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A REVIEW OF THE LITERATURE ON THE USE OF 2, 4-D  
IN FISHERIES

BUREAU OF SPORT FISHERIES AND WILDLIFE

MARCH 1974

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The herbicide 2,4-D [2,4-(dichlorophenoxy)acetic acid] was adapted from terrestrial to aquatic use. Recommended treatment rates vary from 2.24 kg/ha or even higher for submersed species. Many formulations of 2,4-D are available, but the one used most commonly is the dimethylamine salt of 2,4-D (DMA-2,4-D). The ester formulations are also used but are 10 to 20 times as toxic to fish and other aquatic organisms as the dimethylamine salt. A tolerance of 0.1 mg/liter has been issued for DMA-2,4-D that occurs in potable water as a result of applications of DMA-2,4-D to ditch banks in the western United States. Several Federal agencies are presently pursuing the registration of 2,4-D for use on irrigation canal banks and for use in moving water. Since most of the research necessary for registration of 2,4-D has been done, it should be relatively inexpensive to complete the registration procedure.

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(Complete review of literature concerning 2,4-D's use in fisheries)

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A Review of the Literature on the Use of  
2,4-D in Fisheries

by

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## ABSTRACT

The herbicide 2,4-D [2,4-(dichlorophenoxy)acetic acid] was adapted from terrestrial to aquatic use. Recommended treatment rates vary from 2.24 kg/ha to 22.4 kg/ha or even higher for submersed species.

Many formulations of 2,4-D are available, but the one used most commonly is the dimethylamine salt of 2,4-D (DMA-2,4-D). The ester formulations are also used but are 10 to 20 times as toxic to fish and other aquatic organisms as the dimethylamine salt.

A tolerance of 0.1 mg/liter has been issued for DMA-2,4-D that occurs in potable water as a result of applications of DMA-2,4-D to ditch banks in the western United States. Several Federal agencies are presently pursuing the registration of 2,4-D for use on irrigation canal banks and for use in moving water. Since most of the research necessary for registration of 2,4-D has been done, it should be relatively inexpensive to complete the registration procedure.

## HISTORY OF FISH CULTURAL USE

Research on 2,4-D compounds started during World War II, under war-time secrecy. The chemical has a growth-hormone effect at low concentrations (less than one ug/liter) and a herbicidal effect at concentrations of 0.1 mg/liter and greater.

The first recorded aquatic use of 2,4-D was to control waterhyacinth (Hildebrand, 1946). Several years later, Surber (1949) reported that 2,4-D was effective for control of emerged and marginal weeds. Since then, it has been used to control Elodea canadensis (Foret, 1967), Eurasian watermilfoil (Haven, 1963; Smith and Isom, 1967; Whitney, 1970; Wojtalik, et al., 1971) and Hydrilla verticillata (Kleinschmidt, 1969). It also has been tried as a control for Pithophora sp., alligatorweed, duckweed, waterstar grass (Lawrence, 1962), Nymphaea odorata (Pierce, 1960), Brasenia spp. (Pierce, 1961), and, in combination with Endothal, Potamogeton crispus (Pierce, 1969). Martin et al. (1957) noted its use in improving duck marsh habitat. Frank et al. (1963) reported that 2,4-D was useful for controlling water lily, lotus, spatterdock, and penny wort in irrigation canals but would not control Sago pondweed or American pond weed due to its lack of persistence in the soil. The latter authors reported that



2,4-D was somewhat effective for control of the following marginal weeds along irrigation canals: willow, sedge, smartweed, cattail, arrowhead, pickerel weed, bulrush, spikerush, knotgrass, southern watergrass, and needlerush. Bruns (1964, 1967) and Bruns and Clore (1958) investigated possible damage to crops from irrigation water treated with 2,4-D, while DeVaney (1967) reported efficacious dosages for weeds in farm ponds and lakes.

## PHYSICAL AND CHEMICAL PROPERTIES

The herbicide 2,4-D [2,4-(dichlorophenoxy)acetic acid] has a molecular formula of  $C_8H_6Cl_2O_3$ , a molecular weight of 221.0, a specific gravity,  $G$ , of 1.565 (30° C), a melting point of 135-138° C, a boiling point of 160° C at 0.4 mm Hg, and a vapor pressure of 0.4 mm Hg at 160° C (WSSA, 1970). It is a white, crystalline, odorless substance, and is soluble in many organic solvents but quite insoluble in water. Nelson and Faust (1969) determined the acid dissociation constant of 2,4-D by means of potentiometric titration. They found a  $pK_a$  value of 2.73 at an ionic strength of 0.05 for 2,4-D.

The acid is prepared from 2,4-dichlorophenol and monochloroacetic acid in the presence of aqueous base. An aqueous solution of the dimethylamine salt (DMA) is prepared by addition of 40% dimethylamine solution to an aqueous slurry of 2,4-D at pH 8. The butoxyethanol ester is synthesized by the reaction of the free acid of 2,4-D with butoxyethanol (WSSA, 1970).

The free acid may be determined by infrared spectroscopy. Information on analytical methods is contained in the Residue Section of this report.

The teratogenic compound tetrachlorodioxin (dioxin) is found in small amounts in commercially prepared 2,4,5-trichlorophenoxyacetic acid, hence, analyses have been conducted on commercial preparations of 2,4-D. Huston (1972) reported three neutral impurities in commercially prepared 2,4-D which could interfere with gas-liquid chromatographic (glc) analyses for dioxins. She concluded that these contaminants were not dioxins nor deleterious. Plimmer and Klingebiel (1971) concluded that under field conditions, dioxins are unlikely products of the lower chlorinated phenols or phenoxyalkanoic acids.

Several formulations of 2,4-D are in common use. These include: water-soluble salts such as sodium, dimethylamine, ethanolamine, and triethanolamine; oil-soluble amine salts, such as the dodecyl-tetradecyl amine salt and the N-oley-1,3-propylenediamine salt which are soluble in most organic solvents; and esters, such as the iso-octyl, butoxyethanol, and the propyleneglycolbutylether ester.

The formulation most commonly used in aquatic sites is the dimethylamine salt of 2,4-D (DMA-2,4-D). The molecular formula of DMA-2,4-D is  $C_{10}H_{13}Cl_2NO_3$ ; the molecular weight is 266.1; and

the melting point is 85 to 87° C at which temperature it decomposes. DMA-2,4-D is a white, odorless crystal which is highly soluble in water, less soluble in alcohols and acetone, and insoluble in kerosene and diesel oil. It has been reported (Bridges and Sanders, 1963) that DMA-2,4-D will diffuse through polyethylene bags placed in water.

#### Effects on Organisms

Microorganisms and algae. Lamartiniere et al. (1964) reported that 2,4-D affected cross wall formation and cell division in several bacterial species. They found a rapid cellular death in the stationary phase of growth. Butler (1963) noted no decrease in productivity of natural phytoplankton or unialgal cultures of Dunella euchlora or Platymonas sp. during a 4-hour exposure to a 1-mg/liter concentration of DMA-2,4-D. Poorman (1973) found that 24 hours' exposure to 1- to 10-mg/liter concentrations of 2,4-D had no effect on the growth of Euglena gracilis. They also reported no effect after 7 days' exposure to 10 or 50 mg/liter but there was a 26% decrease in growth of Euglena when they were exposed to 100 mg/liter for 24 hours. However, when these cells were diluted and transferred to pesticide-free medium, they recovered.

When the freshwater alga, Scenedesmus quadricaudata, was exposed to 0.1 or 1.0 mg/liter of 2,4-D there was a decrease in cell density at 4-8 days, some indications of carbon fixation stimulation beginning on day 6, and a small reduction in biomass by day 10 (Stadnyk et al., 1971). Walsh et al. (1970) reported that concentrations of 0.1, 1.0, or 10.0 mg/liter BEE-2,4-D did not alter the rate of photosynthesis of a species of unicellular marine alga.

Plants. Lynn and Barrows (1952) found no increase in hydrocyanic acid content of wild pin cherry leaves sprayed with an ester of 2,4-D (see Willard, 1950). Buck et al. (1961) also reported no increase in hydrocyanic acid in Canadian thistle sprayed with 2,4-D. They found that calves and ewes ate the sprayed, dried plants with no ill effects. Williams (1968) found that spraying spring parsley with 2,4-D detoxified the plants, thereby decreasing the plants capacity to photosensitize chickens.

Insects. Adams (1960) found that coccinellid larvae were susceptible to an amine salt of 2,4-D. She stated that the developmental period was lengthened when older age groups were sprayed and that mortality was twice as great in sprayed larvae as in controls up to pupation, although mortality during pupation was no greater in

sprayed than control larvae. Rexrode et al. (1971) reported that trees treated with herbicides were more subject to attack by oak beetles and that beetle numbers were higher on treated trees. However, trees treated with DMA-2,4-D had less fungal mat than those treated with picloram and 2,4-D or picloram and arsenic.

Maxwell and Harwood (1960) reported an increased rate of reproduction by pea aphids fed on broad beans sprayed with non-lethal doses of 2,4-D. This increased rate may have been due to increased amounts of free alanine, aspartic acid, serine, and glutathion in the growth terminals of the bean plant, the area of greatest aphid development.

Aquatic organisms. Butler (1963) found that 5 mg/liter of DMA-2,4-D irritated juvenile blue crabs after 24 and 48 hours of testing but no decrease in oyster shell growth occurred after treatment at 2 mg/liter for 48 hours. When ponds were treated at 5 and 10 mg/liter of 2,4-D, there was a 2-week delay in bluegill spawning (Cope, et al., 1970). Higher treatment levels induced some pathology including depletion of liver glycogen, globular deposits in the blood vessels and stasis and engorgement of the brain circulatory system.

Birds. Whitehead (1973) reported that growth rates of 0- to 8-week old broilers decreased significantly when fed 10, 50, or 100 mg/kg 2,4-D but that food conversion was not affected by any dietary level. Whitehead and Pettigrew (1972) noted a reduction in food consumption and growth levels in chicks fed 250-900 mg/kg dietary 2,4-D. They found that 5,000 mg/kg caused histological changes, but the chicks resumed normal growth when placed on a normal diet. No effect was noticed on plasma calcium or magnesium. Somers et al. (1973) sprayed fertile chick eggs at the recommended and also at 10 and 20 times the recommended application rate. They found no effect on hatching or growth of the chicks. Chickens tolerated 300 mg/kg/day for several weeks without ill effects (Bjorklund and Erne, 1966). The same workers reported that the most noticeable effect was reduced egg production when chickens were given 500 mg/kg in feed or 1,000 mg/liter in water for prolonged periods. In contrast, Lutz-Ostertag and Lutz (1970) reported 40-75% mortalities in eggs of pheasants, gray partridge, and red partridge sprayed with 2,4-D. They also found fetal malformations, reduced fertility, and physiological castration. Sheldon et al. (1964) fed geese 1,000 mg/kg of  $^{14}\text{C}$ -2,4-D and sacrificed geese at intervals from 22 to 230 days. Treated geese gained less than control geese and also had disorganized cellular structure, hepatic cell destruction, and fatty

degeneration in the liver. Two geese treated for 192 days were placed on a control diet and had almost completely recovered after 7 months.

**Mammals.** When Collins and Williams (1971) fed hamsters 20-100 mg/kg daily they found occasional terata, decreased fetal viability, and decreased litter size. However, these anomalies were not dose-related.

Brody (1973) induced myotonia in rat muscles in 45 minutes by injecting 200 mg/kg body weight of 2,4-D. The animals did not appear ill and the myotonia disappeared within 24 hours. He postulated that 2,4-D caused an increase in  $K^+$  conductance which could lead to myotonia through a compensatory decrease in  $Cl^-$  conductance.

Phillipo and Fang (1967) reported that control rats fed  $^{14}C$ -acetate converted more than 90% of it to  $^{14}CO_2$ . When rats were fed 10-20 mg/kg 2,4-D the effect on respired  $^{14}CO_2$  was insignificant, however, a dose of 400 mg/kg decreased the amount of respired  $^{14}CO_2$  and affected the pathway of  $^{14}CO_2$  elimination. Heene (1967) found that 2,4-D inhibited glycogen forming enzymes in rat skeletal muscle. Khera and McKinley (1972) noted fetopathy and an increased incidence of skeletal anomalies following daily per os doses of 100-150 mg 2,4-D/kg. However, weight gain and viability of the offspring were within control limits. Schwetz et al. (1971) administered



doses of 2,4-D esters to pregnant rats 6-15 days after conception. The maximum tabulated dose was 87.5 mg/kg/day. High dose levels resulted in decreased fetal body weight, subcutaneous edema, delayed bone ossification, and formation of lumbar and wavy ribs, all of which were probably dose-related. They found no teratogenic effects at any dose, nor any effect on fertility, gestation, viability, or lactation. Also, neonatal growth and development were not altered by treatment during pregnancy. Rats tolerated 100 mg 2,4-D/kg without ill effect (Bjorklund and Erne, 1966). The same workers also found that 50 mg/kg/day could be toxic to pigs. Pigs fed 500 mg/kg for up to 12 months suffered growth depression, locomotor disturbances, anemia, albuminuria, and moderate hepatic and renal degeneration. A sow fed 2,4-D through the gestation period showed no ill effects, but the piglets were underdeveloped and 10 to 15 of them died within 24 hours of birth. There was retarded growth and increased mortality in the second generation when pregnant rats and their offspring were given 1,000 mg/kg 2,4-D for 10 months. Hansen et al. (1971) fed rats up to 250 mg/kg 2,4-D in the diet for 2 years. They found no significant effect on growth rate, survival rate, organ weights, or hematologic values. No 2,4-D related effects were found in beagles fed up to 500 mg/kg dietary levels for 2 years. In a

three-generation rat litter reproduction study, no deleterious effect was noted from 100 or 500 mg/kg dietary treatment, and, while 1,500 mg/kg did not affect fertility nor litter size, it reduced the percentage of pups surviving to weaning and the weights of weanlings.

Sheep given 100 mg/kg of the alkanolamine salt of 2,4-D were unaffected after 481 doses (Palmer and Radeleff, 1964). Cattle were unaffected by 112 doses or 50 mg/kg but died after 44 doses of 200 mg/kg. Sheep also were unaffected by 481 doses of 100 mg/kg of an ester of 2,4-D while cattle suffered weight loss when given 10 doses of 250 mg/kg of the same ester. Radeleff (1964) stated that 2,4-D was absorbed from the digestive tract and eliminated by the kidneys in mammals.

Walker et al. (1972) reported that 2,4-D inhibited tumor development in Ehrlich ascites. They further noted an increased survival time for tumorigenic mice treated with 2,4-D.

