Biological Report 85(1.9)
August 1986

Contaminant Hazard Reviews
Report No. 9

DIAZINON HAZARDS TO FISH, WILDLIFE, AND INVERTEBRATES: A SYNOPTIC REVIEW

by
Ronald Eisler

Patuxent Wildlife Research Center
U.S. Fish and Wildlife Service
Laurel, MD 20708

SUMMARY

Diazinon (phosphorothioic acid 0,0- diethyl 0- (6-methyl-2-(1-methylethyl)-4-pyrimidinyl) ester) is an organophosphorus compound with an anticholinesterase mode of action. It is used extensively to control flies, lice, insect pests of ornamental plants and food crops, as well as nematodes and soil insects in lawns and croplands. Diazinon degrades rapidly in the environment, with half-time persistence usually less than 14 days. But under conditions of low temperature, low moisture, high alkalinity, and lack of suitable microbial degraders, diazinon may remain biologically active in soils for 6 months or longer.

At recommended treatment levels, diazinon-related kills have been noted for songbirds, honeybees, and especially waterfowl that consume diazinon-treated grass; however, incidents involving agricultural applications may be underreported. Accidental deaths through misapplication of diazinon have also been recorded in domestic poultry, monkeys, and humans. It has been suggested, but not yet verified, that wildlife partially disabled in the field as a result of diazinon poisoning would be more likely to die of exposure, predation, starvation, or dehydration, or face behaviorial modifications, learning impairments, and reproductive declines than would similarly treated domestic or laboratory animals.

Among sensitive aquatic organisms, LC-50 (96 h) values of 1.2 to 2.0 ug/l were derived for freshwater cladocerans, and 4.1 to 5.9 ug/l for marine shrimps; freshwater teleosts were comparatively resistant, with all LC-50 (96 h) values greater than 90 ug/l. Sublethal effects were recorded at 0.3 to 3.2 ug diazinon/l and included reduced emergence of stream insects (0.3 ug/1), reduced fecundity of a marine fish (0.47 ug/1), significant accumulations in freshwater teleosts (0.55 ug/1), daphnid immobilization (1.0 ug/1), potential mutagenicity in a freshwater fish (1.6 ug/1), and spinal deformities in teleosts (3.2 ug/1). Exposure to diazinon during spawning caused temporary, but complete, inhibition of reproduction at concentrations which did not produce this effect in fish exposed continuously since hatch.

Acute oral LD-50's of about 2,500 to 3,500 ug diazinon/kg body weight were determined for goslings *(Anser* spp.), ducks *(Anas* spp.), domestic turkey (*Meleagris gallopavo)*, and the red-winged blackbird (*Agelaius phoeniceus*), the most sensitive birds tested. A dietary LD-50 of 167,000 ug diazinon/kg was determined for Japanese quail (*Corturnix japonica)*. Diazinon produced marked teratogenic effects in embryos of the domestic chicken (*Gallus gallus*) at 6.2 to 25 ug/embryo, reduced egg deposition in the ring-necked pheasant (*Phasianus colchicus*) at more than 1,050 ug/bird, and (empirically) decreased food consumption and increased weight loss in the northern bobwhite (*Colinus virginianus*) at greater than 17,500 ug diazinon/kg diet.

The rat (*Rattus rattus*) was the most sensitive mammal tested in acute oral toxicity screenings, with an LD-50 of 224,000 ug diazinon/kg body weight. Chronic oral toxicity tests with swine (*Sus scrofa*) indicated that death was probable if daily intakes were greater than 5,000 ug diazinon/kg body weight. Measurable adverse effects of diazinon have been recorded in rodents, the most sensitive mammalian group tested, at: 500 ug/kg in diets fed to rats for 5 weeks, causing blood cholinesterase inhibition; 180 ug/kg body weight administered daily to pregnant mice (*Mus musculus*) during gestation, inducing behaviorial modifications and delayed sexual maturity of progeny; and single oral doses of 1,800 and 2,300 ug/kg body weight in rats and white-footed mice (*Peromyscus leucopus*), respectively, which produced altered blood chemistry and brain cholinesterase inhibition.

For protection of sensitive aquatic organisms, diazinon concentrations in water should not exceed 0.08 ug/l; however, more data are needed on effects of fluctuating and intermittent chronic exposures of diazinon on reproduction of fish and aquatic invertebrates. Granular formulations of diazinon seem to be especially hazardous to seed-eating birds, suggesting a need to control or eliminate granular applications when these species are present. For additional protection of birds, diazinon should be used with extreme caution in areas where waterfowl feed, and in large-scale spray applications such as grasshopper control. Diazinon in combination with some agricultural chemicals produced more-than-additive adverse effects on bird growth and fecundity; accordingly, more research is needed on effects of complex mixtures of pesticides that contain diazinon. Most investigators agreed that mammals were less susceptible to diazinon than were birds, at least under controlled environmental regimens. Data are lacking on diazinon impacts to mammals under field conditions; acquisition of these data should constitute a priority research area.

DISCLAIMER

Mention of trade names or commercial products does not constitute endorsement or recommendation for use by the Division of Biological Services, Research and Development, Fish and Wildlife Service, U.S. Department of the Interior.

This report should be cited as:

Eisler, R. 1986. Diazinon hazards to fish, wildlife, and invertebrates: a synoptic review. U.S. Fish and Wildlife Service Biological Report 85(1.9).

TABLES

FIGURES

ACKNOWLEDGMENTS

I thank Lynda J. Garrett, Joyce E. Haber, and Brenda Graff for library services; Irene E. Moore for secretarial help; Joseph B. Hunn, Matthew C. Perry, Kenneth L. Stromborg, and Donald H. White for reviewing the manuscript; and Paul A. Opler and James R. Zuboy for editorial services.

INTRODUCTION

Diazinon, an organophosphorus compound with an anticholinesterase mode of action, was released for experimental evaluation in the early 1950's. Today, diazinon is used extensively by commercial and home applicators in a variety of formulations to control flies, cockroaches, lice on sheep, insect pests of ornamental plants and food crops (especially corn, rice, onions, and sweet potatoes), forage crops such as alfalfa, and nematodes and soil insects in turf, lawns, and croplands (Anon. 1972; Meier et al. 1976; Allison and Hermanutz 1977; Berg 1984; Stone and Gradoni 1985).

Waterfowl and other wildlife may acquire diazinon by drinking contaminated water, by absorbing it through legs and feet, by consuming treated grass or grain, or by ingestion of pesticide-impregnated carrier particles (Stone and Knoch 1982; Stone and Gradoni 1985). Diazinon poisonings of birds--involving 54 incidents in 17 States--have been recorded for at least 23 species, especially among waterfowl feeding on recently treated turfgrass; incidents involving agricultural applications may be less conspicuous, and thus not as well-documented (Stone and Gradoni 1985). Kills of Canada geese (*Branta canadensis*), brant (*Branta bernicla*), mallard (*Anas platyrhynchos*), American black duck (*Anas rubripes*), other species of waterfowl, and songbirds have all been associated with consumption of grass or grain shortly after diazinon application (Schobert 1974; Zinkl et al. 1978; Stone 1980; Stone and Knoch 1982). Fatal diazinon poisonings have also been recorded in humans (Soliman et al. 1982; Lox 1983), domestic chickens (*Gallus gallus*) (Sokkar et al. 1975), domestic ducklings (*Anas* spp.) and goslings (*Anser* spp.) (Egyed et al. 1974, 1976), in laboratory monkey colonies of the tamarin (*Saguinus fuscicollis*) and the common marmoset (*Callithrix jacchus*) (Brack and Rothe 1982), and the honeybee (*Apis mellifera*) (Anderson and Glowa 1984). Mammals seem to be less sensitive than birds to diazinon poisoning (Stone and Gradoni 1985). The lack of reported mammalian mortalities (only one suspected case of a pocket gopher, *Thomomys* sp., found dead in a park at Yakima, Washington, following aerial spraying of diazinon on shade trees) is consistent with the general findings of Grue et al. (1983) for organophosphorus insecticides. Sublethal effects such as reduced hatch, retarded growth, and spinal deformities in fish (Allison and Hermanutz 1977), reduced food consumption and egg production in the ring-necked pheasant (*Phasianus colchicus*) (Stromborg 1977), and behavioral modifications, reduced food intake, alterations in liver enzyme activities, reductions in vitamin concentrations, reduced body temperature, and lowered resistance to cold stress in white-footed mice (*Peromyscus leucopus*) (Montz and Kirkpatrick 1985) have been noted at diazinon concentrations markedly lower than those causing acute mortality. It has been suggested, but not proven, that wildlife partially disabled in the field as a result of diazinon poisoning would be more likely to die of exposure, predation, starvation, or dehydration, or face behaviorial abnormalities learning, impairments, and reproductive declines than would similarly treated domestic or laboratory animals, (Montz 1983; Montz and Kirkpatrick 1985).

