1976	Female	Carcass	29.0	0.8	1.0	1.2
				Brain	—	ND	ND	ND
76B366/367	Found dead	29 October 1976	Male	Carcass	30.4	2.6	1.2	1.3
				Brain	—	ND	ND	ND
76B364/365	Found dead	29 October 1976	Female	Carcass	31.4	3.5	0.66	1.1
				Brain	—	ND	ND	ND
76B372/373	Found dead	29 October 1976	Female	Carcass	28.9	ND	0.4	ND
				Brain	—	ND	ND	ND
Harrison Co.								
77B223	—	1976	—	Guano	—	0.37	0.18	ND
77B224	—	1976	—	Guano	—	0.39	0.17	ND
Wayne Co.								
76B7	—	19 October 1975	—	Guano	—	0.22	0.15	ND
Kentucky								
Edmonson Co. (Dixon Cave)								
77B221	—	1976	—	Guano	—	ND	ND	ND
Edmonson Co. (Long's Cave)								
77B218	—	1976	—	Guano	—	ND	ND	ND
77B219	—	1976	—	Guano	—	ND	ND	ND
77B220	—	1976	—	Guano	—	ND	ND	ND
Missouri								
Franklin Co. (Bear Cave)								
76B32/33	Unknown	13 October 1975	Female	Carcass	39.9	5.9	0.76	1.5
				Brain	—	ND	ND	ND
Franklin Co. (Copper Hollow Sink)								
76B40/41	Unknown	6 October 1975	Female	Carcass	31.3	1.9	0.15	1.3
				Brain	—	ND	0.49	ND
76B42/43	Unknown	6 October 1975	Female	Carcass	37.5	3.8	1.7	0.93
				Brain	—	ND	ND	ND

Appendix I.—Continued

Sample	Origin	Date of collection	Sex	Type of sample	Lipid (%)	Dieldrin	*p,p'*-DDE	Heptachlor epoxide
Missouri								
Iron Co. (Pilot Knob Mine)								
76B330/331	Accidental	12 October 1976	Male	Carcass	7.3	0.51	1.7	0.35
				Brain	—	ND	0.53	ND
76B334/335	Accidental	12 October 1976	Female	Carcass	36.8	2.3	0.70	5.5
				Brain	—	ND	ND	ND
79B21/22	Accidental	28 April 1977	Male	Carcass	0.0	0.40	1.6	0.45
				Brain	—	ND	0.94	ND
79B19/20	Accidental	19 June 1977	Male	Carcass	0.0	0.75	1.0	0.75
				Brain	—	ND	ND	ND
Pulaski Co. (Onyx Cave)								
76B34/35	Accidental	8 October 1975	Male	Carcass	14.2	5.5	1.3	2.1
				Brain	—	ND	ND	ND
76B49/50	Found dead	28 April 1976	Male	Carcass	1.9	8.7	2.0	13.0
				Brain	—	2.6	ND	2.7
76B318/319	Unknown	9 March 1976	Female	Carcass	14.0	4.2	0.47	2.7
				Brain	—	0.92	ND	ND
78B41/42	Found dead	28 April 1976	Male	Carcass	7.1	7.6	1.3	3.4
				Brain	—	0.77	ND	0.46
79B5/6	Unknown	21 Feb 1978	Female	Carcass	22.1	1.8	0.59	1.9
				Brain	—	ND	ND	ND
79B7/8	Found dead	21 Feb 1978	Female	Carcass	17.3	1.1	0.12	1.6
				Brain	—	ND	ND	ND
79B9/10	Found dead	21 Feb 1978	Female	Carcass	14.1	0.66	ND	ND
				Brain	—	ND	ND	ND
Shannon Co. (Bat Cave)								
79B1/2	Found dead	8 April 1977	Male	Carcass	4.2	5.7	1.1	5.4
				Brain	—	2.4	ND	1.6
79B3/4	Found dead	8 April 1977	Female	Carcass	1.3	1.9	0.80	1.7
				Brain	—	ND	ND	ND
Washington Co. (Great Scott Cave)								
76B36/37	Found dead	14 October 1975	Female	Carcass	40.8	2.0	0.69	1.7
				Brain	—	ND	ND	ND
76B38/39	Found dead	14 October 1975	Female	Carcass	40.2	2.7	0.21	2.1
				Brain	—	ND	ND	ND
76B30/31	Found dead	20 October 1975	Female	Carcass	35.6	1.5	0.25	0.64
				Brain	—	ND	ND	ND
76B320/321	Accidental	10 April 1976	Female	Carcass	12.5	3.5	0.51	1.9
				Brain	—	ND	ND	ND
76B324/325	Accidental	22 Sep 1976	Female	Carcass	17.6	3.0	0.54	4.4
				Brain	—	ND	ND	ND
76B326/327	Accidental	22 Sep 1976	Female	Carcass	32.4	0.94	0.73	1.3
				Brain	—	ND	ND	ND
79B11/12	Found dead	24 Feb 1978	Female	Carcass	21.3	0.93	0.29	0.61
				Brain	—	ND	ND	ND
79B13/14	Found dead	24 Feb 1978	Female	Carcass	0.9	0.09	ND	ND
				Brain	—	ND	ND	ND
79B15/16	Found dead	24 Feb 1978	Male	Carcass	10.6	2.3	0.93	1.7
				Brain	—	ND	ND	ND
79B17/18	Found dead	24 Feb 1978	Male	Carcass	0.0	0.10	0.39	0.23
				Brain	—	ND	ND	ND
76B3	—	28 April 1976	—	Guano	—	ND	ND	ND
76B4	—	28 April 1976	—	Guano	—	ND	ND	ND
County not specified (Hamilton Cave)								
78B39/40	Found injured	20 November 1976	Female	Carcass	8.7	1.3	0.87	0.99
				Brain	—	ND	ND	ND