Humans. Sare (1972) reported a case of headaches and double vision in a worker spraying 2,4-D. Berkley and Magee (1962) reported the case of a farmer who had cutaneous exposure and had possibly inhaled DMA-2,4-D. He stated he had tingling of the hands and feet, aching arms, and stiffness in his hands and

knees. His dexterity was so reduced he could not button his shirt or tie his shoes. He also had reduced perception to painful stimuli in the distal extremities. The authors concluded that neuropathy from 2,4-D exposure is rare and those who show it are probably predisposed to neuropathy or susceptibility to the toxin. Another source (Anonymous, 1956) also mentions possible sensitivity of certain individuals to phenolic compounds which are contained in some formulations. Willard (1950) also mentioned the possible allergy of some people to 2,4-D. Berwick (1970) reported an accidental ingestion of 7,200 mg 2,4-D which resulted in fibrillary twitching and paralysis of the intercostal muscles. Bonderman et al. (1971) studied the adaptive responses of some esterase enzymes and found no significant difference in the esterase levels between people who had formulated herbicides for up to 20 years versus controls. Poland et al. (1971) studied 73 male employees in a factory where 2,4-D was manufactured and found chloracne in 18% of those studied. However, the incidence of chloracne was not correlated with job location in the plant. Johnson (1971) noted no genetic effects on 220 men exposed to 30-40 mg 2,4-D/day. Hayes (1971) stated that the oral dose of 2,4-D required to produce illness in man is probably 3-4 g. An intravenous dose of 2.0 g.

produced no illness, but 3.6 g produced coma, fibrillary twitching of some muscles, hyporeflexia, and urinary incontinence. Mammals killed by large doses of 2,4-D are thought to die of ventricular fibrillation. Treatment for 2,4-D poisoning is symptomatic.

### Stability and Degradation

Aly and Faust (1964) decomposed 2,4-D with a UV lamp. They found that decomposition was more rapid at pH 9 than pH 7. They did not feel that UV from sunlight would decompose the herbicide. Daly (1971) found that the butoxyethanol ester of 2,4-D (BEE-2,4-D) and also 2,4-D degraded rapidly under conditions of intense light and high temperature. He found that UV light degraded 50% of a solution of BEE-2,4-D to volatile products in 12 hours.

Crosby (1969), Crosby and Tutass (1966), Crosby and Li (1969) and Crosby and Wond (1973) found that aqueous solutions of 2,4-D are photolyzed through a series of reactive intermediates with replacement of the chlorines by hydroxyl groups and cleavage of the ether bond. Among the photolytic products are 2,4-dichlorophenol, 4-chlorocatechol, 2-hydroxy-4-chlorophenoxyacetic acid, and 1,2,4-benzenetriol. The latter is then converted via a non-photochemical oxidation to a nontoxic polymeric humic acid.

Crosby and Wong (1973) reported that side-chain degradation was oxygen dependent and was more rapid at pH 8 than at pH 2, but that ring reactions did not require oxygen. They further stated that the displacement of chloride from the photo-excited ring by hydroxide ions was pH dependent and the final degradation products (condensation products) were the result of ionic and oxidative combinations with substrates.

## EFFICACY

DMA-2,4-D is recommended for use primarily on broad-leaved plants. The primary aquatic-related uses are for control of weeds on irrigation canal banks, control of waterhyacinth, and control of Eurasian water milfoil. It is also used to control other aquatic plants such as lotus, arrowhead, waterlily, and smartweed, but on a lesser scale. Treatment should be carried out when plants are young and actively growing before the bud or early bloom stage (WSSA, 1970; Corns and Gupta, 1971; DeRigo, 1964; DeVaney, 1967; Thomas and Duffy, 1968). General aquatic application for emerged plants is 2.2 kg/ha in 400-600 liters of water. A "sticking" agent is sometimes employed. Normally, no more than two applications should be made per year except for possible spot treatment. Grover et al. (1972) found that 3-4% of DMA-2,4-D and the butylester drifted off the target area as droplets and an additional 25-30% of the butylester drifted off as a vapor mass within 30 minutes after spraying. Therefore, no treatment should be made when wind velocity exceeds 10 mph or under rainy conditions. Treatment along ditch banks or other moving water should be done in an upstream direction to avoid large concentrations of the chemical. When a body of

water is nearly covered with an undesirable plant, only part of the water should be treated at one time to avoid the possibility of BOD problems from the decomposing vegetation.

There are conflicting reports regarding the disappearance of 2,4-D from water. Aly and Faust (1964) reported that 2,4-D persisted for 120 days in lake water that was aerobically incubated in the laboratory, whereas esters of 2,4-D under similar treatment were hydrolyzed biologically to the acid in 9 days. They further reported that 81-85% of the 2,4-D was decomposed biologically in lake muds within 24 hours but only after extensive microbial adaptation techniques. Faust and Suffett (1966) verified these results. However, Daly (1971) reported that BEE-2,4-D degraded to 2,4-D within 24 hours in systems containing living material, pond water, polluted water, or water with watermilfoil. All systems produced  $^{14}\text{CO}_2$ , but polluted water produced the most.

Rogoff (1961) discussed the oxidation by bacteria of aromatic compounds including 2,4-D. Anderson et al. (1968), utilizing 2,4-D as the sole carbon source, isolated a strain of Aerobacter sp. which decomposed 90% of the 2,4-D in 15 days. Bell (1957, 1960) isolated an Achromobacter species which could decompose 2,4-D. He reported that 99% of the chlorine was released as chloride. He further stated that requirements for rapid oxidation

included a free ortho position, two or less chlorine substitutions on the ring, a para chlorine substitution, and a free side-chain carboxyl group beta to the ether linkage. Others (Robeck et al., 1963; Steenson and Walker, 1956 and 1958; Rogoff and Reid, 1954) have reported degradation of 2,4-D by microorganisms in the genera Flavobacterium, Achromobacter, and Coxyne. Aerobic conditions were found to be essential, and degradation was attributed to the formation of adaptive enzymes. Walker and Newman (1956) reported that a culture of Mycoplasma sp. decomposed 2,4-D, while Medemeyer (1966) found a protozoan and paramecium which could metabolize 2,4-D.

Linscott et al. (1968) stated that the resistance of alfalfa to 2,4-dichlorophenoxybutyric acid (2,4-DB) may result from the synthesis of inactive chlorophenoxy compounds having larger side chains than the parent compound, thus preventing beta-oxidation to the toxic parent chemical.

Faust et al. (1961) reported objectionable odors in water treated with 2,4-D, and concluded the odors resulted from production of phenols from the herbicide. Faust and Aly (1963) reported that 2,4-dichlorophenol (2,4-DCP) was metabolized slowly under either acid or anaerobic conditions. Activated carbon was capable of removing 2,4-D and 2,4-DCP from water, whereas



strongly basic anion-exchange resins sorbed 2,4-DCP and Na-2,4-D but not the esters of 2,4-D (Faust and Suffet, 1966). Inglis and Davis (1968) found that hardnesses ranging from 13-365 mg/liter  $\text{CaCO}_3$  had no significant effect on the toxicity of organic herbicides to fish.

A brief summary of the efficacy of 2,4-D is listed in Table 1. The data are primarily related to aquatic plants. Exceptions are where the applications may have had an effect on vegetable crops (via irrigation) or effects on wildlife. Most emerged broad-leaved aquatic plants are controlled by 2,4-D. Aquatic grasses and submersed species are not controlled or require massive doses of the herbicide for control.

## TOXICITY

The toxicity of 2,4-D depends greatly on the formulation. For example, Sanders (1970) reported the following TL<sub>50</sub> values at 48 hours for Daphnia magna: propyleneglycobutyl ester, 0.1 mg/liter; DMA-2,4-D, 4 mg/liter; and BEE-2,4-D, 5.6 mg/liter.

Lawrence (1962b) reported that 80% of the largemouth bass exposed to 1 mg/liter DMA-2,4-D died in 72 hours. On the other extreme, Stickel (1964) reported an LD<sub>50</sub> of 56,776 mg/kg at 94 days for adult Coturnix quail.

Table 2 is a compilation of toxicity data in which specific amounts of the herbicide and specific mortalities were reported by various authors. The remainder of the toxicity data is summarized on the following pages.

Plants. Elder et al. (1970) reported that 2,4-D exhibited low toxicity to all freshwater and marine algal organisms tested at rates approaching the maximum solubility of the herbicide in water. DeRigo (1964) found that 2,4-D injured commercially grown Chinese waterchestnut. A single annual application of 6.7 kg 2,4-D/ha in diesel fuel was found to be effective for

cattail control with no deleterious effects on the biological activity of sewage lagoons from repeated applications (Corns and Gupta, 1971). Kleinschmidt (1969) found that 2,4-D gave good control of Hydrilla verticillata while others (Daly, 1971; Haven, 1963; Smith and Isom, 1967; Wojtalik et al., 1971) reported control of Eurasian watermilfoil with DMA-2,4-D or BEE-2,4-D. Hildebrand (1946), Schultz and Harman (1974), and Schultz and Whitney (1974) reported that waterhyacinth could be controlled by 2,4-D. Lapham (1964) controlled alligatorweed with a combination of 2,4-D and dichlobenil while Foret (1967) used 2,4-D to control Elodea canadensis. Pierce (1960, 1961) found that 2,4-D eliminated Nymphaea odorata and Brasenia sp., reduced Utricularia purpurea, but seemed to accelerate growth of Potamogeton spp. She also reported that 2,4-D in combination with other herbicides was toxic to Potamogeton crispus and several other aquatic plants (1968, 1969).

Several authors (USDA, 1968; Lawrence, 1962a) have compiled references dealing with the toxicity of herbicides to aquatic plants while others (DeVaney, 1967; Lawrence, 1962b; Mullison, 1970; Gangstad, 1972; and Pimentel, 1971) have reviewed the toxicity of 2,4-D to higher aquatic plants, algae, fish, and non-target organisms.

The toxicity of 2,4-D to terrestrial plants is well known and too lengthy to document. Keith et al. (1959) reported a decrease in pocket gophers due to the reduction of food plants such as dandelion, agoseris, western yarrow, and penstemon. Krefting and Hansen (1963) improved deer habitat by top-killing mountain maple with 2,4-D. The herbicide also increased conifer sprouts and stimulated regrowth of maple sprouts, both of which are excellent deer browse. Hee et al. (1973) found that the oil-soluble amine formulations were absorbed faster by sunflower leaves than the water-soluble amines, thereby decreasing the length of time for death of the plant.

**Microorganisms.** Petruk (1965) added 2,4-D to fish nursery ponds and found an increase in microorganisms in 2 to 3 days. The total numbers decreased after 12 days, but reached the initial value again in 23 to 35 days. He stated that the herbicide stimulated heterotrophic organisms which increased the reservoir productivity.

**Invertebrates.** Butler (1965) found no effect on the growth of oyster shell after 96 hours' exposure to 2 mg/liter of 2,4-D. He also noted no effect of 2 mg/liter on brown shrimp after 24 hours' exposure. In static bioassays, 10 mg/liter of BEE-2,4-D

had no effect on grass shrimp (Hansen et al., 1973). The same authors reported that grass shrimp avoided 2,4-D but not 5 insecticides. Walsh (1971) found that 5 mg/liter of DMA-2,4-D had no effect on blue crabs after 24 or 48 hours' exposure and that 1 mg/liter had no toxic effect on Eastern oysters at 96 hours' exposure.

Walker (1953) reported that 2,4-D decreased the number of bottom organisms in ponds, presumably through removal of cover or feed. No adverse effects were noted on benthos or other aquatic invertebrates in large-scale field applications of BEE-2,4-D (Beaven et al., 1962; Sears and Meehan, 1971; Smith and Isom, 1967; Thomas and Duffy, 1968; Whitney, 1970). In another study, Rawls (1965) found that only the acetamide formulation was toxic to aquatic invertebrates at field application rates.

Adams (1950) and Adams and Drew (1965) reported that applications of 2,4-D could enhance aphid infestations of oat fields by the toxic effects on the coccinellid larvae, which biologically control the aphid.

Moffett and Morton (1971) and Moffett et al. (1972) found that 2,4-D was nontoxic to honeybees when applied in a water carrier, but oil carriers themselves were toxic. Beilman (1950)

sprayed shrubs along roads with 2,4-D and assumed bees could not use the area. Surprisingly, the shrubs were replaced by sweet clover which resulted in good honey production.

There was no effect on the survival of earthworms treated for 2 hours in 100 mg/liter of 2,4-D (Martin and Wiggans, 1959). Neither was the herbicide toxic to wireworms, spring tails, mites, or grasshoppers (Fox, 1964; Putnam, 1949). The reproduction of plant parasitic nematodes was inhibited by 5 mg/liter and impaired by 8 ug/liter of 2,4-D (Webster and Lowe, 1966). Reviews dealing with the effect of 2,4-D on invertebrates include those by USDA (1968), Bohmont (1967), Mullison (1970), and Pimentel (1971).

Fish. The earliest report dealing with the toxicity of 2,4-D to fish is that of King and Penfound (1946). They reported that 1 mg/liter was not toxic to bluegills or largemouth bass and 100 mg/liter was only slightly toxic. The herbicide DMA-2,4-D was not toxic to fry of bluegill, green sunfish, lake chubsucker, or smallmouth bass exposed to 25 mg/liter for 8 days (Hiltebran, 1967). Butler (1965) found no effect on longnose killifish treated for 48 hours with 15 mg/liter of DMA-2,4-D. There were no mortalities in bluegill exposed to 5 mg/liter of 2,4-D for 6 weeks (Cope et al., 1970), and 10 mg/liter was not toxic to squawfish (MacPhee and Ruelle, 1969). Sergeant et al. (1971) found that the acid and salt formulations of 2,4-D were non-toxic to

green sunfish at 110 mg/liter. However, BEE-2,4-D was toxic at the same concentration. There was no toxicity to fish noted in large-scale field applications of BEE-2,4-D or DMA-2,4-D (Beaven et al., 1962; Sears and Meehan, 1971; Whitney, 1970; Wojtalik et al., 1971). In another field study, Smith and Isom (1967) reported that fish temporarily moved out of an area treated with BEE-2,4-D, but no toxicity was noted.

Summaries or references to fish toxicity data are given in USDA, 1968; Bohmont, 1967; Lawrence, 1962a; Mullison, 1970; and Pimentel, 1971.

**Amphibians.** Frogs and turtles showed no toxic effects from 2,4-D treatment of ponds (Pierce, 1961). Sanders (1970) reported that tadpoles of the Western Chorus Frog withstood 50 mg/liter of DMA-2,4-D for 96 hours.

**Birds.** Fertile hen eggs tolerated injected doses of 50 mg/liter of 2,4-D with no effect on hatching (Dunachie and Fletcher, 1970). In the same study, the only teratogenic effect was feather blanching, which was not fatal. Somers et al. (1973) sprayed fertile chick eggs with the recommended field rate, and 10 and 20 times the recommended rate. They found no toxic effects on incubation performance, hatching, or growth of the chicks. Daily doses of

300 mg/kg of 2,4-D for several weeks were not toxic to chickens (Bjorklund and Erne, 1966). Whitehead and Pettigrew (1972) found that chicks tolerated a dietary level of 5,000 mg/kg for 1 week without toxic effects. Schultz and Whitney (1974) reported that field application of 2,4-D had no effect on the hatching of boat-tailed grackle eggs or development of fledglings.