In this account, I summarize available data on the environmental fate and effects of diazinon, with emphasis on fish and wildlife resources. Also included are recommendations for the protection of sensitive species of concern to the U.S. Fish and Wildlife Service. This report is part of a continuing series of synoptic reviews prepared in response to requests for information from Service environmental specialists.

ENVIRONMENTAL CHEMISTRY

Diazinon is a broad spectrum insecticide that is effective against a variety of orchard, vegetable, and soil pests, ectoparasites, flies, lice, and fleas. It exists as a technical grade product, a wettable powder, an emulsifiable concentrate, as granules, and in a variety of other formulations (Negherbon 1959; Anon. 1972; Eberle 1974; Berg 1984). The active ingredient in diazinon is phosphorothioic acid 0,0- diethyl 0- (6-methyl-2-1(methylethyl)-4-pyrimidinyl) ester (Figure 1). Its molecular formula and molecular weight are $C_{12}H_{2l}N_2O_3PS$, and 304.35. The technical grade is light amber to dark brown, and boils at 83 to 84 C. Diazinon is soluble in water to 40 mg/l, and dissolves readily in aliphatic and aromatic solvents, alcohols, and ketones. Diazinon may be stored on the shelf for at least 3 years with negligible degradation. Diazinon is also known as G-24480, Sarolex, Spectracide (Anon. 1972), AG-500, Alfa-tox, Basudin, Dazzel, Diazajet, Diazide, Diazol, ENT 19507, Gardentox, Neocidol, Nucidol, CAS 333-41-5 (Hudson et al. 1984), Diagran, Dianon, DiaterrFos, Diazatol, Dizinon, Dyzol, D.z.n., Fezudin, Kayazinon, Kayazol, Knox Out, and Nipsan (Berg 1984).

Some diazinon formulations contain 0.2 to 0.7% (2,000 to 7,000 mg/kg) of Sulfotep (tetraethyl dithiopyrophosphate) as a manufacturing impurity; Sulfotep is reportedly at least 100 times more toxic than diazinon to some organisms (Jarvinen and Tanner 1982). It seems that additional, research is warranted on diazinon/Sulfotep interactions.

Diazinon degrades rapidly in plants, with half-time persistence usually less than 14 days; however, persistence increases as temperatures decrease, and is longer in crops with a high oil content (Table 1). In water, diazinon breaks down to comparatively nontoxic compounds with little known hazard potential to aquatic species (Meier et al. 1976; Jarvinen and Tanner 1982), although the degradation rate is highly dependent on pH (Table 1). In soils, diazinon seldom penetrates below the top 1.3 cm (Kuhr and Tashiro 1978; Branham and Wehner 1985). But diazinon may remain biologically available in soils for 6 months or longer at low temperature, low moisture, high alkalinity, and lack of suitable microbial degraders (Anon. 1972; Bartsch 1974; Meier et al. 1976; Allison and Hermanutz 1977; Menzie 1978; Forrest et al. 1981; Branham and Wehner 1985). Bacterial enzymes, derived from *Pseudomonas* sp., can be used to hydrolyze diazinon in soil, although costs are prohibitive except in treating emergency situations involving spills of concentrated diazinon solutions. In one case, diazinon was enzymatically hydrolyzed within 24 hours in an agricultural sandy soil at concentrations as high as 10,000 mg/kg (Barik and Munnecke 1982).

Table 1. Persistence of diazinon in plants, soil, and water.

Sample type and other variables	Time for 50% persistence	Reference[a]
Plants		
Cabbage leaves		
Summer	14 days	1
Winter	>14 days	1
Leafy vegetables, forage crops	<2 days	2
Other vegetables, cereal products	<7 days	2
Fruits	4 days	2
Carrots, oil seed plants	> 4 days	2
Grass	7 days	3
Soil	2 to 4 weeks	2, 4
Water		
Lake Superior	30 days (14–184 days)	5
River water	39 days	6
Effect of pH		
3.1	12 h	7
6.0	2 weeks	8
7.4	6 months	8
9.0	4 months	8
10.4	6 days	7

[a]References: 1, Montz 1983; 2, Bartsch 1974; 3, Kuhr and Tashiro 1978; 4, Branham and Wehner 1985; 5, Jarvinen and Tanner 1982; 6, Arthur et al. 1983; 7, Meier et al. 1976; 8, Allison and Hermanutz 1977.

In almost every instance of diazinon poisoning, there has been a general reduction in cholinesterase activity levels, especially in brain and blood. Diazinon exerts its toxicity by binding to the neuronal enzyme acetylcholinesterase (AChE) for a considerable time postexposure (Montz 1983). It is emphasized that all organophosphorus pesticide compounds, in sufficient dose, inhibit AChE in vivo, and all share a common

mechanism of acute toxic action (Murphy 1975). AChE inhibition results in the accumulation of endogenous acetylcholine-in nerve tissues and effector organs, resulting in signs that mimic the muscarinic, nicotinic, and central nervous system (CNS) actions of acetylcholine. The immediate cause of death in fatal organophosphorus compound poisonings, including diazinon, is asphyxia resulting from respiratory failure. Contributing factors are the muscarinic actions of bronchoconstriction and increased bronchial secretions, nicotinic actions leading to paralysis of the respiratory muscles, and the CNS action of depression and paralysis of the respiratory center (Murphy 1975).

Diazinon is not a potent inhibitor of cholinesterase, and must be converted to its oxygen analogues (oxons), especially diazoxon (diethyl 2-isopropyl-6-methylpyrimidin-4-yl phosphate) in vivo before poisoning can occur (Wahla et al. 1976). Diazoxon is about 10,000 times more effective in reducing cholinesterase activity levels than diazinon (Fog and Asaka 1982). At least eight diazinon metabolites have been identified in vertebrates, of which four are oxons (Machin et al. 1975; Menzie 1978; Seguchi and Asaka 1981). It is generally agreed that diazinon is metabolized to diazoxon through the action of liver mixed-function oxidases and nicotinic adenine nucleotide phosphate (Menzie 1978; McLean et al. 1984). Diazinon toxicity will depend to some extent upon the relation between the rates of activation of diazinon to diazoxon, and of decomposition of the latter to harmless products (Fujii and Asaka 1982). Birds are more sensitive to diazinon than mammals, probably because mammalian blood enzymes hydrolyze diazoxon rapidly, whereas bird blood has virtually no hydrolytic activity. It seems that diazoxon stability in blood is a major factor affecting susceptibility of birds and mammals to diazinon poisoning (Machin et al. 1975).

Diazinon poisoning effects in animals can be delayed or prevented by treatment with a variety of compounds. For example, AChE in diazinon-stressed birds can be reactivated by pralidoxime (Egyed et al. 1976; Fleming and Bradbury 1981; Misawa et al. 1982). Furthermore, pretreatment of large white butterfly (*Pieris brassicae*) larvae with methylene dioxyphenyl compounds will inhibit the diazinon to diazoxon activation (Wahla et al. 1976). Added tryptophan and its metabolites may prevent teratogenic defects by maintaining nicotinic adenine nucleotide (NAD) levels in diazinon-treated chicken embryos; diazinon reportedly acts to decrease the availability of tryptophan to bird embryos, subsequently interfering with NAD metabolism and causing birth defects (Henderson and Kitos 1982). NAD metabolism in diazinon-stressed birds may also be maintained with nicotinamide (Misawa et al. 1982). In contrast to many other organophosphorus insecticides, organisms that survive diazinon-inhibited cholinesterase levels can undergo considerable spontaneous reactivation (dephosphorylation), indicating that its dephosphorylation occurs more readily than that of cholinesterase inhibited by other organophosphorus compounds (Fleming and Bradbury 1981).

ACUTE AND CHRONIC TOXICITY

GENERAL

Diazinon toxicity varies widely within and among species, and is modified by organism age, sex, and body size, climatic conditions, pesticide formulation, chemistry of the environment, and other factors (Montz 1983). Nevertheless, several trends are apparent as judged by available data. Among aquatic organisms, for example, freshwater cladocerans and marine shrimps were the most sensitive species tested, with LC-50 (96 h) values of less than 5 ug/l; freshwater teleosts were more resistant, with the lowest LC-50 (96 h) value recorded being 90 ug/l. Diazinon has considerable potential for causing acute avian poisoning episodes; sensitive species of birds, including ducks, turkey (*Meleagris gallopavo*), and red-winged blackbird (*Agelaius phoeniceus*), died at single oral doses of 2 mg of diazinon/kg body weight. Mammals are more resistant than birds to diazinon; the lowest LD-50 (acute oral) value recorded is 224 mg/kg body weight for female rats (*Rattus rattus*). Chronic oral toxicity tests with mammals suggest that daily intake exceeding 5 or 10 mg diazinon/kg body weight is probably fatal over time to swine (*Sus scrofa*) and dogs (*Canis familiaris*), respectively. Finally, 9 mg/kg of dietary diazinon fed during gestation to pregnant mice (*Mus musculus*) was associated with significant mortality of pups prior to weaning.