Appendix II

Amounts (kg) of active ingredients of insecticides applied to all crops combined, including fruits and vegetables, for Missouri and Indiana, in 1992 and 1997

Cotton is grown in extreme southeastern Missouri and contributes heavily to statewide usage patterns for several insecticides. Amounts applied to cotton are indicated separately but also included in Missouri totals. Comparisons should be limited to gross patterns of usage (amount applied) between states and years within states. Individual insecticides vary in toxicity, resistance to degradation, and potential for bioconcentration such that simple comparison of amounts applied among chemicals does not allow evaluation of potential threats to Indiana bats. Data are from the National Center for Food and Agricultural Policy (Gianessi and Marcelli 2000). Blank cells (dashes) indicate that no usage was reported.

	1992			1997		
	Missouri		Indiana	Missouri		Indiana
Insecticide	All crops	Cotton	All crops	All crops	Cotton	All crops
Carbamates						
Carbaryl	63,220	—	20,770	98,989	—	15,195
Carbofuran	88,859	—	66,282	6,223	—	2,834
Formetanate HCl	44	—	1,051	11	—	146
Methomyl	8,083	—	5,029	267	—	232
Oxamyl	1,486	1,486	715	2,694	2,694	1,186
Thiodicarb	3,343	3,343	—	4,138	4,138	—
Trimethacarb	—	—	27,406	—	—	
Organophosphates						
Acephate	2,734	996	2,829	10,250	7,572	4,066
Azinphosmethyl	2,942	1,337	2,155	4,820	2,113	1,442
Chlorethoxyfos	—	—	—	3,701	—	24,250
Chlorpyrifos	148,628	14,858	180,097	128,835	—	160,768
Diazinon	6,319	—	1,899	472	—	555
Dicrotophos	29,717	29,717	—	12,361	12,361	—
Dimethoate	8,870	8,826	5,646	6,289	4,226	2,917
Disulfoton	14,332	14,264	—	11,094	11,094	—
Ethoprop	16	—	447	—	—	—
Ethyl parathion	1,087	—	—	—	—	—
Fonofos	16	—	91,556	—	—	—
Malathion	15,775	—	6,058	27,313	26,414	3,996
Methamidophos	24	—	1,433	553	—	187
Methidathion	31	—	—	—	—	—
Methyl parathion	22,791	—	7,878	109,947	83,204	7,001
Mevinphos	2	—	—	—	—	—
Oxydemeton-methyl	—	—	3,793	—	—	—
Phorate	15,160	—	57,031	25,825	14,721	28,939
Phosmet	147	—	1,495	3,099	—	5,207
Profenofos	1,114	1,114	—	—	—	—
Tebupirimphos	—	—	—	2,082	—	26,271
Terbufos	42,915	—	254,706	10,440	—	252,809
Pyrethroids						
Bifenthrin	918	918	—	—	—	—
Cyfluthrin	594	594	—	1,268	1,268	1,290
Cypermethrin	1,040	1,040	—	1,320	1,320	3
Esfenvalerate	738	297	533	3,112	1,902	164
Fenvalerate	32	—	—	—	—	—
λ-cyhalothrin	891	891	—	7,574	5,177	913
Permethrin	14,829	—	11,986	29,834	—	8,265
Tefluthrin	1,236	—	—	578	—	16,167

Appendix III

Proportion of area of major crops treated with individual insecticides and amount of active ingredient applied in Missouri and Indiana, in 1992 and 1997

Summaries are for dominant crops (Table 1) in counties presumed to include maternity colonies of Indiana bats. Comparisons should be limited to gross patterns of usage between states (percentage of area) and years within states (amount applied). Individual insecticides vary in toxicity, resistance to degradation, and potential for use such that simple comparison of amounts applied among chemicals does not allow evaluation of potential threats to Indiana bats. Data are from the National Center for Food and Agricultural Policy (Gianessi and Marcelli 2000). Blank cells (dashes) indicate that no usage was reported.