Mammals. Hansen et al. (1971) fed rats 1,250 mg/kg and beagles 500 mg/kg of dietary 2,4-D for 2 years with no toxic effects. Seven doses of the alkanolamine salt of 2,4-D at 500 mg/kg was fatal to sheep and 44 doses of 200 mg/kg was fatal to cattle (Palmer and Radeleff, 1964). In the same study, nine doses of the propylene glycol butyl ether ester of 2,4-D at a level of 250 mg/kg was fatal to sheep. Repeated daily doses of 50 mg/kg of 2,4-D were fatal to pigs (Bjorklund and Erne, 1966). No toxicity to cattle was noted when they ingested forage from pastures sprayed with normal or 2 to 4 times the recommended rate of the herbicide (Mitchell et al., 1946; Grigsby and Farwell, 1950). Hassall (1965) stated that animals should be excluded from sprayed areas for 2 weeks to eliminate possible toxic effects of the herbicide and also because naturally poisonous weeds may



be rendered more palatable after spraying. Clegg (1971) reported no effects at 25 mg/kg of 2,4-D and further stated that no problems would be anticipated if dioxin contamination was not a problem.

**Human safety.** Some humans evidently have individual sensitivity to 2,4-D or the other contents of the formulation (Anonymous, 1956; Berkley and Magee, 1962). One individual accidentally ingested about 110 mg/kg body weight without fatal results (Berkwick, 1970). Hayes (1971) stated that the oral dose of 2,4-D required to produce illness in man is about 3 to 4 g. An intravenous dose of 2.0 g produced no illness but an intravenous dose of 3.6 g produced coma, hyporeflexia and urinary incontinence. Recovery was marked in 24 hours and complete in 48 hours. An oral dose of 6.5 g led to convulsions, followed by death. No genetic effects were noted in 220 men exposed to 30 to 40 mg of 2,4-D/day over extended periods (Johnson, 1971).

## RESIDUES

**Methodology.** The various formulations of 2,4-D are recovered from water by treating the water with acid or base, partitioning with an organic solvent, followed by a column cleanup and derivatization for glc analysis (Devine and Zweig, 1969; Faust and Soffet, 1966; Schultz, 1973; Schultz and Harman, 1974; Schultz and Whitney, 1974; Wilder, 1968). Hesselberg and Johnson (1972) and Rodgers and Stalling (1972) used column extractions for 2,4-D residues in fish. Coakley et al. (1964) and Duffy and Sheldon (1967) gave procedures for extracting BEE-2,4-D from shellfish and fish. Procedures for extracting 2,4-D from mud entail the use of methanol, acetone, or ethyl ether combined with other cleanup procedures (Hesselberg and Johnson, 1972; Schultz, 1973; and Woodham et al., 1971).

Procedures for extracting 2,4-D residues from fatty foods, animal products, and animal tissues utilize solvent extraction, partitioning with base, Soxhlet extraction, and cleanup procedures (U.S. Dept. HEW, 1968; Clark, et al., 1967; Crosby and Bowers, 1966; Yip, 1971).

Meagher (1966a, 1966b) gave procedures for extracting 2,4-D, the isopropyl ester of 2,4-D, and a 2,4-D conjugate from citrus peel, while Gutenmann and Lisk (1963) and Yip (1962) gave procedures for determining 2,4-D residues in forage crops.

Rivers et al. (1970) and Shafik et al. (1971) reported methods for determination of 2,4-D in human urine and blood.

Garbrecht (1970) and Scoggins and Fitzgerald (1969) reported esterification procedures for chlorophenoxy acids prior to glc analysis. Purkayastha (1969) detected ionizable herbicides by means of electrophoresis.

Residues. The persistence of 2,4-D in water has been reported by many workers. Averitt (1967) sprayed waters in Louisiana with DMA-2,4-D at the rate of 4.48 kg/ha and found maximum concentrations of 727 ug/liter and 1,020 ug/liter after 1 week in middle and bottom strata of water, respectively. After 3 weeks, concentrations had fallen to 12 ug/liter and 10 ug/liter, respectively. He also sprayed two lagoons at the same rate and found concentrations of 689 ug/liter and 967 ug/liter after 1 day and 11 ug/liter and 19 ug/liter after 31 days. Grzenda (1963) treated ponds with 0.6 ug/liter. After 62 days residues were less than 1 ug/liter. Kleinschmidt (1969) reported detectable levels of residues in water treated with BEE-2,4-D for up

to 22 days but stated that no residues could be detected after 29 days. Frank and Cones (1967) treated ponds with 1.33 mg/liter BEE-2,4-D and found only 0.024 mg/liter 1 day post-treatment and less than 0.001 mg/liter after 36 days. In ponds treated at 8.96 kg/ha, residues declined from 0.34 mg/liter and 0.69 mg/liter after 1 day in Florida and Georgia ponds, respectively, to less than 0.005 mg/liter by 28 days (Schultz and Harman, 1974). In Missouri ponds treated at the same rate it took 56 days for residues to decline to less than 0.005 ug/liter. The highest residue found in water sprayed at 4.48 kg/ha of an oil-soluble amine salt of 2,4-D was 37 ug/liter, 1 day post-treatment (Schultz and Whitney, 1974). When irrigation ditch-banks were treated at 1.56 to 3.36 kg/ha 2,4-D, the maximum concentration found in the canals was 213 ug/liter and 25 to 61 ug/liter in the irrigation water (Bartley and Hatrup, 1970; Frank *et al.*, 1970). Manigold and Schulze (1969) reported a residue of 0.35 ug/liter in 15 out of 20 stations, while Brown and Nishioka (1967) found no herbicide residues from streams analyzed for 2,4-D as part of the National Pesticide Monitoring Program.

Schwartz (1967), utilizing  $^{14}\text{C}$ -2,4-D, reported that 60% of the  $^{14}\text{C}$  remained in sewage effluent for 3 to 6 months. However, he determined only  $^{14}\text{C}$  and not actual 2,4-D. Oysters and clams

contained between 3.5 and 3.7 mg/kg of 2,4-D after exposure for 3 days to an application of 33.6 kg/ha (Coakley et al., 1964). Smith and Isom (1967) reported a maximum concentration of 1.12 mg/kg in mussels taken from water treated at 44.8 to 112 kg/ha of BEE-2,4-D. Oysters contained residues of 1.45 mg/kg after treatment of 44.8 kg/ha of BEE-2,4-D (Thomas and Duffy, 1968). Wojtalik et al. (1971) reported that plankton contained 3.6 ug/kg 4 weeks after treatment at 44.8 kg/ha DMA-2,4-D. Whitney (1970) reported that the highest residue in benthic organisms (grass shrimp, damselfly nymphs, and scud) was 0.23 mg/kg 24 hours post-treatment and that residues in blue crabs never exceeded 0.10 mg/kg.

Sears (1971) found the maximum concentration was 0.5 mg/kg in fish from streams adjacent to cut-over land sprayed with 2,4-D, while Cope et al. (1970) could detect no 2,4-D in fish 4 days after treatment with up to 10.0 mg/liter of 2,4-D in ponds. Walsh (1971) reported no residues of 2,4-D in fish fed 10 mg/kg in their food for 2 weeks and 8.4 mg/liter in fish fed 100 mg/liter for 2 weeks. No residues were detected in the latter fish after being fed a herbicide-free diet for 2 weeks. Rodgers and Stalling (1972) found that whole body or tissue residues in rainbow trout, bluegill, and channel catfish were proportional

to the exposure concentration. Only the liver contained detectable quantities of BEE-2,4-D, whereas the other organs contained only the acid with the highest concentration in bile. Schultz (1973) exposed fish to  $^{14}\text{C}$ -DMA-2,4-D and found radioactive residues ubiquitous for 84 days. However, the actual 2,4-D content was negligible. Among three groups of ponds treated at rates of 2.24, 4.48, or 8.96 kg/ha of 2,4-D, Schultz and Harman (1974) found no residues in fish 14 days post-treatment in Florida ponds, a residue of 0.075 mg/kg in bluegill from Georgia ponds, and no residues 28 days post-treatment in fish from Missouri ponds. Of fish taken from Florida canals sprayed at 4.48 kg/ha of 2,4-D, only 3 contained residues as high as 0.1 mg/kg, 16 had less than 0.01 mg/kg, and 41 had no detectable residues (Schultz and Whitney, 1974). Whitney (1970) reported that the highest residues in fish were 0.23 mg/kg in largemouth bass as a result of treating 200 acres of Eurasian watermilfoil with BEE-2,4-D, while Smith and Isom (1967) reported a maximum residue of 0.20 mg/kg in fish from an area treated for milfoil control.

Breast muscle and liver of Florida gallinules contained 2,4-D residues of 0.30 and 0.68 mg/kg, respectively, 1 day, and no detectable residues 4 days after canals at Loxahatchee National Wildlife Refuge were sprayed with 2,4-D for waterhyacinth control (Schultz and Whitney, 1974).

Residues were detected in mud for up to 6 weeks from ponds treated at 0.1 mg/liter to 10 mg/liter (Cope et al., 1970). Frank and Cumes (1967) reported residues of 4.96 mg/kg, 0.06 mg/kg, and less than 0.005 mg/kg in mud from ponds 1, 36, and 85 days post-treatment with a 1.33-mg/liter concentration of 2,4-D. Mud from Florida and Georgia ponds treated at 2.24, 4.48, or 8.96 kg/ha of 2,4-D contained no residues exceeding 0.05 mg/kg. Mud from Missouri ponds treated similarly contained a maximum residue of 0.17 mg/kg at 3 days and no detectable residue 28 days after treatment (Schultz and Harman, 1974). The maximum residue in mud from Loxahatchee Refuge was 0.005 mg/kg 15 days post-treatment with 4.48 kg/ha of 2,4-D (Schultz and Whitney, 1974). Wojtalik et al. (1971) reported residues of 0.30 mg/kg in mud 2 months after treatment at 22.4 or 44.8 kg/ha of DMA-2,4-D and residues less than 0.10 mg/kg 6 months post-treatment.

Cope et al. (1970) reported that vegetation from ponds treated with 0.1 to 10.0 mg/liter of 2,4-D contained residues for up to 6 weeks. Smith and Isom (1967) found that Eurasian watermilfoil contained 8.26 mg/kg 24 hours after treatment at either 44.8 or 112 kg/ha of BEE-2,4-D.

When rats were fed 100 mg  $^{14}\text{C}$ -2,4-D, all tissues and organs examined contained radioactive material 6 to 8 hours after the dose. This radioactive residue persisted for 17 hours. The largest amount of residue was 1,690 ug/0.1 g dry tissue found in the stomach of rats 24 hours after a dosage of 80 mg (Khanna and Fang, 1966). Zielinski and Fishbein (1967) reported that esters of 2,4-D disappeared more rapidly than the free acid in mice fed various 2,4-D formulations. Only about 10% of the 2,4-D was recoverable 24 hours after treatment. No 2,4-DCP was detected in animals injected with 2,4-D or its butyl or iso-octyl ester.

Less than 0.05 mg/kg 2,4-D or 2,4-DCP was found in the milk or cream of cows fed 30 to 1,000 mg/kg dietary 2,4-D for 2 to 3 weeks and then placed on untreated feed for 1 week (Leng, 1972). Negligible residues (less than 0.1 mg/kg) were found in the muscle and fat of slaughtered animals fed 2,4-D for 4 weeks at 300 mg/kg. Cows fed 300, 1,000, or 3,000 mg/kg of 2,4-D contained residues of 1.1, 4.9, and 5.3 mg/kg, respectively, in their kidneys. Residues in sheep were similar to those found in cows. There was no withdrawal period in the latter two studies (Leng, 1972). When a steer was fed 113.5 mg of 2,4-D, 100.65 ug was recovered in the urine (Lisk et al., 1963). Gutermann et al.



(1963) fed a Jersey cow 5 mg/kg of 2,4-D in grain for 5 days. No residues were detected in daily samples of milk or feces. They also fed a heifer 5 mg/kg and took samples from a fistula over a 23-hour period. Recovery of 2,4-D dropped from 3.5 initially to 0.5 mg/kg at the end of the experimental period.

**Metabolites and degradation products.** Extensive studies have been conducted on the degradation and metabolism of 2,4-D by bacteria. Bell (1957, 1960) reported that an Achromobacter species isolated from soil would degrade 2,4-D via oxidation. He reported that about 94% of the chlorine from 2,4-D was released as chloride. Anderson and Okrend (1968) found an Aerobacter species which degraded 2,4-D by 90% in 15 days. A species of Arthrobacter cleaved the ether linkage of 2,4-D acetate resulting, through oxidation, in 2,4-DCP (Tiedje and Alexander, 1969). The enzyme responsible for this cleavage converted <sup>14</sup>C-acetate-2,4-D to alanine and a volatile product. The authors proposed that glyoxylate was the initial product formed in cleavage of the side chain and that alanine was produced by condensation of two molecules of either glyoxylate or glycine. Tiedje et al. (1969) reported that 2,4-D was metabolized by an enzyme preparation to succinic acid. An enzyme from an extract of an Arthrobacter species dehalogenated 2,4-D and produced 2,4-DCP which was, in turn, further metabolized (Loos et al., 1967). Other workers found that extracts from an Arthrobacter

species would degrade 2,4-DCP, and other secondary degradation products of 2,4-D (Bollag et al., 1968a, 1968b, Duxbury et al., 1970).

Faulkner and Woodcock (1964) stated that Aspergillus niger would metabolize 2,4-D to 2,4-dichloro-5-hydroxyphenoxy-acetic acid while Steenson and Walker (1958) reported transformation of 2,4-D into 2,4-DCP by a strain of Flavobacterium peregrinum. Rogoff and Reid (1954) reported a Coxys bacterium which metabolized 2,4-D in buffered solution with a quantitative yield of chloride ion. Two Pseudomonas strains from soil utilized 2,4-D as a sole carbon source (Evans et al., 1971). One of the pseudomonads converted 2,4-D to 2,4-DCP, 2-chlorophenol, 3,5-dichlorophenol, and alpha-chloromuconate. The second Pseudomonad metabolized 2,4-D to 2,4-dichloro-5-hydroxyphenoxy-acetate, 2,4-DCP, 3,5-dichlorocatechol and alpha-gamma-dichloromuconate. The authors found that dechlorination had to occur at the 4 (para) position before ring fission could occur. Gaunt and Evans (1971) and Gamar and Gaunt (1971) found that a 2,4-D analog was metabolized in similar fashion to that above. Apparently, the formation of adaptive enzymes is necessary for the rapid degradation of 2,4-D (Robeck et al., 1963; Schwartz, 1967; Steenson and Walker, 1956; and Walker and Newman, 1956).