AQUATIC ORGANISMS

Freshwater cladocerans and marine crustaceans were the most sensitive groups tested, with LC-50(96 h) values of less than 5 ug/l for the more sensitive species (Table 2). Rainbow trout (*Salmo gairdneri*) and bluegill (*Lepomis macrochirus*) seemed to be the least resistant freshwater teleosts tested, with LC-50(96 h) values

between 90 and 120 ug/l; however, the postlarval and juvenile stages of the striped knifejaw (*Oplegnathus fasciatus*) --a marine fish cultured intensively in Japan-- were unusually sensitive (Table 2). In general, technical grade formulations of diazinon seem to be more toxic than emulsifiable concentrates, dusts, and oil solutions (Table 2). Also, large variations in acute toxicity values were evident, even among closely related species (Table 2).

Outward signs of diazinon poisoning in fish included lethargy, forward extension of pectoral fins, darkened areas on posterior part of body, hyperexcitability when startled, sudden rapid swimming in circles, and severe muscular contractions (Goodman et al. 1979). Internally, physiological mechanisms in teleosts preceding death involved the following sequence: cholinesterase inhibition, acetylcholine accumulation, disruption of nerve functions, respiratory failure, and asphyxia (Sastry and Sharma 1980).

Limited data indicated that the yellowtail (*Seriola quinqueradiata*), a marine teleost, was 84X more sensitive to diazinon than were 4 species of freshwater fishes, as judged by LC-50(48 h) values, and by its inability to biotransform diazinon to nontoxic metabolites within one hour (Fujii and Asaka 1982). Diazinon has not been detected in marine waters, but the potential exists for contamination of estuarine areas from agricultural and urban runoff (Goodman et al. 1979).

BIRDS

Diazinon adversely affects survival of developing mallard embryos when the eggshell surface is subjected for 30 seconds to concentrations 25 to 34 times higher than recommended field application rates; mortality patterns were similar for solutions applied in water or in oil (Table 3). This laboratory finding suggests that eggs of mallards, and probably other birds, are protected when diazinon is applied according to label directions. Chickens dipped in solutions containing 1,000 mg of diazinon/l, an accidentally high formulation, experienced 60% mortality within 3 days; no other deaths occurred during the next 4 months (Sokkar et al. 1975).

Results of 5-day feeding trials with 2-week-old Japanese quail (*Coturnix japonica*), followed by 3 days on untreated feed, showed an LD-50 of 167 mg diazinon/kg diet -- a concentration considered "very toxic". No deaths were observed at dietary levels of 85 mg diazinon/kg, but 53% died at 170 mg/kg, and 87% at 240 mg/kg (Hill and Camardese 1986).

Diazinon has a potential for causing acute avian poisoning episodes (Schafer et al. 1983). Ingestion of 5 granules of Diazinon 14G (14.3% diazinon) killed 80% of house sparrows (*Passer domesticus*), and all red-winged blackbirds to which they were administered (Balcomb et al. 1984). Ingestion of fewer than 5 granules of Diazinon 14G, each containing about 215 ug of diazinon, could be lethal to sparrow-sized birds (i.e., 15 to 35 g body weight), especially juveniles of seed-eaters (Hill and Camardese 1984). Acute oral LD-50's indicate that 15 mg of diazinon/kg body weight is fatal to virtually all species tested, and that 2 to 5 mg/kg is lethal to the more sensitive species (Table 4). Signs of diazinon poisoning in birds included muscular incoordination, wing spasms, wing-drop, hunched back, labored breathing, spasmodic contractions of the anal sphincter, diarrhea, salivation, lacrimation (tear production), eyelid drooping, prostration, and arching of the neck over the back (Hudson et al. 1984). Most of these signs have been observed in birds poisoned by compounds other than diazinon; these compounds also act via an anticholinesterase mode of action (Hudson et al. 1984).

Table 2. Acute toxicity of diazinon to aquatic organisms. All values shown are in micrograms of diazinon (active ingredients)/liter of medium fatal to 50% in 96 hours.

Ecosystem, taxonomic group organism, and other variables	LC-50 (96 h), in µg/L	Reference[a]
Freshwater		
Invertebrates		
Daphnid, *Daphnia magna*		
Dust (27%)	1.2	1
Emulsifiable concentrate (47.5%)	1.3	1
Technical grade (91.9%)	2.0	1
Oil solution (0.5%)	13.0	1
Cladoceran, *Simocephalus serrulatus*	1.4[b]	2
Stonefly, *Pteronarcys californica*	25	2
Amphipod, *Gammarus fasciatus*	200	2
Daphnid, *Daphnia pulex*	800[b]	2
Fish		
Rainbow trout, *Salmo gairdneri*	90–400	2, 3
Technical grade	110	1
Emulsifiable concentrate	3,000	1
Dust	3,200	1
Oil solution	19,000	1
Bluegill, *Lepomis macrochirus*	120–670	2, 3, 4
Technical grade	120	1
Emulsifiable concentrate	530	1
Dust	170	1
Oil solution	160	1
Lake trout, *Salvelinus namaycush*	602	2
Brook trout, *Salvelinus fontinalis*	770	4
Flagfish, *Jordanella floridae*	1,600	4
Cutthroat trout, *Salmo clarki*	1,700	2
Murrel, *Channa punctatus*	3,100	5
Fathead minnow, *Pimephales promelas*	5,100–15,000	4, 6
Goldfish, *Carassius auratus*	9,000	3
Amphibians		
Bullfrog, *Rana catesbeiana*	>2,000,000[c]	7
Marine		
Invertebrates		
Mysid shrimp, *Mysidopsis bahia*	4.8	8

Penaeid shrimp, *Penaeus aztecus*	28[b]	8
Fish		
Sheepshead minnow, *Cyprinodon variegatus*	1,470	9
Striped knifejaw, *Opelgnathus fasciatus*		
Egg	3,200[d]	10
Prelarvae	5,500[d]	10
Postlarvae	25.1[d]	10
Juvenile	27.8[d]	10

[a]References: 1, Meier et al. 1976; 2, Johnson and Finley 1980; 3, Anon. 1972; 4, Allison and Hermanutz 1977; 5, Sastry and Malik 1982; 6, Jarvinen and Tanner 1982; 7, Hudson et al. 1984; 8, Nimmo et al. 1981; 9, Goodman et al. 1979; 10, Seikai 1982.

[b]48 h value.

[c]Single oral dose, in g/kg body weight.

[d]24 h value.

Table 3. Mortality of mallard embryos after immersion for 30 seconds in graded strength diazinon solutions (after Hoffman and Eastin 1981).

Age of eggs (days)	Solution vehicle (water or oil)	Diazinon concentration (mg/L)	Percent dead	Approximate field application rate
3	Water	11	none	0.5
3	Water	110	3	5
3	Water	542	50	25
8	Water	597	50	27
3	Oil	13	none	0.6
3	Oil	133	7	6
3	Oil	648	50	29
8	Oil	741	50	34

MAMMALS

Signs of diazinon poisoning in mammals included a reduction in blood and brain cholinesterase activity, diarrhea, sweating, vomiting, salivation, cyanosis, muscle twitches, convulsions, loss of reflexes, loss of sphincter control, and coma (Anon. 1972). Other compounds that produce their toxic effects by inhibiting AChE, such as organophosphorus pesticides and many carbamates, show similar effects (Murphy 1975). Two species of marmoset accidentally poisoned by diazinon exhibited, prior to death, high-pitched voices, trembling, frog-like jumping, a stiff gait, and pale oral mucous membranes; internally, bone marrow necrosis and hemorrhages in several organs were evident (Brack and Rothe 1982). Internal damage was also observed in swine and dogs that died following controlled administration of diazinon. Swine showed histopathology of liver and intestinal tract, and duodenal ulcers; dogs showed occasional rupture of the intestinal wall, and testicular atrophy (Earl et al. 1971).

Results of acute oral toxicity tests indicated that the rat was the most sensitive mammalian species tested, with an acute oral LD-50 of 224 mg diazinon/kg body weight (Table 4). It is clear that mammals are significantly more resistant to acute oral poisoning by diazinon than birds (Table 4). Diazinon was also toxic to mammals when administered dermally, through inhalation, and in the diet (Table 5). The lowest dermal LD-50 recorded was 600 mg diazinon/kg body weight for rabbits (*Lepus* sp.) using an emulsifiable (4E) formulation. The single datum for inhalation toxicity indicated that 27.2 mg of diazinon/l of air killed 50% of test rabbits after exposure for 4 hours (Table 5). Pregnant mice fed diets containing 9 mg of diazinon/kg during gestation all survived, but some pups died prior to weaning (Table 5). Results of chronic oral toxicity tests of diazinon indicated that death was probable if daily doses exceeded 5 mg/mg body weight for swine, or 10 mg/kg for dogs (Table 5).