	1992				1997			
	Missouri		Indiana		Missouri		Indiana	
Crop/Insecticide	Percentage of area (%)	Amount of insecticide applied (kg)	Percentage of area (%)	Amount of insecticide applied (kg)	Percentage of area (%)	Amount of insecticide applied (kg)	Percentage of area (%)	Amount of insecticide applied (kg)
Corn								
Carbamates								
Carbaryl	2	33,703	—	—	—	—	—	—
Carbofuran	3	30,332	2	60,294	—	—	—	—
Methomyl	1	5,617	—	—	—	—	—	—
Trimethacarb	—	—	1	27,407	—	—	—	—
Organophosphates								
Chlorethoxyfos	—	—	—	—	2	3,701	6	24,250
Chlorpyrifos	7	91,223	6	171,017	7	123,882	6	159,141
Fonofos	—	—	3	91,264	—	—	—	—
Methyl parathion	—	—	—	—	4	23,134	—	—
Phorate	1	11,234	2	55,909	—	—	1	28,039
Tebupirimphos	—	—	—	—	2	2,082	8	26,271
Terbufos	3	40,106	8	254,332	1	10,410	8	252,605
Pyrethroids								
Cyfluthrin	—	—	—	—	—	—	8	1,213
λ-cyhalothrin	—	—	—	—	10	2,313	2	505
Esfenvalerate	1	337	—	—	1	1,041	—	—
Permethrin	9	14,155	2	9,866	25	28,918	2	6,568
Tefluthrin	1	11,234	3	9,044	1	578	8	16,166
Sorghum								
Carbamates								
Carbaryl	5	16,308	—	—	1	2,169	—	—
Carbofuran	4	10,959	—	—	1	1,446	—	—
Methomyl	1	1,631	—	—	—	—	—	—
Organophosphates								
Chlorpyrifos	2	4,631	—	—	—	—	—	—
Diazinon	2	3,262	—	—	—	—	—	—
Malathion	3	9,785	—	—	—	—	—	—
Phorate	1	3,653	—	—	—	—	—	—
Terbufos	1	2,609	—	—	—	—	—	—
Pyrethroids								
Permethrin	—	—	—	—	1	144	—	—
Soybeans								
Organophosphates								
Chlorpyrifos	2	19,252	—	—	—	—	—	—
Alfalfa								
Carbamates								
Carbaryl	1	2,265	3	4,485	—	—	—	—
Carbofuran	20	36,240	5	3,737	2	1,567	2	1,383
Organophosphates								
Chlorpyrifos	12	16,580	5	3,737	—	—	—	—

Appendix III. — Continued

Crop/Insecticide	1992 Missouri Percentage of area (%)	1992 Missouri Amount of insecticide applied (kg)	1992 Indiana Percentage of area (%)	1992 Indiana Amount of insecticide applied (kg)	1997 Missouri Percentage of area (%)	1997 Missouri Amount of insecticide applied (kg)	1997 Indiana Percentage of area (%)	1997 Indiana Amount of insecticide applied (kg)
Alfalfa								
Organophosphates—continued								
Diazinon	1	2,265	—	—	—	—	—	—
Dimethoate	—	—	7	3,453	2	1,474	2	1,474
Ethyl parathion	2	1,087	—	—	—	—	—	—
Malathion	2	5,889	2	4,485	—	—	1	948
Methyl parathion	4	4,530	—	—	3	1,175	3	4,512
Phosmet	—	—	1	1,495	—	—	2	2,105
Pyrethroids								
Cyfluthrin	—	—	—	—	—	—	1	1,504
λ-cyhalothrin	—	—	—	—	1	1,703	1	45
Permethrin	—	—	3	448	1	1,703	1	255
Wheat								
Carbamates								
Carbaryl	1	6,972	—	—	3	14,347	—	—
Methyl parathion	6	17,613	—	—	1	2,391	—	—
Other Hay								
Carbamates	—	—	—	—	—	—	—	—
Carbaryl	—	—	—	—	4	77,401	4	8,075