Daly (1971) found that BEE-2,4-D was degraded to 2,4-D in systems containing living material, pond water, polluted water, and water with milfoil, while Rodgers and Stalling (1972) reported that the hydrolysis of BEE-2,4-D to 2,4-D was accelerated by the presence of fish. The sodium salt of 4-(2,4-dichlorophenoxybutyric acid) [4-(2,4-DB)], an analog of 2,4-D, was converted to 2,4-D by fish (Gutermann and Lisk, 1965). Also Lisk et al. (1963), reported that steers fed 4-(2,4-DB) eliminated it in the urine as 2,4-D. When fish were exposed to DMA-2,4-D, one of the major metabolites was a glucuronide (Schultz, 1973).

The majority of the 2,4-D fed to rats was excreted unchanged in the urine (Shafik et al., 1971). Mitchell et al. (1946) reported finding 2,4-D (probably as a salt) in the blood serum of cows fed forage treated with 2,4-D. Eisner et al. (1971) reported finding 2,5-dichlorophenol in the defensive froth of grasshoppers and postulated that it was a degradation product of the 2,4-D applied to vegetation. Sheep fed  $^{14}\text{C}$ -2,4-D yielded approximately 96% of the intact molecule in the urine in 12 hours while less than 1.4% was excreted in the feces (Clark et al., 1964). These authors found little radioactivity in edible tissue and concluded that 2,4-D was excreted essentially unchanged by sheep.

In an early report of 2,4-D breakdown, Winston and Ritty (1961) stated that under field conditions 2,4-D was not degraded to 2,4-DCP but, rather, to CO<sub>2</sub>, HCl, and H<sub>2</sub>O. Thomas et al. (1964) reported that phenoxyacetic acids with a chlorine atom at position 4 were not readily hydroxylated. A heat-labile, insoluble, conjugated form of 2,4-D was found in citrus peel by Meagher (1966b). He speculated that the herbicidal molecule was probably conjugated with pectin. Linscott et al. (1968) and Hagin et al. (1970) reported that 4-(2,4-DB) and 2,4-D were converted to inactive compounds by resistant grasses. These inactive compounds had longer side chains than the parent compound thus preventing beta-oxidation and also preventing subsequent translocation to the site of action. Freed and Montgomery (1969) reported that the principal routes of metabolism of phenoxyacetic acids in plants were via conjugation and hydroxylation. Menzie (1969) and Loos (1969), compiled extensive reviews of the degradation of 2,4-D in soil, plants, bacteria, and animals.

## REGISTRATION STATUS

In the April 27, 1972 Federal Register (Vol. 37, No. 82) the Environmental Protection Agency and Food and Drug Administration (FDA) announced the issuance of proposed tolerances for dimethylamine salt, 2,4-D (DMA-2,4-D) used in control of weeds on irrigation ditch banks. A 0.1-mg/liter (negligible residue) tolerance was issued for DMA-2,4-D that may be present in potable water only as a result of application of DMA-2,4-D to ditch banks in western United States in programs of the following groups: Bureau of Reclamation; cooperating water user organizations; the Bureau of Sport Fisheries and Wildlife, USDI, and the Corps of Engineers, USDD (21 CFR Part 121). Livestock are to be excluded from treated ditch banks, and there is no reasonable expectancy of 2,4-D residues in milk or meat as a result of ditch bank treatment.

At present there is a request for a label for use of DMA-2,4-D for control of weeds on irrigation canal banks. This petition is sponsored by the Bureau of Reclamation and cooperating water users and the Bureau of Sport Fisheries and Wildlife, USDI, Agricultural Research Service, USDA; and the Corps of Engineers, USDD.

The Corps of Engineers, USDD is also presently seeking a label for the use of DMA-2,4-D for control of waterhyacinth in moving waters. Petitions have also been supported by, or submitted by, Amchem Products, Inc. and the Dow Chemical Company for other formulations of 2,4-D.

A copy of one proposed label has been appended to this manuscript.

## Literature Cited

- Adams, J. B. 1960. Effects of spraying 2,4-D amine on coccinellid larvae. *Can. J. Zool.* 38(2): 285-288.
- Adams, J. B., and H. E. Drew. 1965. Grain aphids in New Brunswick. III. Aphid populations in herbicide-treated oat fields. *Can. J. Zool.* 43(5): 789-794.
- Alabaster, J. S. 1959. Toxicity of weedkillers, algicides, and fungicides to trout. *Proc. 4th Brit. Weed Contr. Conf.* 2 pp.
- Alabaster, J. S. 1969. Survival of fish in 164 herbicides, insecticides, wetting agents and miscellaneous substances. *Int. Pest. Contr.*, March-April, 8 p.
- Aly, O. M., and S. D. Faust. 1964. Studies on the fate of 2,4-D and ester derivatives in natural surface waters. *J. Agr. Food Chem.* 12(6): 541-546.
- Anderson, M. O., and H. Okrend. 1968. Degradation of 2,4-D by *Aerobacter aerogenes*. *Bacteriol. Proc.*, A-25.
- Anonymous. 1956. Letter to the editor about possible 2,4-D acid poisoning in a farmer. *J. Amer. Med. Ass.* 162: 1296.

- Averitt, W. K. 1967. An evaluation of the persistence of 2,4-D amine in surface waters in the State of Louisiana. Proc. S. Weed Conf. 20: 342-347.
- Bartley, T. R., Jr., and A. R. Hattrup, Jr. 1970. 2,4-D contamination and persistence in irrigation water. Proc. West. Soc. Weed Sci. 23: 10-33.
- Beaven, G. F., C. K. Rawls, and G. E. Beckett. 1962. Field observations upon estuarine animals exposed to 2,4-D. Proc. Northeast. Weed Contr. Conf. 16: 449-458.
- Beilman, A. P. 1950. Weed killers and bee pasture. Amer. Bee J. 90(12): 542-543.
- Bell, G. R. 1957. Some morphological and biochemical characteristics of a soil bacterium which decomposes 2,4-dichlorophenoxyacetic acid. Can. J. Microbiol. 3: 821.
- Bell, G. R. 1960. Studies on a soil Achromobacter which degrades 2,4-dichlorophenoxyacetic acid. Can. J. Microbiol. 6: 325.
- Berkley, M. C., and K. R. Magee. 1962. Neuropathy following exposure to a dimethylamine salt of 2,4-D. Arch. Int. Med.: 351-352.



- Berwick, P. 1970. 2,4-dichlorophenoxyacetic acid poisoning in man. *J. Amer. Med. Ass.* 214(6):1114-1117.
- Bjorklund, N., and K. Erne. 1966. Toxicological studies of phenoxyacetic herbicides in animals. *Acta. Vet. Scand.* 7: 364-390.
- Bohmont, B. L. 1967. Toxicity of herbicides to livestock, fish, honey bees, and wildlife. *Proc. West. Weed Contr. Conf.* 21: 25-27.
- Bollag, J. M., G. G. Briggs, J. E. Dawson, and M. Alexander. 1968a. 2,4-D metabolism. Enzymatic degradation of chlorocatechols. *J. Agr. Food Chem.* 16: 829-833.
- Bollag, J. M., C. S. Helling, and M. Alexander. 1968b. 2,4-D metabolism. Enzymatic hydroxylation of chlorinated phenols. *J. Agr. Food Chem.* 16: 826-828.
- Bonderman, D. P., D. L. Dick, and K. R. Long. 1971. Occupational exposure to Aldrin, 2,4-D, and 2,4,5-T, and its relationship to esterases. *Ind. Med. Surg.* 40(6): 23-27.
- Bridges, W. R., and H. O. Sanders. 1963. Diffusion of herbicides through plastic film. *Prog. Fish-Cult.* 25(4): 213-214.
- Brody, I. A. 1973. Myotonia induced by monocarboxylic aromatic acids. *Arch. Neur.* 28: 243-246.

- Brown, E., and Y. A. Nishioka. 1967. Pesticides in selected western streams. A contribution to the national program. *Pestic. Monit. J.* 1: 38-46.
- Bruns, V. F. 1954. The response of certain crops to 2,4-dichlorophenoxyacetic acid in irrigation water. Part I. Red Mexican Beans. *Weeds* 3: 359-376.
- Bruns, V. F. 1957. The response of certain crops to 2,4-dichlorophenoxyacetic acid in irrigation water. Part II. Sugar beets. *Weeds* 5: 250-258.
- Bruns, V. F., and W. J. Clore. 1958. The response of certain crops to 2,4-dichlorophenoxyacetic acid in irrigation water. Part III. Concord grapes. *Weeds* 6: 187-193.
- Buck, W. B., W. Binns, L. James, and M. C. Williams. 1961. Results of feeding herbicide-treated plants. *J. Am. Vet. Med. Ass.* 138(6): 320-323.
- Butler, P. A. 1963. Commercial Fisheries Investigations. In Pesticide-wildlife studies. A review of fish and wildlife service investigations during 1961 and 1962. U.S. Fish and Wildl. Serv., Circular 167. 109 pp.
- Butler, P. A. 1965. Effects of herbicides on estuarine fauna. *Proc. S. Weed Conf.* 18: 576-580.

- Clark, D. E., J. E. Young, R. L. Younger, L. M. Hunt, and J. K. McLaran. 1964. The fate of 2,4-dichlorophenoxyacetic acid in sheep. *J. Agr. Food Chem.* 12(1): 43-45.
- Clark, D. E., F. C. Wright, and L. M. Hunt. 1967. Determination of 2,4-D residues in animal tissues. *J. Agr. Food Chem.* 15(1): 171-173.
- Clegg, D. J. 1971. Embryotoxicity of chemical contaminants of foods. *Food Cosmet. Toxicol.* 9: 195-205.
- Coakley, J. E., J. E. Campbell, and E. F. McFarren. 1964. Determination of butoxyethanol ester of 2,4-dichlorophenoxyacetic acid in shellfish and fish. *J. Agr. Food Chem.* 12(3): 262-265.
- Collins, T. F. X., C. H. Williams, and G. L. Gray. 1971. Teratogenic studies with 2,4,5-T and 2,4-D in the hamster. *Bull. Environ. Contam. Toxicol.* 6: 559-567.
- Cope, O. B., E. M. Wood., and G. H. Walen. 1970. Some chronic effects of 2,4-D on the bluegill (Lepomis macrochirus). *Trans. Am. Fish. Soc.* 99(1): 1-12.
- Corns, W. G., and R. K. Gupta. 1971. Chemical control of cattail, Typha latifolia. *Can. J. Plant Sci.* 51(6): 491-497.

- Crosby, D. G. 1969. The nonmetabolic decomposition of pesticides. *Ann. N. Y. Acad. Sci.* 160(1): 82-96.
- Crosby, D. G., and J. B. Bowers. 1966. Determination of 2,4-D residues in animal products. *Bull. Environ. Contam. Toxicol.* 1(3): 104-107.
- Crosby, D. G., and M. Li. 1969. Herbicide photodecomposition. pp. 321-363. In P. C. Kearney and D. D. Kaufman (Ed), *Degradation of Herbicides*. Marcel Dekker, Inc., N. Y. 394 pp.
- Crosby, D. G., and R. K. Tucker. 1966. Toxicity of aquatic herbicides to Daphnia magna. *Sci.* 154(3746): 209-291.
- Crosby, D. G., and H. O. Tutass. 1966. Photodecomposition of 2,4-dichlorophenoxyacetic acid. *J. Agr. Food Chem.* 14: 596-599.
- Crosby, D. G., and A. S. Wong. 1973. Photodecomposition of p-chlorophenoxyacetic acid. *J. Agr. Food Chem.* 21(6): 1049-1052.
- Daly, R. W., Jr. 1971. Degradation of 2,4-DBEE in an aquatic environment. Ph.D. Dissertation, Auburn Univ., Auburn, Ala. 158 pp.
- Davis, H. C., and H. Hudu. 1969. Effects of pesticides on embryonic development of clams and oysters and on survival and growth of the larvae. U.S. Fish and Wildl. Serv., *Fish. Bull.* 67(2): 393-404.

- Davis, J. T., and W. S. Hardcastle. 1959. Biological assay of herbicides for fish toxicity. *Weeds* 7: 397-404.
- De Rigo, H. T. 1964. Pre- and post-emergence chemical weed control in Chinese waterchestnut. *Proc. S. Weed Conf.* 17: 333-336.
- De Vaney, T. E. 1967. Chemical vegetation control manual for fish and wildlife management programs. U.S. Fish and Wildl. Serv., Resource Publ. 48, 42 p.
- Devine, J. M., and G. Zweig. 1969. Note on the determination of some chlorophenoxy herbicides and their esters in water. *J. Ass. Offic. Anal. Chem.* 52(1): 187-189.
- De Witt, J., D. G. Crabtree, R. B. Finley, and J. L. George. 1962. Effects on wildlife. In *Effects of pesticides on fish and wildlife: A review of investigations during 1960.* U.S. Fish and Wildl. Serv., Circular 143. 52 p.
- De Witt, J. B., W. H. Stickel, and P. F. Springer. 1963. Wildlife Studies, Patuxent Wildlife Research Center. In *Pesticide-wildlife studies. A review of fish and wildlife service investigations during 1961 and 1962.* U.S. Fish and Wildl. Serv., Circular 167. 109 p.

- Duffy, J. R., and P. Shelfoo. 1967. Determination of 2,4-D and its butoxyethanol ester in oysters by gas chromatography. *J. Ass. Offic. Anal. Chem.* 50(5): 1098-1102.
- Dunachie, J. F., and W. W. Fletcher. 1970. Toxicity of certain herbicides to hens' eggs assessed by the egg-injection technique. *Ann. Appl. Biol.* 66(3): 515-520.
- Duxbury, J. M., J. M. Tiedje, M. Alexander, and J. E. Dawson. 1970. 2,4-D metabolism: Enzymatic conversion of chloromethylacetic acid to succinic acid. *J. Agr. Food Chem.* 18: 199-201.
- Eisner, T., L. B. Hendry, D. B. Peakall, and J. Meinwald. 1971. 2,5-dichlorophenol (from ingested herbicide?) in defensive secretion of grasshopper. *Science* 172: 277-278.
- Elder, J. H., C. A. Lembi, and D. J. Morre. 1970. Toxicity of 2,4-D and picloram to fresh water algae. U.S. Clearing House, Fed. Sci. Tech. Inform. PB Rep. No. 199114, 13 pp.

- Evans, W. C., B. S. Smith, H. W. Fernley, and J. I. Davies. 1971. Bacterial metabolism of 2,4-dichlorophenoxyacetate. *Biochem. J.* 122: 543-551.
- Faulkner, J. K., and D. Woodcock. 1964. Metabolism of 2,4-dichlorophenoxyacetic acid (2,4-D) by Aspergillus niger von Tiegh. *Nature* 203: 865.
- Faust, S. D., and O. M. Aly. 1963. Some effects of 2,4-D and 2,4-DCP on drinking water quality. *Proc. Northeast. Weed Contr. Conf.* 17: 460-470.
- Faust, S. D., and I. H. Suffet. 1966. Recovery, separation, and identification of organic pesticides from natural and potable waters. *Residue Reviews* 15: 44-116.
- Faust, S. D., R. J. Tucker, and O. M. Aly. 1961. A preliminary report on the effect of some aquatic herbicides on water quality. *Proc. Northeast. Weed Contr. Conf.* 15: 546-548.
- Foret, J. A. 1967. Response of Elodea canadensis to treatment with phenoxy herbicides and organic acids. *Proc. S. Weed Conf.* 20: 288-289.