TERRESTRIAL INVERTEBRATES

Accidental spraying of beehives in Connecticut with diazinon resulted in a complete kill of resident honeybees. Dead bees contained up to 3 mg/kg of diazinon (Anderson and Glowa 1984). Diazinon is an an effective insecticide. LD-50 values for diazinon and adult houseflies (*Musca domestica*), applied topically, were 0.4 ug/insect, or 4.6 mg/kg body weight (Negherbon 1959). LD-50 values for larvae of the large white butterfly, applied topically, were 8.8 mg/kg body weight for diazinon, and 11.0 mg/kg body weight for diazoxon (Wahla et al. 1976). Pretreatment of larvae with methylene dioxyphenyl compounds antagonized the action of diazinon by a factor of about 2, but synergized the action of diazoxon by an order of magnitude (Wahla et al. 1976).

Table 4. Acute oral toxicity of diazinon to birds and mammals. All values shown are in milligrams of diazinon/kg body weight fatal to 50% after a single oral dose.

Taxonomic group, organism and other variables	LD-50 (range), in mg/kg body weight	Reference[a]
Birds		
Turkey, *Meleagris gallopavo*	2.5	1
Red-winged blackbird, *Agelaius phoeniceus*	2.6	2
Goslings, *Anser* spp.	2.7	1
Turkey	3.5	3
Ducks, *Anas* spp.	3.5	3
Mallard, *Anas platyrhynchos*	3.5 (2.4–5.3)	4, 5
European quail, *Coturnix coturnix*	4.2	2
Ring-necked pheasant, *Phasianus colchicus*	4.3 (3.0–6.2)	4, 5
Northern bobwhite, *Colinus virginianus*	5.0[b]	6
Chicks, *Gallus gallus*	5.0[c]	1
Chicken, *Gallus gallus*	9.0	3
Turkey	10.0[c]	1
Ducklings	14.0	1
Northern bobwhite	14.7	6
Northern bobwhite	25.0[c]	6
European starling, *Sturnus vulgaris*	213	2
Mammals		

Rat, *Rattus rattus*	425	3, 5
Technical grade	350	7
AG 500 (granule)	327	7
4 E (emulsion)	542	7
4 S (spray)	735	7
50 W (wettable)		
Males	521	7
Females	224	7
Pig, *Sus scrofa*	400	3
Guinea pig, *Cavia cobaya*	450	3
Dog, *Canis familiaris*	>500	8
Sheep, *Ovis aries*	>1,000	3

[a]References: 1, Egyed et al. 1974; 2, Schaefer et al. 1983; 3, Machin et al. 1975; 4, Hudson et al. 1984; 5, Zinkl et al. 1978; 6, Hill et al. 1984; 7, Anon. 1972; 8, Earl et al. 1971.

[b]No mortality seen. [c]All animals tested died.

Table 5. Toxicity of diazinon to laboratory animals via dermal, inhalation, dietary, and chronic oral routes of administration.

Mode of administration, units, organism, formulations, and other variables	Dose	Effect	Reference[a]
Dermal, in mg/kg body weight			
Rabbit, *Lepus* sp.			
AG-500 (granule	900	LD-50	1
4 E (emulsion)	600	LD-50	1
4 S (spray)	735	LD-50	1
14 G (granule)	>15,400	LD-50	1
50 W (wettable)	>2,000	LD-50	1
Mice, *Mus musculus*			
Technical diazinon	2,750	LD-50	2
Inhalation, in mg/L air			
Rabbit[b]	27.2	LC-50	1
Dietary, in mg/kg diet, during gestation only			
Mice	0.18	No pup deaths at weaning	3
"	9	12% of pups dead prior to weaning	3
Chronic oral, in mg/kg body weight daily			
Dog, *Canis familiaris*	10	None dead in 8 months	4
"	20	All dead in 30 days	4
"	25	None dead in 15 days	4

"	50	None dead in 4 days	4
Swine, *Sus scrofa*	5	None dead in 8 months	4
"	10	75% dead in 30 days	4

[a]References: 1, Anon., 1972; 2, Skinner and Kilgore 1982; 3, Barnett et al. 1980; 4, Earl et al. 1971.

[b]Exposure for 4 h to 4% aqueous suspension.

SUBLETHAL EFFECTS

GENERAL

Among sensitive species of aquatic organisms, diazinon was associated with reduced growth and reproduction in marine and freshwater invertebrates and teleosts, spinal deformities in fish, reduced emergence in stream insects, measurable accumulations in tissues, increased numbers of stream macroinvertebrates carried downstream by currents (drift), possible mutagenicity in fish, and interference with algal-invertebrate interactions. In birds, diazinon is a known teratogen; it also is associated with reduced egg production, decreased food intake, and loss in body weight. Diazinon fed to pregnant mice resulted in offspring with brain pathology, delayed sexual maturity, and adverse behavioral modifications that became apparent late in life. For all groups tested, diazinon directly or indirectly inhibited cholinesterase activity.

AQUATIC ORGANISMS

Spinal deformities, mostly lordosis and scoliosis, were among the more insidious effects documented for diazinon. Malformations were observed in fathead minnows (*Pimephales promelas*) after 19 weeks in water containing 3.2 ug diazinon/l (Allison and Hermanutz 1977), in yearling brook trout (*Salvelinus fontinalis*) within a few weeks at 4.8 ug/l (Allison and Hermanutz 1977), and in various species of freshwater teleosts after exposure for 7 days to 50 ug diazinon/l (Kanazawa 1978).

Diazinon is a noncarcinogen and reportedly has no significant mutagenic activity in microbial systems, yeast, and mammals including humans (as quoted in Vigfusson et al. 1983). However, Vigfusson et al. (1983) have measured a significant increase in the frequency of sister chromatid exchange in central mud minnows (*Umbra limi*) that were exposed *in vivo* for 11 days to solutions containing 0.16 to 1.6 ug of diazinon/l. This finding requires verification.

Diazinon in water is bioconcentrated by brook trout at levels as low as 0.55 ug/l, but tissue residues for all aquatic organisms did not exceed 213 times that of ambient water, even after months of continuous exposure (Table 6). Diazinon and its metabolites are excreted rapidly posttreatment; the loss rate is approximately linear (Kanazawa 1978). The enzyme system responsible for diazinon metabolism in fish liver microsomes required NADPH and oxygen for the oxidative desulfuration of diazinon to diazoxon (Hogan and Knowles 1972). Fish with high fat content contained greater residues of diazinon in fatty tissues than fish with comparatively low lipid content (Seguchi and Asaka 1981), and this could account, in part, for inter-and intraspecies variations in uptake and depuration. Some organisms, such as the sheepshead minnow (*Cyprinodon variegatus*), have measurable diazinon residues during initial exposure to 6.5 ug/l, but no detectable residues after lengthy exposure (Goodman et al. 1979), suggesting that physiological adaptation resulting in rapid detoxication is possible.

Freshwater and marine alga were unaffected at water diazinon concentrations, that were fatal (i.e., 1,000 ug/l) to aquatic invertebrates (Stadnyk and Campbell 1971; Shacklock and Croft 1981). However, diazinon at 1.0 ug/l induced extensive clumping of a freshwater alga (*Chlorella pyrenoidosa*) onto the antennae of *Daphnia magna* within 24 hours (Stratton and Corke 1981). The affected daphnids were immobilized and settled to the bottom of the test containers. The causes of particulate matter adhesion are open to speculation, and additional research is merited.

Freshwater macroinvertebrates were comparatively sensitive to diazinon (Table 7). Results of large scale experimental stream studies (Arthur et al. 1983) showed that dose levels of 0.3 ug diazinon/l caused a 5 to 8-fold reduction in emergence of mayflies and caddisflies within 3 weeks; after 12 weeks, mayflies, damselflies, caddisflies, and amphipods were absent from benthic samples. Elevated (and catastrophic) drift of stream

invertebrates also was documented in diazinon-treated streams, especially for amphipods, leeches, and snails (Arthur et al. 1983).

Freshwater fish populations can be directly damaged by prolonged exposure to diazinon at concentrations up to several hundred times lower than those causing acute mortality (Sastry and Sharma 1980; Sastry and Malik 1982; Table 7). Impaired reproduction and AChE inhibition occurs concurrently in teleosts during long-term exposure to diazinon, but reproduction can be impaired for at least 3 weeks after fish are placed in uncontaminated water, even though AChE is normal and they contained no detectable diazinon residues (Goodman et al. 1979). Furthermore, diazinon exposure during spawning caused complete, but temporary, inhibition of reproduction at concentrations which did not produce this effect in fish exposed since hatch (Allison 1977). This could severely impact aquatic species with a short reproductive period (Allison 1977).

BIRDS

Diazinon produces visible Type I and II teratisms when injected into chicken embryos (Misawa et al. 1981, 1982; Henderson and Kitos 1982; Wyttenbach and Hwang 1984). Type I teratisms (related to tissue NAD depression) included abnormal beaks, abnormal feathering, and shortened limbs. Type II teratisms, which included short and wry neck, leg musculature hypoplasia, and rumplessness were associated with disruptions in the nicotinic cholinergic system. The severity of effects depended on embryo age and was dose-related. Chick embryos (age 48 hours) receiving 25 ug or more of diazinon/embryo had cervical notochord and neural tube malformations at 96 hours, and short neck at 19 days (Wyttenbach and Hwang 1984). Wry neck occurred at doses ranging from 6.2 to 100 ug/embryo, but was more frequent at higher doses. Type II teratisms were attributed to disruption of notochord sheath formation. Coinjection of 2-pyridinealdoxime methochloride (2-PAM) along with 200 ug of diazinon/embryo markedly reduced notochord and neural tube deformations (Wyttenbach and Hwang 1984). Similarly, the copresence of tryptophan--or its metabolites L-kynurenine, 3-hydroxyanthronilic acid, quinolinic acid--maintained NAD levels of diazinon-treated embryos close to, or above, normal, and significantly alleviated the symptoms of Type I teratisms (Henderson and Kitos 1982).