- Fox, C. J. S. 1964. The effects of five herbicides on the numbers of certain invertebrate animals in grassland soil. *Can. J. Plant Sci.* 44: 405-409.
- Frank, P. A., and R. D. Comes. 1967. Herbicidal residues in pond water and hydrosol. *Weeds* 15: 210-213.
- Frank, P. A., R. J. Demint, and R. D. Comes. 1970. Herbicides in irrigation water following canal-bank treatment for weed control. *Weed Sci.* 18(6): 687-692.
- Frank, P. A., R. H. Hodgson, and R. D. Comes. 1963. Evaluation of herbicides applied to soil for control of aquatic weeds in irrigation canals. *Weeds* 11(2): 124-128.
- Freed, M. L., and V. H. Montgomery. 1969. Metabolism of herbicides. *Ann. N. Y. Acad. Sci.* 160(1): 133-139.
- Gamar, Y., and J. K. Gaunt. 1971. Bacterial metabolism of 4-chloro-2-methylphenoxyacetate. *Biochem. J.* 122: 527-531.
- Gangstad, E. O. 1972. Herbicidal control of aquatic plants. *J. Sanit. Eng; Div. Amer. Soc. Civ. Eng.* 98(SA-2): 397-406.



- Garbrecht, T. P. 1970. Rapid esterification of dicamba and chlorophenoxy acids with N,O-bis(trimethylsilyl) acetamide for gas chromatographic analysis. *J. Ass. Offic. Anal. Chem.* 53(1): 70-73.
- Gaunt, J. K., and W. C. Evans. 1971. Metabolism of 4-chloro-2-methylphenoxyacetate by a soil pseudomonad. *Biochem. J.* 122: 519-526.
- Grigsby, B. H., and E. D. Farwell. 1950. Some effects of herbicides on pasture and on grazing livestock. *Michigan Agri. Exp. Sta., Quart. Bull.*, 32(3): 378-385.
- Grover, R., J. Maybank, and K. Yoshida. 1972. Droplet and vapor drift from butyl ester and dimethylamine salt of 2,4-D. *Weed Sci.* 20(4): 320-324.
- Grzenda, A. R. 1963. Public health affect of weed control in potable water supplies. *Proc. S. Weed Conf.* 16: 420-424.
- Gutenmann, W. H., D. D. Hardee, R. F. Holland, and D. J. Lisk. 1963. Residue studies with 2,4-dichlorophenoxyacetic acid herbicide in the dairy cow and in a natural and artificial rumen. *J. Dairy Sci.* 46: 1287-1288.

- Gutenmann, W. H., and D. J. Lisk. 1963. Rapid determination of 4(2,4-DB) and a metabolite, 2,4-D, in treated forage by electron affinity spectroscopy. *J. Agr. Food Chem.* 11(4): 304-306.
- Gutenmann, W. H., and D. J. Lisk. 1965. Conversion of 4-(2,4-DB) herbicide to 2,4-D by bluegills. *N. Y. Fish and Game J.* 12(6): 100-111.
- Hagin, R. D., D. L. Linscott, and J. E. Dawson. 1970. 2,4-D metabolism in resistant grasses. *J. Agr. Food Chem.* 18(5): 848-350.
- Hansen, D. J., S. C. Schimmel, and J. M. Keltner, Jr. 1973. Avoidance of pesticides by grass shrimp (*Palaemonetes pugio*). *Bull. Environ. Contam. Toxicol.* 9(3): 129-133.
- Hansen, W. H., M. L. Qurife, R. T. Haberman, and O. G. Fitzhugh. 1971. Chronic toxicity of 2,4-dichlorophenoxyacetic acid in rats and dogs. *Toxicol. Appl. Pharmacol.* 20: 122-129.
- Harrisson, J. W. E., and E. W. Rees. 1946. 2,4-D toxicity - 1. Toxicity towards certain species of fish. *Amer. J. Pharm.* 118(12): 422-425.
- Hassall, K. A. 1965. Pesticides: Their properties, uses and disadvantages. II. Fungicides and herbicides. Pesticides in relation to animals. *Brit. Vet. J.* 121: 199-211.

- Haven, D. 1963. Mass treatment with 2,4-D of milfoil in tidal creeks in Virginia. Proc. S. Weed Conf. 16: 345-350.
- Hayes, W. J. 1971. Insecticides, rodenticides, and other economic poisons. In Drills' Pharmacology in Medicine: 1256-1276.
- Hee, S. S. Q., and R. G. Sutherland. 1973. Penetration of amine salt formulations of 2,4-D into sunflower. Weed Sci. 21(2): 115-118.
- Heene, R. 1967. Inhibition of glycogen-forming enzymes by 2,4-dichlorophenoxyacetate (2,4-D) on cryostat sections of the skeletal muscle of warm-blooded animals. Histochemie 8: 45-53. (Translated from German).
- Hesselberg, R. J., and J. L. Johnson. 1972. Column extraction of pesticides from fish, fish food and mud. Bull. Environ. Contam. Toxicol. 7(2/3): 115-120.
- Hildebrand, E. H. 1946. Herbicidal action of 2,4-dichlorophenoxyacetic acid on the water hyacinth, Eichornia crassipes. Science 103: 477-479.
- Hiltibran, R. C. 1967. Effect of some herbicides on fertilized fish eggs and fry. Trans. Am. Fish Soc. 96(4): 414-416.

- Hughes, J. S. 1973. Acute toxicity of thirty chemicals to striped bass (Morone saxatilis). Presented at: West. Ass. of State Game and Fish Commissioners. Salt Lake City, Utah, July, 1973.
- Hughes, J. S., and J. T. Davis. 1962. Median tolerance limits reported in parts per million of bluegill sunfish to herbicides tested during 1961-1962. La Wild. and Fish. Comm., Monroe, La. 3 pp.
- Hughes, J. S., and J. T. Davis, 1963. Variations in toxicity to bluegill sunfish of phenoxy herbicides. Weeds 11(1): 50-53.
- Hughes, J. S., and J. T. Davis. 1964. Effects of selected herbicides on bluegill sunfish. Proc. 18th Ann. Conf. Southeast. Ass. Game and Fish Comm.: 480-482.
- Hughes, J. S., and J. T. Davis. 1966. Toxicity of pesticides to bluegill sunfish tested during 1961-1966. La Wildl. and Fish. Comm., Monroe, La. 7 pp.
- Huston, B. L. 1972. Identification of three neutral contaminants in production grade 2,4-D. J. Agr. Food Chem. 20(3): 724-727.
- Inglis, A., and E. L. Davis. 1968. The effect of water hardness on the toxicity to fish of several organic and inorganic herbicides. Abs., Weed Sci. Soc. of Am. Ann. Mtg., New Orleans, La., p. 47.

- Johnson, J. E. 1971. The public health implications of widespread use of the phenoxy herbicides and picloram. *Bio-Science* 21(17): 899-905.
- Jones, G. D., and J. U. Connell. 1954. Studies of the toxicity to worker honey-bees (*Apis mellifera* L.) of certain chemicals used in plant protection. *Ann. Appl. Biol.* 41(2): 271-279.
- Keith, J. O., R. M. Hansen, and L. Ward. 1959. Effect of 2,4-D on abundance and foods of pocket gophers. *J. Wildl. Mgmt.*, 23(2): 137-145.
- Khanna, S., and S. C. Fang. 1966. Metabolism of C<sup>14</sup>-labeled 2,4-dichlorophenoxyacetic acid in rats. *J. Agr. Food Chem.* 14(5): 500-503.
- Khera, K. S., and W. P. McKinley. 1972. Pre- and post-natal studies on 2,4,5-trichlorophenoxyacetic acid, 2,4-dichlorophenoxyacetic acid and their derivatives in rats. *Toxicol. Appl. Pharmacol.* 22: 14-28.
- King, J. E., and W. T. Penfound. 1946. Effects of two of the new formagenic herbicides on bream and largemouth bass. *Ecology* 27(4): 372-374.

- Kleinschmidt, H. E. 1969. Effect of granular 2,4-D on some waterweeds and its persistence. *J. Agr. Anim. Sci.* 26(4): 587-592.
- Krefting, L. W., and H. L. Hansen. 1963. Use of phytocides to improve deer habitat in Minnesota. *Proc. S. Weed Conf.* 16: 209-215.
- Lamartiniere, C. A., C. T. Hart, and A. D. Larson. 1969. Delayed lethal effect of 2,4-dichlorophenoxyacetic acid on bacteria. *Bull. Environ. Contam. Toxicol.* 4(2): 113-119.
- Lapham, V. T. 1964. Notes of the control of alligatorweed using dichlobenil. *Proc. S. Weed Conf.* 17: 325-327.
- Lawrence, J. M. 1962a. Aquatic herbicide data. Agriculture Handbook No. 231. Wash., D. C. ARS. USDA. 133 pp.
- Lawrence, J. M. 1962b. Research to discover means of controlling and destroying certain obnoxious aquatic plants by means of chemicals, and their toxicological effects on fish under controlled conditions. Final Report, Contract No. DA-01-076-CIVENG-60-215 between Dept. of Army, CE, and Agr. Exp. Stn., Auburn Univ., Auburn, Alabama. 122 pp.

- Leng, M. L. 1972. Residues in milk and meat and safety to livestock from the use of phenoxy herbicides in pasture and rangeland. *Down Earth* 20(1): 12-20.
- Linscott, D. L., R. D. Hagin, and J. E. Dawson. 1968. Conversion of 4-(2,4-dichlorophenoxy)butyric acid to homologs by alfalfa. *J. Agr. Food Chem.* 16(5): 844-848.
- Lisk, D. J., W. H. Gutermann, C. A. Bache, R. G. Warner, and D. G. Wagner. 1963. Elimination of 2,4-D in the urine of steers fed 4-(2,4-DB) or 2,4-D. *J. Dairy Sci.* 46: 1435.
- Loos, M. A. 1969. Phenoxyalkanoic acids. pp. 1-49. In P. C. Kearney and D. D. Kaufmann (Ed) *Degradation of Herbicides*. Marcel Dekker, Inc., N. Y. 394 pp.
- Loos, M. A., J. M. Bollag, and M. Alexander. 1967. Phenoxyacetate herbicide detoxication by bacterial enzymes. *J. Agr. Food Chem.* 15(5): 858-860.
- Lutz-Ostertng, Y., and H. Lutz. 1970. Detrimental effect of the herbicide 2,4-D on the embryonic development and fecundity of wildfowl (game birds). *Compt. Rend. Acad. Sci. Paris (Serie D)*, 2418-2421. (Translated from French).

- Lynn, G. E., and K. C. Barrows. 1952. The hydrocyanic acid (HCN) content of wild cherry leaves sprayed with a bush killer containing low volatile esters of 2,4-D and 2,4,5-T. Proc. Northeast. Weed Contr. Conf. 6: 331-333.
- MacPhee, C., and R. Ruelle. 1969. Lethal effects of 1888 chemicals upon four species of fish from western North America. Univ. of Idaho Bull. No. 3, 112 pp.
- Manigold, D. B., and J. A. Schulze. 1969. Pesticides in selected western streams - A progress report. Pest. Monit. J. 3(2): 124-135.
- Martin, A. C., R. C. Erickson, and J. H. Steenis. 1957. Improving duck marshes by weed control. U.S. Fish and Wildl. Serv., Circular 19. 49 pp.
- Martin, L. W., and S. C. Wiggins. 1959. The tolerance of earthworms to certain insecticides, herbicides, and fertilizers. Okla. State Univ. Exp. Sta., Proc. Ser. P-334.
- Maxwell, R. C., and R. F. Harwood. 1960. Increased reproduction of pea aphids on broad beans treated with 2,4-D. Ann. Entomol. Soc. Amer. 53: 199-205.



- Meagher, W. R. 1966a. Determination of 2,4-dichlorophenoxyacetic acid and 2-(2,4,5-trichlorophenoxy)propionic acid in citrus by electron capture gas chromatography. *J. Agr. Food Chem.* 14: 374-377.
- Meagher, W. R. 1966b. A heat-labile insoluble conjugated form of 2,4-dichlorophenoxyacetic acid and 2-(2,4,5-trichlorophenoxy)propionic acid in citrus peel. *J. Agr. Food Chem.* 14(6): 599-601.
- Menzie, C. M. 1969. Metabolism of pesticides. *Bur. Sport Fish. and Wildl., Spec. Sci. Report -- Wildl. No. 127*: 109-114.
- Mitchell, J. W., R. E. Hodgson, and C. F. Gaetjens. 1946. Tolerance of four animals to feed containing 2,4-dichlorophenoxyacetic acid. *J. Ani. Sci.* 5: 226-232.
- Moffett, J. O., and H. L. Morton. 1971. Toxicity of airplane applications of 2,4-D, 2,4,5-T, and a cotton desiccant to colonies of honeybees. *Amer. Bee J.* 111(10): 382-383.
- Moffett, J. O., H. L. Morton, and R. H. McDonald. 1972. Toxicity of some herbicidal sprays to honeybees. *J. Econ. Entomol.* 65(1): 32-36.

- Mount, D. I., and C. E. Stephan. 1967. A method for establishing acceptable toxicant limits for fish - Malathion and the butoxy-ethanol ester of 2,4-D. *Trans. Am. Fish. Soc.* 96: 185-193.
- Mullison, W. R. 1970. Effect of herbicides on water and its inhabitants. *Weed Science* 18(6): 738-750.
- Nelson, N. H., and S. D. Faust. 1969. Acid dissociation constants of selected aquatic herbicides. *Environ. Sci. Tech.* 3: 1186-1188.
- Palmer, J. S., and R. D. Radeleff. 1964. The toxicologic effect of certain fungicides and herbicides on sheep and cattle. *Ann. N. Y. Acad. Sci.* 111(2): 729-736.
- Petruk, G. F. 1965. Effect of herbicides on heterotrophic microorganisms of ponds. *Mikrobiologiya*, 1964, 33: 1018-1021. In: *Weed Abstr.* 14: 330, No. 1837.
- Philleo, W. W., and S. C. Fang. 1967. Effect of 2,4-dichlorophenoxyacetic acid on the in vivo metabolism of acetate in adult rats. *J. Agr. Food Chem.* 15: 256-260.
- Pierce, M. E. 1960. A study of the effect of the weed killer, 2,4-D granular on three experimental plots of Long Pond, Dutchess County, N. Y. *Proc. Northeast. Weed Contr. Conf.* 14: 483-487.

- Pierce, M. E. 1961. A study of the effect of the weed killer, 2,4-D aqua granular on six experimental plots of Long Pond, Dutchess County, N. Y. Proc. Northeast. Weed Contr. Conf. 15: 539-544.
- Pierce, M. E. 1968. The effect of several herbicides on eight test areas in Nobska Pond, Woods Hole, Massachusetts. Proc. Northeast. Weed Contr. Conf. 22: 195-203.
- Pierce, M. E. 1969. The effect of a combined formulation of 2,4-D and endothal upon a small pond. Proc. Northeast. Weed Contr. Conf. 23: 376-379.
- Pimentel, D. 1971. Ecological effects of pesticides on non-target species. Exec. Off. of the Pres., Off. of Sci. and Technology, U.S. Govt. Printing Office, No. 4106-0029. 220 pp.
- Plimmer, J. R., and U. I. Klingebiel. 1971. Riboflavin photosensitized oxidation of 2,4-dichlorophenol: Assessment of possible chlorinated dioxin formation. Science 174(4007): 407-408.
- Poland, A. P., D. Smith, G. Metter, and P. Possick. 1971. A health survey of workers in a 2,4-D and 2,4,5-T plant with special attention to chloracne, porphyria cutanea tarda, and psychologic parameters. Arch. Environ. Health 22: 316-327.

- Poorman, A. E. 1973. Effects of pesticides on Euglena gracilis.  
I. Growth studies. Bull. Environ. Contam. Toxicol. 10(1):  
25-28.
- Purkayastha, R. 1969. Direct detection of ionizable herbicides  
by electrophoresis. Bull. Environ. Contam. Toxicol. 4(4):  
246-255.
- Putnam, C. G. 1949. The survival of grasshopper nymphs on vege-  
tation treated with 2,4-D. Sci. Agric. 29(8): 396-399.
- Radeleff, R. D. 1964. Veterinary toxicology. Lea and Febiger,  
Philadelphia, Pa. pp. 241-261 (Chapter 8), 314 pp.
- Rawls, C. K. 1965. Field tests of herbicide toxicity to certain  
estuarine animals. Chesapeake Sci. 16(3): 150-161.
- Rexrode, C. O., R. P. True, and R. R. Jones. 1971. Influence of  
three herbicides on mat production and bark beetle attack in  
oak wilt trees. Plant Dis. Rep. 55: 1106-1107.
- Rivers, J. B., W. L. Yauger, Jr., and H. W. Klemmer. 1970. Simul-  
taneous gas chromatographic determination of 2,4-D and dicamba  
in human blood and urine. J. Chromatog. 50: 334-337.
- Robeck, G. G., J. M. Cohen, W. T. Sayers, and P. L. Woodward. 1963.  
Degradation of ABS and other organics in unsaturated soils.  
J. Water Poll. Cont. Fed. 35: 1225.

- Rodgers, C. A., and D. L. Stalling. 1972. Dynamics of an ester of 2,4-D in organs of three fish species. *Weed Sci.* 20(1): 101-105.
- Rogoff, M. H. 1961. Oxidation of aromatic compounds by bacteria. *Adv. Appl. Microbiol.* 3: 193.
- Rogoff, M. H., and J. J. Reid. 1954. Bacteriological decomposition of 2,4-D. *Bacteriol. Proc. (Soc. Am. Bacteriologists)* 54: 21
- Rowe, V. R., and T. A. Hymas. 1954. Summary of toxicological information on 2,4-D and 2,4,5-T type herbicides and on evaluation of the hazards to livestock associated with their use. *Amer. J. Vet. Res.* 15: 622-629.
- Sanders, H. O. 1969. Toxicity of pesticides to the crustacean Gammarus lacustris. *USDI, Fish and Wildl. Serv. Bull.* 25, 18 p.
- Sanders, H. O. 1970a. Pesticide toxicities to tadpoles of the Western Chorus Frog Pseudacris triseriata and Fowler's Toad, Bufo woodhousii fowleri. *Copeia* 2: 246-251.
- Sanders, H. O. 1970b. Toxicities of some herbicides to six species of freshwater crustaceans. *J. Water Poll. Cont. Fed.* 8, Part 1: 1544-1550.

- Sanders, H. O., and O. B. Cope. 1968. The relative toxicities of several pesticides to naiads of three species of stoneflies. *Limn. Oceanogr.* 13(1): 112-117.
- Sare, W. M. 1972. The weedicide 2,4-D as a cause of headaches and diplopia. *N. Z. Med. J.* 75: 173-174
- Schultz, D. P. 1973. Dynamics of a salt of (2,4-dichlorophenoxy)-acetic acid in fish, water, and hydrosol. *J. Agr. Food Chem.* 21(2): 186-192.
- Schultz, D. P., and P. D. Harman. 1974. Residues of 2,4-D in pond waters, mud, and fish. *Pestic. Monit. J.* (in press).
- Schultz, D. P., and E. W. Whitney. 1974. Monitoring 2,4-D residues at Loxahatchee National Wildlife Refuge. *Pestic. Monit. J.* (in press).
- Schwartz, H. G., Jr. 1967. Microbial degradation of pesticides in aqueous solution. *J. Water Poll. Contr. Fed.* 39: 1701-1714.
- Schwetz, B. A., G. L. Sparschu, and P. J. Gehring. 1971. The effect of 2,4-dichlorophenoxyacetic acid (2,4-D) and esters of 2,4-D on rat embryonal, foetal, and neonatal growth and development. *Food Cosmet. Toxicol.* 9: 801-817.

- Scoggins, J. E., and C. H. Fitzgerald. 1969. Rapid methylation of chlorophenoxyacetic acid herbicides with dimethyl sulfate for gas chromatographic analysis. *J. Agr. Food Chem.* 17: 156-157.
- Sears, H. S., and W. R. Meehan. 1971. Short-term effects of 2,4-D on aquatic organisms in the Nakwasina River watershed, South-eastern Alaska. *Pestic. Monit. J.* 5(2): 213-217.
- Sergeant, M., D. Blazek, J. H. Elder, C. H. Lembi, and D. J. Morre. 1971. Toxicity of 2,4-D and picloram herbicides to fish. *Proc. Ind. Acad. Sci.* 80: 114-123.
- Shafik, M. T., H. Sullivan, and H. F. Enos. 1971. A method for determination of low levels of exposures to 2,4-D and 2,4,5-T. *Int. J. Environ. Anal. Chem.* 1: 23-33.
- Sheldon, M. G., P. Johnson, J. E. Peterson, and W. H. Robison. 1964. *Pesticide-Wildlife studies, 1963. A review of fish and wildlife service investigations during the calendar year.* U.S. Fish and Wildl. Serv., Circular 199. 130 pp.
- Smith, G. E., and B. G. Isom. 1967. Investigations of effects of large-scale applications of 2,4-D on aquatic fauna and water quality. *Pestic. Monit. J.* 1(3): 16-21.

- Somers, J. D., E. T. Moran, Jr., and B. S. Reinhart. 1973. Effect of external applications of 2,4-D and picloram to the fertile egg on hatching success and early chick performance. *Down Earth* 29(3): 15-17.
- Stadnyk, L., R. S. Campbell, and B. T. Johnson. 1971. Pesticide effect on growth and  $^{14}C$  assimilations in a freshwater alga. *Bull. Environ. Cont. Toxicol.* 6(1): 1-8.
- Stenson, T. I., and N. Walker. 1956. Observations of the bacterial oxidation of chlorophenoxyacetic acid. *Plant and Soil* 8: 17.
- Stenson, T. I., and N. Walker. 1958. Adaptive pattern in the bacterial oxidation of 2,4-dichloro and 4-chloro, 2-methylphenoxyacetic acid. *J. Gen. Microbiol.* 18: 692.
- Stickel, L. 1964. *Wildlife Studies*, Patuxent Wildlife Research Center. In *Pesticide-Wildlife Studies*, 1963. A review of Fish and Wildlife Service investigations during the calendar year. U.S. Fish and Wildl. Serv., Circular 199, 130 pp.
- Sudak, F. N., and C. L. Claff. 1960. Survival of *Uca pugnax* in sand, water, and vegetation contaminated with 2,4-dichlorophenoxyacetic acid. *Proc. Northeast. Weed Contr. Conf.* 14: 508-510.
- Surber, E. W. 1949. Control of aquatic plants in ponds and lakes. 1949. U.S. Fish and Wildl. Serv., Leaflet 344. 20 pp.



- Thomas, E. W., B. C. Loughman, and R. G. Powell. 1964. Metabolic fate of some chlorinated phenoxyacetic acids in the stem tissue of Avena sativa. *Natura* 204: 286.
- Thomas, M. L. H., and J. R. Duffy. 1968. Butoxyethanol ester of 2,4-D in the control of eelgrass (Zostera marina L.) and its effect on oysters (Crossostrea virginica Gruehin) and other benthos. *Proc. Northeast. Weed. Contr. Conf.* 22: 186-193.
- Tiedje, J. M., and M. Alexander. 1969. Enzymatic cleavage of the ether bond of 2,4-dichlorophenoxyacetate. *J. Agr. Food Chem.* 17(5): 1080-1084.
- Tiedje, J. M., J. M. Duxburg, M. Alexander, and J. E. Dawson. 1969. 2,4-D metabolism: Pathway of degradation of chlorocatechols by Arthrobacter sp. *J. Agr. Food Chem.* 17: 1021-1026.
- Tucker, R. K., and D. G. Crabtree. 1970. Handbook of toxicity of pesticides to wildlife. U.S. Bur. of Sport Fish. and Wildl. Resource Publication No. 84. 131 pp.
- U.S. Department of Agriculture, National Agricultural Library. 1968. The toxicity of herbicides to mammals, aquatic life, soil microorganisms, beneficial insects, and cultivated plants, 1950-1965. A list of selected references. *Library List No.* 37: 1-161.

- U.S. Department of Health, Education, and Welfare, Food and Drug Administration. 1968. Pesticide Analytical Manual, Vol. I. Loose-leaf pub. n.p.
- Walker, C. R. 1963. Toxicological effects of herbicides. Proc. 8th Ann. Air and Water Poll. Conf. p. 17-33. Ed by Ralph H. Luebbers. Engineering Extension Series Bull. No. 2. The Univ. of Mo. Bull.
- Walker, E. M., Jr., R. H. Gadsden, L. M. Atkins, and G. R. Gale. 1972. Some effects of 2,4-D and 2,4,5-T on Ehrlich Ascites tumor cells in vivo and in vitro. *Ind. Med. Surg.* 41(1): 22-27.
- Walker, R. L., and A. S. Newman. 1956. Microbial decomposition of 2,4-dichlorophenoxyacetic acid. *Appl. Microbiol.* 4: 201.
- Walsh, G. E., J. M. Keltner, Jr., and E. Mathews. 1970. Effects of herbicides on marine algae. In Progress Reprint of the Bureau of Commercial Fisheries Center for Estuarine and Menhaden Research, Pesticide Field Station, Gulf Breeze, Fla. U.S. Fish and Wildl. Serv., Circular 335. 33 pp.
- Walsh, G. E. 1971. Effect of DMA-2,4-D on estuarine crustaceans. Personal communication.

- Webster, J. M., and D. Lowe. 1966. The effect of the synthetic plant-growth substance, 2,4-dichlorophenoxyacetic acid, on the host-parasite relationship of some plant-parasitic nematodes in monoxenic callus culture. *Parasitology* 56: 313-322.
- Wedemeyer, G. 1966. Microorganisms and pesticides at Seattle. In Progress in sport fishery research, 1965. *Eur. Sport Fish. and Wildl. Resource Publ.* 17: 54-55.
- Weed Science Society of America, *Herbicide Handbook.* 1970. W. F. Humphrey Press, Inc., Geneva, N. Y. 368 pp.
- Whitehead, C. C. 1973. Growth depression of broilers fed on low levels of 2,4-dichlorophenoxyacetic acid. *Brit. Poult. Sci.* 14: 425-427.
- Whitehead, C. C., and R. J. Pettigrew. 1972. The subacute toxicity of 2,4-dichlorophenoxyacetic acid and 2,4,5-trichlorophenoxyacetic acid to chickens. *Toxicol. Appl. Pharmacol.* 21: 348-354.
- Whitney, E. W. 1970. Expanded project for aquatic plant control, Currituck Sound, N. C. U.S. Fish and Wildl. Serv. Report. 20 pp.

- Wilder, E. T. 1968. Determination of the herbicide dimethylamine salt of 2,4-dichlorophenoxyacetic acid in surface water. *J. Amer. Water Works Ass.* 60(7): 827-831.
- Willard, C. J. 1950. Indirect effects of herbicides. *Proc. North Central Weed Contr. Conf.* 7: 110-112.
- Williams, M. C. 1968. Effects of herbicides on the capacity of spring parsley to photosensitize chickens. *Weed Sci.* 16(3): 350-352.
- Winston, A. W., Jr., and P. M. Ritty. 1961. What happens to phenoxy herbicides when applied to a watershed area. *Proc. Northeast. Weed Contr. Conf.* 15: 396-401.
- Wojtalik, T. A., T. F. Hall, and L. O. Hill. 1971. Monitoring ecological conditions associated with wide scale applications of DMA 2,4-D to aquatic environments. *Pestic. Monit. J.* 4(4): 184-203.
- Woodham, D. W., W. G. Mitchell, C. D. Loftis, and C. W. Collier. 1971. An improved gas chromatographic method for the analysis of 2,4-D free acid in soil. *J. Agr. Food Chem.* 19(1): 186-188.

- Yip, G. 1962. Determination of 2,4-D and other chlorinated phenoxy alkyl acids. *J. Ass. Offic. Anal. Chem.* 45(2): 367-376.
- Yip, G. 1971. Improved method for determination of chlorophenoxy acid residues in total diet samples. *J. Ass. Offic. Anal. Chem.* 54(4): 966-969.
- Zielinski, W. L., Jr., and L. Fishbein. 1967. Gas chromatographic measurement of disappearance rates of 2,4-D and 2,4,5-T acids and 2,4-D esters in mice. *J. Agr. Food Chem.* 15(5): 841-844.

Table 1.-- Efficacy data for 2,4-D on plants. Arranged alphabetically by authors.