Table 6. Accumulation of diazinon by aquatic organisms.

Ecosystem, taxonomic group, organism, and other variables	Diazinon concentration in water (ug/l)	Exposure period (d = days, m = months)	Concentration factor	Reference[a]
Freshwater				
Invertebrates				
Crayfish, *Procambarus clarkii*				
Whole	10	7 d	5	1
Pond snail, *Cipangopaludina malleata*				
Whole	10	7 d	6	1
Red snail, *Indoplanorbis exustus*				
Whole	10	7 d	17	1
Shrimp, *Penaeopsis joyneri*				
Whole	20	14 d	3	2
Whole	20	14 d + 7 d posttreatment (pt)	<1	2
Fish				
4 spp., whole	10	7 d	18–152	1
3 spp., whole	20	14 d	26–120	2

3 spp., whole	20	14 d + 7 d pt	<1	2
Topmouth gudgeon,				
Pseudorasbora parva				
Whole	10	14 d	173	1
Whole	10	14 d + 1 d pt	72	1
Whole	10	14 d + 4 pt	8	1
Whole	10	14 d + 8 d pt	<1	1
Brook trout, *Salvelinus fontinalis*				
Adult				
Muscle	0.55	8 m	25	3
Blood	1.1	6 m	17	3
Muscle	1.1	8 m	25	3
Muscle	2.4	8 m	35	3
Blood	4.8	6 m	13	3
Muscle				
Mature male	4.8	8 m	24	3
Spawned female	4.8	8 m	19	3
Immature male				
Muscle	4.8	8 m	51	3
Adult female				
Egg	9.6	8 m	151	3
Muscle	9.6	8 m	34	3
Marine				
Fish				
Sheepshead minnow,				
Cyprinodon variegatus				
Whole	1.8	4 d	147	4
Whole	3.5	4 d	147	4
Whole	6.5	4 d	213	4
Whole	6.5	4 d + 8 d pt	<1	4
Whole	6.5	108 d	<1	4
Egg	<0.98	LC[b]	<1	4
Egg	1.8–6.5	LC[b]	10–13	4

[a]References: 1, Kanazawa 1978; 2, Seguchi and Asaka 1981; 3, Allison and Hermanutz 1977; 4, Goodman et al. 1979.

[b]LC = life cycle.

Table 7. Lowest tested diazinon concentrations that produce significant biological effects to aquatic organisms.

Ecosystem, and taxonomic group	Water concentration in µg/L	Effect	Reference[a]
Freshwater			
Invertebrates			
Insects	0.3	Lowered emergence	1
Amphipods	0.3	Elevated drift	1
Daphnids	1.0	Immobilization	2
Fish			
Brook trout, *Salvelinus fontinalis*	0.55	Reduced growth of progeny	3
Fathead minnow, *Pimephales promelas*	3.2	Reduced hatching success	3
Flagfish, *Jordanella floridae*	14.0	Reduced larval growth	4
Marine			
Invertebrates			
Mysid shrimp, *Mysidopsis bahia*	3.2	Reduced growth and reproduction	5
Fish			
Sheepshead minnow, *Cyprinodon variegatus*	0.47	Reduced fecundity	3

[a]References: 1, Arthur et al. 1983; 2, Stratton and Corke 1981; 3, Goodman et al. 1979; 4, Allison and Hermanutz 1977; 5, Nimmo et al. 1981.

Reduced egg production, depressed food consumption, and loss in body weight have been observed in ring-necked pheasants at daily diazinon intakes greater than 1.05 mg/bird; a dose-related delay in recovery of egg laying was noted after termination of diazinon treatment (Stromborg 1977, 1979). Threshold levels in ring-necked pheasants of 1.05 and 2.1 mg of diazinon daily corresponded to 1/16 and 1/8 of daily ration (70 g) treated at commercial application rates. Food consumption of ring-necked pheasants was reduced significantly when only food treated with diazinon was available; pheasants avoided diazinon-treated food if suitable alternatives existed (Stromborg 1977; Bennett and Prince 1981). Dietary levels above 50 mg/kg were associated with reduced food consumption, weight loss, and reduction in egg production in northern bobwhites (Stromborg 1981). If food reduction is important, then diets containing more than 17.5 mg diazinon/kg (based on, empirical calculations) were potentially harmful to bobwhites (Stromborg 1981). The mechanisms accounting for reduction in egg deposition are not clear, but are probably related primarily to decreased food intake. They may also be associated with diazinon-induced pituitary hypofunction at the level of the hypothalamus, resulting in reduced synthesis and secretion of gonadotrophic, thyrotrophic, and adrenocorticotrophic hormones (Sokkar et al. 1975).

MAMMALS

Diazinon exerts its toxic effects by binding to the neuronal enzyme acetylcholinesterase (AChE) for long periods after exposure. Diazinon, in turn, is converted to diazoxon, which has a higher affinity for AChE (and thus greater toxicity) than the parent compound. There is a latent period in white-footed mice in reduction of

cholinesterase activities, sometimes up to 6 hours, until diazinon is converted to diazoxon (Montz 1983). Effects of multiple doses of diazinon to mammals are not clear, e.g., rats exposed to a high dose of diazinon did not respond fully to a second dose until one month later (Kikuchi et al. 1981). It is difficult to ascertain when complete recovery of diazinon-poisoned animals has occurred. It is speculated, but not verified, that wildlife recovering from diazinon poisoning may face increased predation, aberrant behavior, learning disabilities, hypothermia, and reproductive impairments (Montz 1983). Data are now lacking on recovery aspects of diazinon-poisoned native mammal populations (Montz and Kirkpatrick 1985).

Diazinon is rapidly biotransformed and excreted in mammals. Estimated half-times of diazinon persistence were 6 to 12 hours in rats (Anon. 1972) and dogs (Iverson et al. 1975). Most of the diazinon metabolites were excreted in the urine as diethyl phosphoric acid and diethyl phosphorothioic acid in dogs (Iverson et al. 1975), and as hydroxy diazinon and dehydrodiazinon in sheep (Machin et al. 1974).

Determination of AChE activity in selected tissues following diazinon exposure provided an estimate of potential toxicity, but tissue sensitivity varied widely between and among taxa. In sheep, brain cholinesterase inhibition was pronounced after diazinon insult, and metabolism of diazinon in, or close to, the brain was the most likely, source of toxicologically effective diazoxon (Machin et al. 1974, 1975). In rat, diazinon effectively reduced blood cholinesterase levels, with inhibition significantly more evident in erythrocytes than in plasma (Tomokuni and Hasegawa 1985). All mammalian bloods hydrolyze diazoxon rapidly, whereas birds have virtually no hydrolytic activity in their blood, and, as a result, were more susceptible than mammals. The stability of diazoxon in the blood appears to be a primary factor in susceptibility to diazinon poisoning (Machin et al. 1975). In species lacking blood oxonases, the liver was probably the most important site of diazinon metabolism (Machin et al. 1975). Diazinon that accumulated in rat liver was biotransformed, usually within 24 hours, by microsomal mixed-function oxidases and glutathione S-transferases; however, diazinon residues in rat kidney were almost 500 times those in liver (and 11 times brain), and were measurable in kidney but not in liver (Tomokuni and Hasegawa 1985). It now seems that diazinon residues in kidney, and cholinesterase inhibition in erythrocytes are the most useful indicators of acute diazinon poisoning in mammals.

Sublethal effects of diazinon have been recorded in rodents, the most sensitive mammal group tested. Effects were measured at 0.5 mg diazinon/kg in diets of rats for 5 weeks, at 0.18 mg/kg body weight administered daily to pregnant mice, and at single doses of 1.8 mg/kg body weight for rat and 2.3 mg/kg body weight for white-footed mice (Table 8). Many variables modify diazinon-induced responses, including the organism's sex. For example, female rats and dogs were more sensitive to diazinon than males (Earl et al. 1971; Davies and Holub 1980a, 1980b; Kikuchi et al. 1981), but male swine were more sensitive than females (Earl et al. 1971).

Table 8. Sublethal effects of diazinon to selected mammals.