Species	Concentration	Exposure	Formulation	Comments	Citations
Red Mexican beans	2.24 kg/ha 6.72 kg/ha		Acid	0.91 kg during seedling stage injured top growth and roots but didn't reduce yield. 2.73 kg reduced yield 40%. 0.91 kg at bloom stage did not reduce yield. 6.72 kg reduced yield 29%.	Bruns, 1954
Sugar beets	0.91 & 2.72 kg in water for irrigation		Acid	Both concentrations lethal during seedling stage and reduced stands in 1 in. diameter stage. 2.72 kg at 1 in. diameter stage reduced sucrose.	Bruns, 1957
Concord grape	2.24, 4.48, 8.96, 17.9 kg/ha		Acid	Lethal to 13, 33, 65, & 54% of plants respectively, lesser conc. No effect. Root systems of injured plants partially destroyed.	Bruns & Clore, 1958
Cattail	6.7 kg/ha		BEE	Single annual application in 112.3 liters diesel fuel/ha was effective for control. No effect on biological activity at nearby sewage lagoons.	Corns & Gupta, 1971
Eurasian watermilfoil	4.48 kg/ha		BEE		Daly, 1971
Lotus	2.24-4.48 kg/ha		DMA		DeVaney, 1967
Spatterdock	6.72-8.96 kg/ha				
Water chestnut	6.72-7.84 kg/ha				
Arrowarum					
Needlerush	33.6 kg/ha				
Hibiscus	7.84 kg/ha				
Groundselbush					
Hightide bush	1.81 kg/378.5				
Swamp loosestrife	liter water				
Willow					
Elodea Canadensis	4.48 and 6.72 kg/ha	2 months	DMA combined with molasses.	4.48 kg/ha gave 50% control in 2 months. 6.72 kg/ha gave 74% control in 2 months	Foret, 1967

Table 1.-- Contd

Species	Concentration	Exposure	Formulation	Comments	Citations
Waterlily, lotus, spatterdock, pennywort, willow, sedge, smartweed, cattail, arrowhead, pickerel weed, bullrush, spikerush, knot grass, needlerush, southern water grass, Sago pondweed, American pondweed	Not given		Acid	Not good control for the last two. Not good for use in irrigation canals due to lack of persistence in the soil. The first four were well controlled by 2,4-D. On all plants between the first 4 and the last 2 control was marginal.	Frank <i>et al.</i> , 1963
Eurasian watermilfoil	Not given		2-ethylhexyl and Granular BEE	Some reduction in amphipods and molluscs probably due to secondary effects such as smothering and habitat changes.	Haven, 1963
Waterhyacinth	1:1430 (acid:water W/V) 1:1140 (acid:water W/V)		Acid	Got 90-100% kill	Hildebrand, 1946
Dandelion Agoseris Western yarrow Rydberg Penstemon	Not given			Reduction was by 91%, 99%, 85%, and 88% respectively.	Keith <i>et al.</i> , 1959
Hydrilla	97 kg/ha		Granular containing 20% BEE of 2,4-D	Good control	Kleinschmidt, 1969
Mountain maple	0.91 kg/13.25 liter water aerial spray		BEE	Used to top kill maple and stimulate undergrowth used for deer browse.	Krefting and Hansen, 1963
Alligatorweed	8.96 kg/ha		DMA	Excellent control after 16 weeks when used with dichlobenil.	Lapham, 1964
Pithophora, Alligatorweed, waterhyacinth, duckweed, waterstar grass	5 mg/l in water, 100 mg/l in water, 5 mg/l sprayed on plants		DMA	% control for 5 mg/l in H <sub>2</sub> O was 40, 40, 90, 40, 90 respectively. % control for 100 mg/l in H <sub>2</sub> O was 95, 90, 100, 100, 100 respectively. % control for 5 mg/l sprayed on plants was 35, 80, 100, 45, 95, respectively.	Lawrence, 1962b

Table 1.-- Contd

Species	Concentration	Exposure	Formulation	Comments	Citations
Hyacinth Alligatorweed	2.24-4.48 kg/ha 8.96 kg/ha		Amine salt - DMA	Used for control in 1945 & 1948-50 Used in 1950-52 with spotty results.	Martin <i>et al.</i> , 1957
Nymphaea odorata Potamogeton Utricularia purpurea	1.5 mg/l. 3.1 mg/l	11 months	Granular	Nymphaea odorata eliminated by all 3 concentrations. Potamogeton accelerated by 1.5 mg/l. 3.1 and 6.2 mg/l reduced but did not eliminate U. purpurea.	Pierce, 1960
Nymphaea odorata Brasenia sp. Utricularia purpurea	10% a.e. at 0.9, 1.3 & 1.8 mg/l; 20% a.e. at 1.8, 2.6, & 3.6 mg/l.		Aqua granular	N. odorata and Brasenia sp. eliminated at all concentrations. U. purpurea reduced but not eliminated by 1.8, 2.6, & 3.6 mg/l.	Pierce, 1961
Surface plants	1 mg/l; 2 mg/l.	8 weeks		Applied in combination with fenac. Resulted in 50% reduction in surface weeds. Sprayed in combination with endothal at 2 mg/l. 50% clearing of surface noted in 8 weeks. No effect in Chara, Potamogeton, common sedge, fish or inverts.	Pierce, 1968
Potamogeton crispus	2 mg/l.	3 weeks		Applied with endothal. Hastened winter bud formation and/or death of the plant in 3 weeks.	Pierce, 1969
Waterhyacinth	4.48 kg/ha		Amine(oil)- Emuls- amine 3 DMA-2,4-D	General application was with the Emulsamine 3 and spot treatment was with DMA-2,4-D	Schultz and Whitney, 1974
Eurasian watermilfoil	44.8-112 kg/ha.		2,4-D BEE	No adverse effects noted on mussels, clams, aquatic fauna or water quality. Fish moved out of treated area.	Smith & Isom, 1967
Eurasian watermilfoil	10.16 metric tons 20% a.e./80.94 ha.		BEE	Improved Habitat. Destroyed the milfoil. Residues in various flora and fauna from the treated area are given.	Whitney, 1970
Spring parsley	Not given		DMA, PGBEE	2,4-D and 2,4,5-T controlled and detoxified spring parsley.	Williams, 1968
Eurasian watermilfoil	22.4-44.8 kg/ha		DMA-2,4-D	No distinguishable response noted in zoo or phytoplankton, benthic macroinvertebrates or fish.	Wojtalik <i>et al.</i> , 1971



Table 2.-- Toxicity of 2,4-D.

Organisms	Concentration		Exposure	Water Temp	Loading	Testing regime	Comments	Citations
	Treatment	Toxicity						
Concord grapes	2.24 kg/ha	13% killed					Lesser concentrations showed no effect. Roots partially destroyed.	Bruns & Clore, 1968
	4.48 "	33% "						
	8.96 "	65% "						
	17.9 "	54% "						
Oyster larva		% mortality					First 4 mortality figures from tests with a 2,4-D ester. The rest of mortalities from tests with an unnamed 2,4-D salt.	Davis & Hidu, 1969
	0.025 mg/l	-5						
	0.05 "	-3						
	0.10 "	-20						
	0.50 "	-2						
	0.025 "	-16						
	0.05 "	-2						
	0.10 "	-7						
	0.25 "	-8						
	0.50 "	-12						
	1.00 "	-11						
	10.00 "	-3						
50.00 "	-48							
100.0 "	-55							
Daphnia magna		IC50- >100 ug/g				Immobilization concentration	Crosby & Tucker, 1966 Sanders, 1970b	
Daphnia magna	TL50-0.10 mg/l		48 hr.	21° C		PGBE		
	" -4.0 "		"	"		DMA		
	" -5.6 "		"	"		BEE		
Seed shrimp	TL50-0.32 "		48 hr.	21° C		PGBE		
	" -8.0 "		"	"		DMA		
	" -1.8 "		"	"		BEE		
Scud	TL50-2.6 "		"	"		PGBE		
	" ->100.0 "		"	"		DMA		
	" -2.2 "		"	"		BEE		
Sowbug	TL50-2.2 "		48 hr.	15.5° C		PGBE		
	" ->100.0 "		"	"		DMA		
	" -3.9 "		"	"		BEE		

Table 2.-- Contd.

Organisms	Concentration		Exposure	Water Temp.	Loading	Testing regime	Comments	Citations
	Treatment	Toxicity						
Glass shrimp		TL50-2.7 mg/l	48 hr.	21° C			PGBE	
		" - >100.0 "	"	"			DMA	
		" - 1.4 "	"	"			BEE	
Crayfish		TL50->100.0 "	48 hr.	15.5° C			PGBE	
		>100.0	"	"			DMA	
		>100.0	"	"			BEE	
Gammarus lacustris (Scud)		LC50-1400 ug/l	24 hr.	21° C			2,4-D-BEE	Sanders, 1969
		" -2100 "	"	"			" -PGBE	
		" -6800 "	"	"			" -IOE	
		LC50- 760 "	48 hr.	"			2,4-D-BEE	
		" -1800 "	"	"			" -PGBE	
		" -4600 "	"	"			" -IOE	
		" - 440 "	96 hr.	"			2,4-D-BEE	
	" -1600 "	"	"			" -PGBE		
	" -2400 "	"	"			" -IOE		
						There was no effect from 100mg/l DMA after 96-hr.		
Fiddler crab (Uca pugnax)	10,000 mg/kg	100%(50%/72 hrs)	108 days			Removed and washed after 12-hrs.	2,4-D Acid. Injections of the Na-Salt showed toxicity to be greater than 0.4 mg/g.	Sudak & Claff, 1960
	5,000 "	100%(50%/96 hrs)						
	2,500 "	100%	10 days					
	1,000 "	100%(10-20%/2 wks)	17 days					
Stone fly naiad		LC50-8.50 mg/l	24 hr.				2,4-D BEE	Sanders & Cope, 1968
		" -1.84 "	48 hr.				"	
		" -1.60 "	96 hr.				"	
		" -56.0 "	24 hr.				2,4-D	
		" -44.0 "	48 hr.				"	
	" -15.0 "	96 hr.				"		
Coccinellid larvae	8 oz ae/acre	31 of 77 died.					Used mixed amine salts. Mortality more than 2 times as great in treated than in controls up to pupation. During pupation mortality was no greater. Deformity was greater in larvae sprayed at later stage of development.	Adams, 1960

Table 2.-- Contd

Organisms	Concentration		Exposure	Water Temp.	Loading	Testing regime	Comments	Citations
	Treatment	Toxicity						
Honey Bee ( <i>Apis mellifera</i> )	LD20-71.45 ug. LD50-104.50 " LD90-186.60 "	24 hr. " "					Na-Salt of 2,4-D toxicant was fed to the bees	Jones & Connell, 1954
Harlequin fish and Rainbow trout	TLm-7000 mg/l " - 4800 "	24 hr. 48 hr.					Shell 2,4-D QR pellets	Alabaster, 1969
Harlequin fish	" - 1160 "	24 hr.					2,4-D NaSalt	
Rainbow trout	" - 250 "	24 hr.					2,4-D triethanol amine	
Harlequin fish	" - 210 "	48 hr.					2,4-D BEE	
Harlequin fish	" - 1.0 "	24 hr.						
Harlequin fish	" - 1.0 "	48 hr.						
Trout	TLm-250 mg/l " - 210 mg/l	24 hr. 48 hr.					2,4-D-triethanol amine salt	Alabaster, 1959
Fathead minnow eggs	TLm-5.6 mg/l " - 1.5 mg/l	96 hr. 48 hr.				Static	Egg mortality was 100%.	Mount & Stephan, 1967
Killifish	LD50-2000 mg/kg			20-25° C	1 fish/gal		2,4-D acid	Harrison & Rees, 1946
Bream ( <i>Lepomis gibbosus</i> )	LD50-1000 mg/kg			"	"			
Bullhead ( <i>Ictalurus nebulosus</i> )	LD50-2000 mg/kg			"	"			
Striped bass ( <i>Morone saxatilis</i> )	LC <sub>0</sub> -0.1 larva LC50-0.15 " LC100-0.25 "	2.0 fry 3.0 " 4.0 "	96 hr. " "				2,4-D Butyl ester safe for use with fry. Nothing safe with larva.	Hughes, 1973
Green sunfish	5X10 <sup>-4</sup> M		60 min.				BEE	Sergeant, 1971
Bluegill	TL50->100.0 mg/l " - 1.1 "	48 hr. "		24° C "			DMA BEE	Sanders, 1970b

Organisms	Concentration		Exposure	Water temp.	Loading	Testing regime	Comments	Citation
	Treatment	Toxicity						
Bluegill	Tlm	650 mg/l	550	24 and			Weedar-64	Hughes & Davis, 1966
	"	600 "	500	48 hr.			"	
	"	475 "	450				Crop Rider 6D-2	
	"	230 "	230				Crop Rider Amine 4D-2	
	"	470 "	425				Ortho 2,4-D	
	"	327 "	327				2,4-D Amine 4	
	"	265 "	265				Amine	
	"	200 "	200			Weed Rhap A-4	Producers given. TLM based on active ingredients DMA-2,4-D	
Bluegill	TLM	542 mg/l	458	24. and	25° C.	10 fish/25 l	lake water	Hughes & Davis, 1963
	"	500 "	416	48 hr.				
	"	390 "	353					
	"	273 "	273					
	"	220 "	220					
	"	166 "	160					
Bluegill	TLM-542	mg/l	650	24 hr.			Weedar 64	Hughes & Davis, 1962
	"	500 "	600	"			"	
	"	394 "	475	"			Crop Rider 6D-2	
	"	390 "	470	"			Ortho 2,4-D	
	"	273 "	327	"			2,4-D Amine-4	
	"	220 "	265	"			2,4-D Amine	
	"	166 "	200	"			Weed Rhap A-4	
	"	458 "	550	48 hr.			Weedar 64	
	"	416 "	500	"			"	
	"	373 "	450	"			Crop Rider 6-D-2	
	"	353 "	425	"			Ortho 2,4-D	
	"	273 "	327	"			2,4-D Amine 4	
	"	220 "	265	"			2,4-D Amine	
	"	166 "	200	"			Weed Rhap A-4	
						The toxicity figures given are based on the acid equivalent and the active ingredients of DMA 2,4-D. Producers are also listed.		
Bluegill	TLM-188	mg/l		24 hr.			DMA-2,4-D	Hughes & Davis, 1964
	"	"		48 hr.				

Organisms	Concentration		Exposure	Water temp.	Loading	Testing regime	Comments	Citation
	Treatment	Toxicity						
Bluegill		TLm-350 mg/kg	24 hr.	25° C	10 fish/25 l	Water from Ouachita River (24) & Bayou DeSiard (24 & 48). Water aerated.	DMA TLm's based on 4 reps.	Davis & Hardcastle, 1959
		" 390 "	"	"				
Largemouth bass		" 375 "	48 hr.	"				
		" 350 "	24 hr.	"				
		" 375 "	24 hr.	"				
		" 350 "	48 hr.	"				
Largemouth bass		% mortality						
	1 mg/l	80	72 hrs.				DMA-2,4-D	Lawrence, 1962b.
	"	80	96 hrs.				Fathead minnows showed	
	5 mg/l	50	72 hrs.				no mortality after 96 hr.	
	"	70	96 hrs.				at 1, 5, and 10 mg/l	
	10 mg/l	40	72 hrs.					
	"	60	96 hrs.					
Frog		TL50-100 mg/l	96 hrs.			Static assay	Weedar 64 (DMA)	Sanders, 1970 a
Bobwhite quail, young	5,000 mg/kg	LD50-28,000 mg/kg					DMA-2,4-D. Treatment level	DeWitt <u>et al.</u> , 1962
adult		" > 38,000 "	<100 days				is based on amount in diet.	
							LD50 based on amount eaten	
Mallard, young	5,000 mg/kg	" 8,250 "					by the time 50% mortality oc-	
adult	>2,500 "	" >35,000 "					curred. Treatment concentration	
Adult pheasant		" >16,500 "					is the amount of DMA-2,4-D in	
							the diet in mg/kg.	
Bobwhite quail								
Young	5,000 mg/kg	LD50-8,250 mg/kg	<10 days				LD50's are based on average amount	DeWitt <u>et al.</u> , 1963
Ring neck pheasant							eaten by the time 50% mortality oc-	
Young	5,000 mg/kg	LD50-19,780 mg/kg	<100 days				curred. Treatment concentration is the	
Adult	>5,000 mg/kg	" >6,500 "	<100 days				amount of DMA-2,4-D in the diet in mg/kg.	
Mallard, young	2,500 "	" 22,100 "	<100 days					

Table 2.-- Contd.