Organism and dose (D = mg/kg diet; BW = mg/kg body weight daily)	Exposure period	Effect	Reference[a]
Rat, *Rattus rattus*			
0.009 (BW)	5 weeks	No effect	1
0.1 (D)	5 weeks	No effect	1
0.5 (D)	5 weeks	Depressed plasma cholinesterase	1
1.8 (BW)	Single dose	Elevated serum glucuronidase	2
2 (D)	1 week	Depressed plasma cholinesterase (females only)	3
3.8 (BW)	Single dose	Altered blood chemistry	4
10 (D)	2 years	Cholinesterase inhibition	5
1,000 (D)	2 years	Reduced growth	5
1,000 (D)	3 generations	No malformations, no effect on reproduction	5
Mice, *Mus musculus* (pregnant)			
0.18 (BW)	2.8 weeks	Altered behavior and delayed sexual maturity of progeny	6
9 (BW)	Throughout gestation	Reduced growth and altered serum immunoglobulins of progeny; some deaths	7
Mice (juveniles)			
0.18 (BW)	14.4 weeks	Impaired endurance and coordination	6
9 (BW)	14.4 weeks	Brain pathology	6
White-footed mice, *Peromyscus leucopus*			
2.3 (BW)	Single dose	9% depression in brain AChE in 24 h	8
17.3 (BW)	Single dose	60% depression in brain AChE in 6 h	9
Dog, *Canis familiaris*			
4 (BW)	Single dose	39% reduction in serum cholinesterase in 10 min; 50% reduction in 3.5 h	10
4.3–5.3 (BW)	43 weeks	Cholinesterase inhibition	5

10 (BW)	8 months	Testicular atrophy, cholinesterase inhibition	11
75 (BW)	Single dose	Acute pancreatitis	12
Swine, *Sus scrofa*			
5 (BW)	8 months	Cholinesterase inhibition, duodenal ulcers, liver pathology	11
Sheep, *Ovis aries*			
Sprayed with 100 ppm diazinon solutions	4 min	Effective lice control for 3 weeks, partial protection for 8.6 weeks	13
450–650 (BW)	Single dose	Flesh unfit for human consumption for several weeks (high fat residues of 333–520 mg/kg)	14
Monkeys, several species			
0.5 (BW)	2.04 years	None	5
5 (BW)	2.04 years	Cholinesterase inhibition	5

[a]References: 1, Davies and Holub 1980a; 2, Kikuchi et al. 1981; 3, Davies and Holub 1980b; 4, Lox 1983; 5, Anon. 1972; 6, Spyker and Avery 1977; 7, Barnett et al. 1980; 8, Montz 1983; 9, Montz and Kirkpatrick 1985; 10, Iverson et al. 1975; 11, Earl et al. 1971; 12, Dressel et al. 1982; 13, Wilkinson 1980; 14, Machin et al. 1974.

Behavioral deficits observed in offspring of mice exposed to diazinon during gestation indicated that prenatal exposure may produce subtle dysfunctions not apparent until later in life (Spyker and Avery 1977). Pregnant mice given a daily dose of 0.18 or 9 mg diazinon per kg body weight throughout gestation gave birth to viable, overtly normal, offspring. But, pups born to mothers of the 9 mg/kg groups grew more slowly than controls and were significantly smaller at 1 month than controls (Spyker and Avery 1977). Offspring of mothers receiving 0.18 mg/kg body weight exhibited significant delays in the appearance of the contact placing reflex, and in descent of testes or vaginal opening. Mature offspring of mothers exposed to either dose level displayed impaired endurance and coordination on rod cling and inclined plane tests of neuromuscular function (Table 8). In addition, offspring of the 9 mg/kg dose had slower running speeds and less endurance in a swimming test than controls. At 101 days, forebrain neuropathology was evident in the 9 mg/kg dose, but not in the 0.18 mg/kg group. The mechanisms responsible for these effects are unknown (Spyker and Avery 1977).

Diazinon is nonmutagenic to mammals, as judged by its inability to induce sister chromatid exchanges (SCE) in Chinese hamster ovary cells (CHOC) at 80 mg/kg culture medium; most organophosphorus insecticides tested induced SCE in CHOC at this concentration (Nishio and Uyeki 1981; Chen et al. 1982). Diazoxon, an oxygen analog of diazinon, did produce SCE at 304 mg/kg, but was 3 to 10X less effective than oxygen analogs of other organophosphorus compounds screened (Nishio and Uyeki 1981).

TERRESTRIAL INVERTEBRATES

Tobacco hornworms (*Manduca sexta*) from a field sprayed with 840 mg diazinon/ha contained no detectable residues of diazoxon. Only one sample, collected about 4 hours after spraying, exceeded 1.0 mg diazinon/kg body weight. No diazinon residues in these insects were detectable after 18 days. It was concluded that the potential hazard to birds eating hornworms was minimal (Stromborg et al. 1982). In contrast, diazinon residues in molluscan slugs (*Agriolimax reticulatus*), collected from plats of spring wheat sprayed with 8,000 mg diazinon/ha, increased linearly to about 200 mg/kg at 6 weeks postapplication, then declined to background levels after 16 weeks (Edwards 1976). During this same period, soil residues decreased from about 4 mg/kg immediately after application, to about 1 mg/kg at 6 weeks, and were not detectable after 12 weeks. The high

residues observed in slugs may be due, in part, to physical adsorption of diazinon to slug mucus. Edwards (1976) concluded that slugs heavily contaminated by diazinon constituted a serious danger to birds and mammals feeding on them.

Depuration rates of diazinon differed significantly for two species of nematodes, *Panagrellus redivivus* and *Bursaphelenchus xylophilus* (Al-Attar and Knowles 1982). Both species showed maximum uptake of radiolabeled diazinon between 6 and 12 hours, and both metabolized diazinon to diazoxon and pyrimidinol. By 96 hours, 95% of the diazinon in *P. redivivus* had been metabolized, but only 26% was transformed in *B. xylophilus*, again demonstrating variability in diazinon metabolism between related species.

RECOMMENDATIONS

Certain aquatic organisms were impacted by diazinon water concentrations between 0.3 and 1.2 ug/l; effects included lowered emergence and elevated drift of stream insects (0.3 ug/l), reduced fecundity of marine minnows (0.47 ug/l), accumulations in freshwater teleosts (0.55 ug/l), and daphnid immobilization (1.0 ug/l) and death (1.2 ug/l). For protection of sensitive aquatic organisms, Arthur et al. (1983) recommended that water diazinon levels should not exceed 0.08 ug/l. This value represents a safety factor of about 4 over the lowest recorded adverse effect level of 0.3 ug/l. The safety factor may require adjustment, probably upwards, as additional data become available. Establishment of safe levels is complicated by the fact that diazinon can persist many months in neutral or basic waters, including seawater (Kanazawa 1978), but hydrolyzes rapidly in acidic waters (Allison and Hermanutz 1977). Data on chronic effects of fluctuating and intermittent exposures of fishes and invertebrates to diazinon are also needed, and these will aid in the establishment of safe concentrations for this organophosphorus pesticide (Allison and Hermanutz 1977).

Granular formulations were especially hazardous to seed-eating birds; ingestion of fewer than 5 granules of a Diazinon 14G formulation could be lethal (Hill and Camardese 1984). A reduction in diazinon content of existing granular formulations may become necessary in application areas frequented by high densities of seed-eating birds. Stone and Gradoni (1985) recommend that diazinon should not be used in areas where waterfowl feed, especially turfgrass. Suggested alternatives to diazinon for turfgrass use include Dursban (0,0- diethyl 0-(3,5,6-trichloro-2- pyridyl)-phosphorothioate), Dylox (dimethyl (2,2,2-trichloro-l-hydroxyethyl) phosphonate), Carbaryl (1-naphthyl N-methylcarbamate), and Lannate (S-methyl-N-((methylcarbamoyl) oxy)- thioacetimidate) (Stone 1980; Stone and Gradoni 1985). Diazinon should be used with caution in large-scale spray applications--such as grasshopper control--as judged by some deaths of horned larks (*Eremophila alpestris*), lark buntings (*Calamospiza melanocorys*), western meadowlarks (*Sturnella neglecta*). and chestnut-collared longspurs (*Calcarius ornatus*) when used for this purpose in Wyoming (McEwen et al. 1972). Diazinon applications to agricultural crops comprised a relatively small percentage of the reported mortality incidents, but it is likely that this category is underreported since such incidents were probably less conspicuous than those noted on lawns and golf courses (Stone and Gradoni 1985). Also, diazinon interactions with other agricultural chemicals, such as Captan (cis -N-((trichloromethyl)thio)-4-cyclohexene- 1,2-dicarboximide), may produce more-than-additive (but reversible) adverse effects on food consumption and egg production of ring-necked pheasants (Stromborg 1977). More research is needed on complex mixtures of agricultural pesticides that contain diazinon.

In female rats, the no-observable-effect level (NOEL) is 0.1 mg/kg of dietary diazinon; at 0.5 mg/kg there was a marked lowering of plasma cholinesterase activity in 5 weeks (Davies and Holub 1980a). But studies with male rats indicate that the NOEL is 2 mg/kg of dietary diazinon, or about 20X higher than female rats (Davies and Holub 1980b). Accordingly, future studies should consider sex as a variable in toxicity evaluation of diazinon. It is generally agreed that mammals are more resistant than birds to diazinon owing, in part, to their ability to rapidly metabolize diazoxon. However, data are missing on the effects of diazinon to native mammals under field conditions, and this should constitute a priority research area.