Organisms	Concentration		Exposure	Water temp.	Loading	Testing regime	Comments	Citations
	Treatment	Toxicity						
Young Bobwhite quail	2,500 mg/kg	% mortality	10 days		24/pen		Treatment conc. based on amount of DMA-2,4-D in feed. % mortality based on number dead after 10, 30 or 100 days. Exposure time for LC50 is time to 50% mortality.	Stickel, 1964
		12	30 days					
		29	100 days					
Young Coturnix	2,500 "	12	10 days		25/pen			
		12	30 "					
Adult Coturnix	5,000 "	7	10 "		15/pen			
		27	30 "					
		33	100 "					
" "	2,500 mg/kg	100	98 days		16/pen			
		LD50-56,776 mg/kg	94 days					
		8	10 "					
Young pheasant	5,000 mg/kg	84	30 "		25/pen			
		LD50-15,998 mg/kg	19 days					
" "	2,500 mg/kg	28	30 "					
		36	100 "					
		LD50->1000 mg/kg						
Mallards		LD50->2025 "				Male & female	Technical acid	Tucker & Crabtree, 1970
"		LD50-ca2000 "				3-5 mo. old	" Na salt.	
"						Male-7 mo.	4 lbs a.e./gal. amine	
Pheasants		" -472(340-654)mg/kg				Male-3 mo-4 mo.	Technical acid	
Coturnix		" -668(530-842) "				Male-2 months	" "	
Pigeons		" -668(530-842) "				Male & female	" "	
Mule deer		" -400-800 "				Female 8-11 mo.	" "	
Rats, mice, guinea pigs, rabbits		" - 300-1000 mg/kg				Fed in H <sub>2</sub> O, olive oil, corn oil, capsule or plain	Dow formulations	Rowe & Hymas, 1954
Rat, Guinea pig		LD50-375-666 mg/kg						Johnson, 1971
		" -1000 "						
Man	6.5 g						Orally administered. Convulsions precede death. Death thought to be from ventricular fibrillation.	Hayes, 1971

## APPENDIX A - COMMON AND TECHNICAL NAMES OF ORGANISMS

## PLANTS

<u>Common name</u>	<u>Technical name</u>
WATER PLANTAINS Arrowhead	ALISMACEAE <u>Sagittaria latifolia</u>
AMARANTHS Alligatorweed	AMARANTHACEAE <u>Alternanthera philoxeroides</u>
ARUMS Arrowarum	ARACEAE <u>Peltandra virginica</u>
SEDGES Softstem bulrush Sedge Slender spikerush	CYPERACEAE <u>Scirpus validus</u> <u>Cyperus spp.</u> <u>Eleocharis acicularis</u>
GRASSES Southern watergrass	GRAMINEAE <u>Hydrochloa carolinensis</u>
WATER MILFOILS Eurasian watermilfoil	HALORAGIDACEAE <u>Myriophyllum spicatum</u>
FROGBITS Florida elodea	HYDROCHARITACEAE <u>Hydrilla verticillata</u>
DUCKWEEDS Duckweed	LEMNACEAE <u>Lemna minor</u>
BLADDERWORTS Bladderwort	LENTIBU LARIACEAE <u>Utricularia vulgaris</u>
LOOSESTRIFES Swamp loosestrife	LYTHRACEAE <u>Decodon verticillatus</u>
MALLOWS Hibiscus	MALVACEAE <u>Hibiscus moscheutos</u> <u>Hibiscus militaris</u> <u>Hibiscus lasiocarpus</u>

<u>Common name</u>	<u>Technical name</u>
PONDWEEDS	NAJADACEAE
Curlyleaf pondweed	<u>Potamogeton crispus</u>
Sago pondweed	<u>Potamogeton pectinatus</u>
American pondweed	<u>Potamogeton nodosus</u>
WATERLILIES	NYMphaeACEAE
Fragrant waterlily	<u>Nymphaea odorata</u>
Watershield	<u>Brasenia schreberi</u>
Lotus	<u>Nelumbo lutea</u>
Spatterdock	<u>Nuphar advena</u>
BUCKWHEATS	POLYGONACEAE
Smartweed	<u>Polygonum amphibium</u>
PICKEREL WEEDS	PONTADERIACEAE
Waterstar grass	<u>Heteranthera dubia</u>
Waterhyacinth	<u>Eichornia crassipes</u>
Pickereelweed	<u>Pontaderia cordata</u>
WATER CHESTNUTS	TRAPACEAE
Water chestnut	<u>Trapa natans</u>
CATTAILS	TYPHACEAE
Cattail	<u>Typha latifolia</u>
CARROTS	UMBELLIFERAE
Water pennywort	<u>Hydrocotyle umbellata</u>



## ANIMALS

<u>Common name</u>	<u>Technical name</u>
MOLLUSKS	MOLLUSCA
Eastern oyster	<u>Crassostrea virginica</u>
CRUSTACEANS	CRUSTACEA
DAPHNIA	DAPHNIDAE
Daphnia	<u>Daphnia magna</u>
OSTRACODS	
Seed shrimp	
ASELLIDAE	
Sowbug	<u>Asellus spp</u>
ASTRACIDAE	
Crayfish	<u>Orconectes spp</u>
PALAEEMONIDAE	
Grass shrimp	<u>Palaemonetes kadiakensis</u>
GAMMARIDAE	
Scud	<u>Gammarus lacustris</u>
DECAPODS	DECAPODA
Blue crab	<u>Callinectes sapidus</u>
Fiddler crab	<u>Uca pugnax</u>
Brown shrimp	<u>Penaeus aztecus</u>
INSECTS	INSECTA
Pteronardidae	
Stonefly naiads	<u>Pteronarcys californica</u>
ACRIDADAE	
Grasshoppers	<u>Romalea microptera</u>
APIDAE	
Honey bees	<u>Apis mellifera</u>

## FISHES

<u>Common name</u>	<u>Technical name</u>
TROUTS	SALMONIDAE
Rainbow trout	<u>Salmo gairdneri</u>
MINNOWS AND CARPS	CYPRINIDAE
Squaw fish	<u>Ptychocheilus spp</u>
Fathead minnows	<u>Pimephales promelas</u>
SUCKERS	CATOSTOMIDAE
Lake chubsuckers	<u>Erismyzon sucetta</u>
FRESHWATER CATFISHES	ICTALURIDAE
Channel catfish	<u>Ictalurus punctatus</u>
Brown bullhead	<u>Ictalurus nebulosus</u>
KILLIFISHES	CYPRINODONTIDAE
Longnose killifish	<u>Fundulus similis</u>
TEMPERATE BASSES	PERCICHTHYIDAE
Striped bass	<u>Morone saxatilis</u>
SUNFISHES	CENTRARCHIDAE
Green sunfish	<u>Lepomis cyanellus</u>
Pumpkinseed	<u>Lepomis gibbosus</u>
Bluegill	<u>Lepomis macrochirus</u>
Smallmouth bass	<u>Micropterus dolomieu</u>
Largemouth bass	<u>Micropterus salmoides</u>
AMPHIBIANS	
FROGS	HYLIDAE
Western Chorus Frog	<u>Pseudacris triseriata</u>
BIRDS	
PARTRIDGES	PERDIDIDAE
Red partridge	<u>Perdix rufa</u>
Gray partridge	<u>Perdix perdix</u>

<u>Common name</u>	<u>Technical name</u>
PHEASANTS Pheasant	PHASIANIDAE <u>Phasianus colchicus</u>
RAILS Florida gallinule	RALLIDAE <u>Gallinula chloropus</u>
GRACKLES Boat-tailed grackle	ICTERIDAE <u>Cassidix mexicanus</u>

APPENDIX B

PROPOSED LABEL COPY (front panel)

2,4-D AMINE WEED KILLER

FOR

CONTROL OF WEEDS ON IRRIGATION CANAL BANKS

CAUTION

KEEP OUT OF THE REACH OF CHILDREN

See other cautions on right panel

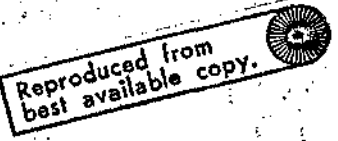
Active Ingredient:

\*Dimethylamine salt of 2,4-dichlorophenoxyacetic acid . . . . 49.5%

Inert Ingredients: . . . . . 50.5%

\*2,4-dichlorophenoxyacetic acid equivalent 41.2% by weight

or 4 pounds per gallon



Manufactured for use of the Bureau of Reclamation and cooperating water users organizations and the Bureau of Sport Fisheries and Wildlife, U.S. Department of the Interior; Agricultural Research Service, U.S. Department of Agriculture; and the Corps of Engineers, U.S. Department of Defense.

## PROPOSED LABEL COPY (left panel)

## GENERAL DIRECTIONS

Recommendations made are for noncrop use to control annual and perennial broadleaf herbaceous weeds and woody plants on banks of irrigation canals and irrigation drainage ditches.

Control of Broadleaf Annual and Perennial Weeds. Apply 1 to 2 quarts of 2,4-D Amine Weed Killer per acre in a sufficient volume of water to provide good spray coverage of vegetation; 20 to 100 gallons per acre is usually sufficient. Treat when weeds are young and actively growing before the bud or early bloom stage. For harder-to-kill weeds, a repeat spray may be needed after 3 to 4 weeks for maximum results. Apply no more than 2 treatments per season.

Control of Woody Brush and Patches of Perennial Broadleaf Weeds. Mix 1 gallon of 2,4-D Amine Weed Killer in 150 gallons of water. Wet foliage thoroughly using about 1 gallon of solution per square rod.

Spraying Instructions. Apply with low-pressure (optimum 10 psi and not over 40 psi) high-volume power spray equipment mounted on truck, tractor, or boat. Spray operation is to be made traveling upstream to avoid concentrating chemical in the water. Spray when the air is fairly calm (5 mph or less). Do not use on small canals (less than 10 cubic feet per second), when water will be used for drinking purposes. Boom spraying onto the water surface must be held to a minimum. When spraying shoreline weeds, avoid spraying over the water in the stream except to cover shoreline vegetation. Allow no more than 2 feet of spray onto water with an average of less than 1 foot of spray over the water surface to prevent introduction of greater than negligible amounts of chemical in water. Avoid cross-stream spraying to the opposite bank. Do not spray the banks on both sides of the stream at the same time. Spray each bank separately.

Convert pounds 2,4-D acid equivalent recommendations into terms of 2,4-D AMINE WEED KILLER by the following table:

2,4-D	1 lb	3/4 lb	1/2 lb	3/8 lb	1/4 lb	1/6 lb
2,4-D AMINE WEED KILLER	2 pt	1-1/2 pt	1 pt	3/4 pt	1/2 pt	3/8 pt

## PROPOSED LABEL COPY. (right panel)

Typical ditchbank weeds controlled by 2,4-D Amine Weed Killer are as follows:

## Annual and Biennial Weeds

beggarsticks  
 bull thistle  
 cocklebur  
 kochia  
 lambsquarter  
 lettuce (wild)  
 mallow  
 morningglory  
 marsh elder  
 mustards

parsnip  
 peppergrass  
 pigweed  
 prickly lettuce  
 primrose  
 ragweed  
 Russian thistle  
 sunflower  
 vetch

## Perennial Weeds

bindweed  
 catnip  
 Canada thistle  
 dandelion  
 dogbane  
 dock

goldenrod  
 hoary cress  
 nettles  
 plantains  
 water hemlock

## CAUTION

Do not allow dairy animals to graze on treated areas for at least 7 days after spraying. Water within treated banks should not be fished. Harmful if swallowed. Avoid contact with skin, eyes, or clothing. Avoid spray drift to susceptible plants, such as: cotton, beans, tomatoes, and ornamentals, as this produce may cause injury. Coarse sprays are less likely to drift. Thoroughly clean spray equipment with suitable chemical cleaner before using for other purposes (or do not use spray equipment for other purposes). Do not store near fertilizers, seeds, insecticides, or fungicides.

DO NOT USE THIS HERBICIDE AT RATES OR FOR METHODS OF APPLICATION OTHER THAN THOSE RECOMMENDED ON THIS LABEL TO PREVENT SERIOUS CONTAMINATION OF IRRIGATION WATER. DO NOT ADD INGREDIENTS OTHER THAN WATER TO THE HERBICIDE FORMULATION.

**DIRECTIONS FOR USE OF 2,4-D AMINE  
WEED KILLER FOR CONTROL OF WEEDS  
ON IRRIGATION CANAL BANKS**

The amine formulation of 2,4-D has been used for over 20 years to control broad-leaved weeds on irrigation canal banks of the western United States. The following directions summarize treatment procedures used by the Bureau of Reclamation and cooperation water users organizations.

Amount of Rates of Application

**Broad-leaved Annual and Perennial Weeds.** Apply 1 to 2 quarts per acre in sufficient volume of water to provide adequate coverage of vegetation; 20 to 100 gallons per acre is usually sufficient. Treat when weeds are young and actively growing before the bud or early bloom stage. Repeat treatments may be needed in 3 or 4 weeks for maximum control. For control of woody brush and patches of perennial broadleaf weeds, mix 1 gallon of 2,4-D with 150 gallons of water. Apply with hand gun or similar type of spot application equipment. Wet foliage thoroughly using about 1 gallon of solution per square foot.

Frequency and Timing of Application

Most irrigated areas of the western United States require only one application of 2,4-D amine in spring to early summer to control broad-leaved ditchbank weeds. Treatment timing will vary locally but should be made when the weeds are young and actively growing before the bud or early bloom stage. A second application may be required after 3 or 4 weeks for maximum results or when higher than normal rainfall encourages a second crop of ditchbank weeds. The second seasonal treatment can often be managed by spot treatment of the weedy vegetation, particularly perennial weeds or woody plants. Intervals between treatments will vary according to local conditions ranging from as little as 3 to 4 weeks or as much as 6 weeks to 2 months.

In areas where 2,4-D sensitive crops such as cotton are grown, make the first general ditchbank treatment so that it will precede emergence of the crops concerned.

Restrictions on Use and Application Procedures - Special Application Instructions

Apply with coarse low-pressure (optimum 10 psi and not over 40 psi) power spray equipment mounted on truck, tractor, or boat. Spraying operations are to be made traveling upstream to avoid concentrating chemical spray that gets into the stream. To prevent introduction of greater than negligible levels of chemical into the stream, boom spray and manually operated spray gun patterns should be made so as not to spray over the water surface more than 24 inches from the edge of the water.