LITERATURE CITED

Al-Attar, H.J., and C.O. Knowles. 1982. Diazinon uptake, metabolism, and elimination by nematodes. Arch. Environ. Contam. Toxicol. 11:669-673.

Allison, D.T. 1977. Use of exposure units for estimating aquatic toxicity of organophosphate pesticides. U.S. Environ. Protection Agency Rep. 600/3-77-077. 25 pp.

Allison, D.T., and R.O. Hermanutz. 1977. Toxicity of diazinon to brook trout and fathead minnows. U.S. Environ. Protection Agency Rep. 600/3-77-060. 69 pp.

Anderson, J.W., and W. Glowa. 1984. Insecticidal poisoning of honey bees in Connecticut. Environ. Entomol. 13:70-74.

Anon. 1972. Diazinon insecticide. Tech. Bull., CIBA-GEIGY, Agric. Div., Ardsley, New York.

Arthur, J.W., J.A. Zischke, K.N. Allen, and R.O. Hermanutz. 1983. Effects of diazinon on macroinvertebrates and insect emergence in outdoor experimental channels. Aquat. Toxicol. 4:283-301.

Balcomb, R., R. Stevens, and C. Bowen II. 1984. Toxicity of 16 granular insecticides to wild-caught songbirds. Bull. Environ. Contam. Toxicol. 33:302-307.

Barik, S., and D.M. Munnecke. 1982. Enzymatic hydrolysis of concentrated diazinon in soil. Bull. Environ. Contam. Toxicol. 29:235-239.

Barnett, J.B., J.M. Spyker-Cranmer, D.L. Avery, and A.M. Haberman. 1980. Immunocompetence over the lifespan of mice exposed in utero to carbofuran or diazinon: I. Changes in serum immunoglobulin concentrations. J. Environ. Pathol. Toxicol. 4(5&6):53-63.

Bartsch, E. 1974. Diazinon. II. Residues in plants, soil, and water. Residue Rev. 51:37-68.

Bennett, R.S., Jr., and H.H. Prince. 1981. Influence of agricultural pesticides on food preference and consumption by ring-necked pheasants. J. Wildl. Manage. 45:74-82.

Berg, G.L. (ed.). 1984. Farm chemicals handbook. Meister Publ. Co., Willoughby, Ohio. 501 pp.

Brack, M., and H. Rothe. 1982. Organophosphate poisoning in marmosets. Lab. Anim. 16:186-188.

Branham, B.E., and D.J. Wehner. 1985. The fate of diazinon applied to thatched turf. Agron. J. 77:101-104.

Chen, H.H., S.R. Sirianni, and C.C. Huang. 1982. Sister chromatid exchanges in Chinese hamster cells treated with seventeen organophosphorus compounds in the presence of a metabolic activation system. Environ. Mutagen. 4:621-624.

Davies, D.B., and B.J. Holub. 1980a. Toxicological evaluation of dietary diazinon in the rat. Arch. Environ. Contam. Toxicol. 9:637-650.

Davies, D.B., and B. J. Holub. 1980b. Comparative subacute toxicity of dietary diazinon in the male and female rat. Toxicol. Appl. Pharmacol. 54:359-367.

Dressel, T.D., R.L- Goodale, B. Zweker, and J.W. Borner. 1982. The effect of atropine and duct decompression on the evaluation of diazinon - induced acute canine pancreatitis. Ann. Surg. 195:424-434.

Earl, F.L., B.E. Melveger, J.E. Reinwall, G.W. Bierbower, and J.M. Curtis. 1971. Diazinon toxicity - comparative studies in dogs and miniature swine. Toxicol. Appl. Pharmacol. 18:285-295.

Eberle, D.O. 1974. Diazinon. I. Analysis of technical grade product, formulations, and residues. Residue Rev. 51:1-35.

Edwards, C.A. 1976. The uptake of two organophosphorus insecticides by slugs. Bull. Environ. Contam. Toxicol. 16:406-410.

Egyed, M.N., M. Malkinson, A. Eilat, and A. Shlosberg. 1974. Basudin (diazinon) poisoning in goslings. Refuah Veterin. 31:22-26.

Egyed, M.N., A. Shlosberg, M. Malkinson, and A. Eilat. 1976. Some considerations in the treatment of diazinon poisoned goslings. Clin. Toxicol. 9:245-249.

Fleming, W.J., and S.P. Bradbury. 1981. Recovery of cholinesterase activity in mallard ducklings administered organophosphorus pesticides. J. Toxicol. Environ. Health 8:885-897.

Forrest, M., K.A. Lord, N. Walker, and H.C. Woodville. 1981. The influence of soil treatments on the bacterial degradation of diazinon and other organophosphorus insecticides. Environ. Pollut. 24A:93-104.

Fujii, Y., and S. Asaka. 1982. Metabolism of diazinon and diazoxon in fish liver preparations. Bull. Environ. Contam. Toxicol. 29:453-460.

Goodman, L.R., D.J. Hansen, D.L. Coppage, J.C. Moore, and E. Matthews. 1979. Diazinon: chronic toxicity to, and brain acetylcholinesterase inhibition in, the sheepshead minnow, *Cyprinodon variegatus.* Trans. Am. Fish. Soc. 108:479-488.

Grue, C.E., W.J. Fleming, D.G. Bushy, and E.F. Hill. 1983. Assessing hazards of organophosphate insecticides to wildlife. Trans. North Am. Wildl. Nat. Res. Conf. 48:200-220.

Henderson, M., and P.A. Kitos. 1982. Do organophosphate insecticides inhibit the conversion of tryptophan to NAD+ in ovo? Teratology 26:173-181.

Hill, E.F., and M.B. Camardese. 1984. Toxicity of anticholinesterase insecticides to birds: technical grade versus granular formulations. Ecotoxicol. Environ. Safety 8:551-563.

Hill, E.F., and M.B. Camardese. 1986. Lethal dietary toxicities of environmental contaminants and pesticides to coturnix. U.S. Fish Wildl. Serv. Tech. Rep. 2. 147 pp.

Hill, E.F., M.B. Camardese, G. H. Heinz, J.W. Spann, and A.B. DeBevec. 1984. Acute toxicity of diazinon is similar for eight stocks of bobwhite. Environ. Toxicol. Chem. 3:61-66.

Hoffman, D.J., and W.C. Eastin, Jr. 1981. Effects of malathion, diazinon, and parathion on mallard embryo development and cholinesterase activity. Environ. Res. 26:472-485.

Hogan, J.W., and C.O. Knowles. 1972. Metabolism of diazinon by fish liver microsomes. Bull. Environ. Contam. Toxicol. 8:61-64.

Hudson, R.H., R.K. Tucker, and M.A. Haegele. 1984. Handbook of toxicity of pesticides to wildlife. U.S. Fish Wildl. Serv. Resour. Publ. 153. 90 pp.

Iverson, F., D.L. Grant and J. Lacroix. 1975. Diazinon metabolism in the dog. Bull. Environ. Contam. Toxicol. 13:611-618.

Jarvinen, A.W., and D.K. Tanner. 1982. Toxicity of selected controlled release and corresponding unformulated technical grade pesticides to the fathead minnow *Pimephales promelas*. Environ. Pollut. 27A:179-195.

Johnson, W.W., and M.T. Finley. 1980. Handbook of acute toxicity of chemicals to fish and aquatic invertebrates. U.S. Fish Wildl. Serv. Resour. Publ. 137. 98 pp.

Kanazawa, J. 1978. Bioconcentration ratio of diazinon by freshwater fish and snail. Bull. Environ. Contam. Toxicol- 20:613-617.

Kikuchi, H., Y. Suzuki, and Y. Hashimoto. 1981. Increase of B-glucuronidase activity in the serum of rats administered organophosphate and carbamate insecticides. J. Toxicol. Sci. 6:27-35.

Kuhr, R.J., and H. Tashiro. 1978. Distribution and persistence of chlorpyrifos and diazinon applied to turf. Bull. Environ. Contam. Toxicol. 20:652-656.

Lox, C.D. 1983. Effects of acute pesticide poisoning on blood clotting in the rat. Ecotoxicol. Environ. Safety 7:451-454.

Machin, A.F., P.H. Anderson, and C.N. Hebert. 1974. Residue levels and cholinesterase activities in sheep poisoned experimentally with diazinon. Pestic. Sci. 5:49-56.

Machin, A.F., H. Rogers, A.J. Cross, M.P. Quick, L.C. Howells, and N.F. Janes. 1975. Metabolic aspects of the toxicology of diazinon. I. Hepatic metabolism in the sheep, cow, pig, guinea-pig, rat, turkey, chicken and duck. Pestic. Sci. 6:461-473.

McEwen, L.C., C.E. Knittle, and M.L. Richmond. 1972. Wildlife effects from grasshopper insecticides sprayed on short-grass range. J. Range Manage. 25:188-195.

McLean, S., M. Sameshina, T. Katayama, J. Iwata, C.E. Olney, and K.L. Simpson. 1984. A rapid method to determine microsomal metabolism of organophosphate pesticides. Bull. Jpn. Soc. Sci. Fish. 50:1419-1423.

Meier, E.P., M.C. Warner, W.H. Dennis, W.F. Randall, and T.A. Miller. 1976. Chemical degradation of military standard formulations of organophosphate and carbamate pesticides. I. Chemical hydrolysis of diazinon. U.S. Army Med. Bioengin. Res. Dev. Lab., Fort Detrick, Frederick, MD. Tech. Rep. 7611. 32 pp.

Menzie, C.M. 1978. Metabolism of pesticides. Update II. U.S. Fish Wild. Serv. Spec. Sci. Rep. - Wildl. No. 212. 381 pp.

Misawa, M., J. Doull, P.A. Kitos, and E.M. Uyeki. 1981. Teratogenic effects of cholinergic insecticides in chick embryos. I. Diazinon treatment on acetylcholinesterase and choline acetyltransferase activities. Toxicol. Appl. Pharmacol. 57:20-29.

Misawa, M., J. Doull, and E.M. Uyeki. 1982. Teratogenic effects of cholinergic insecticides in chick embryo-T. III. Development of cartilage and bone. J. Toxicol. Environ. Health 10:551-563.

Montz, W.E., Jr. 1983. Effects of organophosphate insecticides on aspects of reproduction and survival in small mammals. Ph.D. Thesis. Virginia Polytechnic Institute and State University, Blacksburg. 176 pp.

Montz, W.E., Jr., and R.L. Kirkpatrick. 1985. Temporal patterns of brain cholinesterase activities of white-footed mice (*Peromyscus leucopus*) following dosing with diazinon or parathion. Arch. Environ. Contam. Toxicol. 14:19-24.

Murphy, S.D. 1975. Pesticides. Pages 408-453 *in* L.J. Casarett and J. Doull (eds.). Toxicology: the basic science of poisons. Macmillan Publ. Co., New York.

Negherbon, W.O. 1959. Handbook of toxicology. Volume III. Insecticides, a compendium. Natl. Acad. Sci., Natl. Res. Coun., Div. Biol. Agric., WADC Tech. Rep. 55-16:252-261.

Nimmo, D.R., T.L. Hamaker, E. Matthews, and J.C. Moore. 1981. An overview of the acute and chronic effects of first and second generation pesticides on an estuarine mysid. Pages 3-19 *in* J. Vernberg, A. Calabrese, F.P. Thurberg and W.B. Vernberg (eds.). Biological monitoring of marine pollutants. Academic Press, New York.

Nishio, A., and E.M. Uyeki. 1981. Induction of sister chromatid exchanges in Chinese hamster ovary cells by organophosphate insecticides and their oxygen analogs. J. Toxicol. Environ. Health 8:939-946.

Sastry, K.V., and P.V. Malik. 1982. Acute and chronic effects of diazinon on the activities of three dehydrogenases in the digestive system of a freshwater teleost fish *Channa punctatus*. Toxicol. Lett. 10:55-59.

Sastry, K. V., and K. Sharma. 1980. Diazinon effect on the activities of brain enzymes from *Ophiocephalus* (*Channa*) *punctatus*. Bull. Environ. Contam. Toxicol. 24:326-332.

Schafer, E.W., Jr., W.A. Bowles, Jr., and J. Hurlbert. 1983. The acute oral toxicity, repellency, and hazard potential of 998 chemicals to one or more species of wild and domestic birds. Arch. Environ. Contam. Toxicol. 12:355-382.

Schobert, E.E. 1974. Ingested insecticide poisoning in waterfowl. J. Zoo Anim. Med. 5:20.

Seguchi, K., and S. Asaka. 1981. Intake and excretion of diazinon in freshwater fishes. Bull. Environ. Contam. Toxicol. 27:244-249.

Seikai, T. 1982. Acute toxicity of organophosphorus insecticides on the developmental stages of eggs, larvae and juveniles of Japanese striped knifejaw *Oplegnathus fasciatus*. Bull. Jpn. Soc. Sci. Fish. 48:599-603.

Shacklock, P.F., and G.B. Croft. 1981. Effect of grazers on *Chondrus crispus* in culture. Aquaculture 22:331-342.

Skinner, C.S., and W. W. Kilgore. 1982. Acute dermal toxicities of various organophosphate insecticides in mice. J. Toxicol. Environ. Health 9:491-497.

Sokkar, S.M., M. A. Mohammed, and F.A. Soliman. 1975. Toxic effects of diazinon on the gonads of fowl. Zbl. Vet. Med. 22A:557-563.

Soliman, S.A., G.W. Sovocool, A. Curley, N.S. Ahmed, S. El-Fiki, and A-K. El-Sebae. 1982. Two acute human poisoning cases resulting from exposure to diazinon transformation products in Egypt. Arch. Environ. Health 37:207-212.

Spyker, J.M., and D.L. Avery. 1977. Neurobehavioral effects of prenatal exposure to the organophosphate diazinon in mice. J. Toxicol. Environ. Health 3:989-1002.

Stadnyk, L., and R..S. Campbell. 1971. Pesticide effect on growth and assimilation in a freshwater alga. Bull. Environ. Contam. Toxicol. 6:1-8.

Stone, W.B. 1980. Bird deaths caused by pesticides used on turfgrass. N.Y. State Turfgrass Conf. Proc. 4:58-62.

Stone, W.B., and P.B. Gradoni. 1985. Wildlife mortality related to use of the pesticide diazinon. Northeast. Environ. Sci. 4:30-38.

Stone, W.B., and H. Knoch. 1982. American brant killed on golf courses by diazinon. N.Y. Fish Game J. 29:95-96.

Stratton, G.W., and C.T. Corke. 1981. Interaction of permethrin with *Daphnia magna* in the presence and absence of particulate material. Environ. Pollut. 24A:135-144.

Stromborg, K.L. 1977. Seed treatment pesticide effects on pheasant reproduction at sublethal doses. J. Wildl. Manage. 41:632-642.

Stromborg, K.L. 1979. Pheasant food habits in spring and consumption of seed treatment pesticides. J. Wildl. Manage. 43:214-219.

Stromborg, K.L. 1981. Reproductive tests of diazinon on bobwhite quail. Pages 19-30 *in* P.W. Lamb and E.E. Kenaga (eds.). Avian and mammalian wildlife toxicology: second conference. ASTM STP 757, American Society for Testing and Materials. Philadelphia.

Stromborg, K.L., W.N. Beyer, and E. Kolbe. 1982. Diazinon residues in insects from sprayed tobacco. Pages 93-97 *in* Chemistry in ecology, Vol. 1. Gordon and Breach Science Publishers, Great Britain.

Tomokuni, K., and T. Hasegawa. 1985. Diazinon concentrations and blood cholinesterase activities in rats exposed to diazinon. Toxicol. Lett. 25:7-10.

Vigfusson, N.V., E.R. Vyse, C.A. Pernsteiner, and R.J. Dawson. 1983. In vivo induction of sister-chromatid exchange in *Umbra limi* by the insecticides endrin, chlordane, diazinon and guthion. Mutat. Res. 118:61-68.

Wahla, M.A., G. Gibbs, and J.B. Ford. 1976. Diazinon poisoning in large white butterfly larvae and the influence of sesamex and piperonyl butoxide. Pestic. Sci. 7:367-371.

Wilkinson, F.C. 1980. The uptake of dipping fluid and persistence of diazinon on shower-dipped sheep. Aust. Veterin. J. 56:561-562.

Wyttenbach, C.R., and J.D. Hwang. 1984. Relationship between insecticide-induced short and wry neck and cervical defects visible histologically shortly after treatment of chick embryos. J. Exp. Zool. 229:437-446.

Zinkl, J.G., J. Rathert, and R.R. Hudson. 1978. Diazinon poisoning in wild Canada geese. J. Wildl. Manage. 42:406-408.

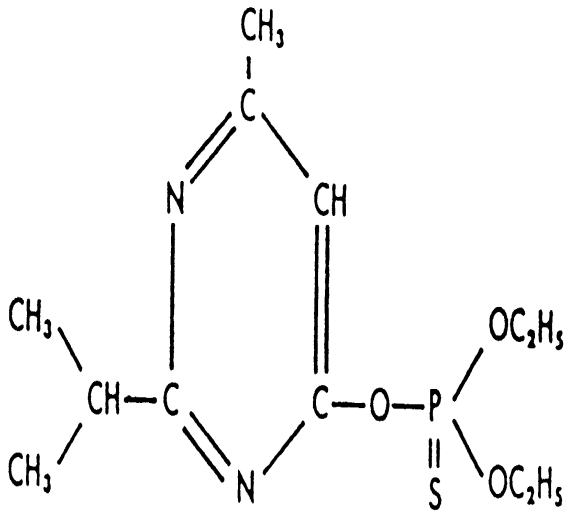

Figure 1. Structural formula of diazinon.