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ENVIRONMENTAL TOXICITY OF TCDD

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ABSTRACT

Data on toxicity of TCDD to organisms, concentrations in the organisms, and concentrations in the media in and on which the organisms live can help determine the importance of measured levels of TCDD in the environment as part of the hazard assessment process. These toxicity and environmental fate data relationships vary depending on the media (soil, food, water, air) in which TCDD occurs. These variations are indicated by differences among bioconcentration factors, and the no-effect concentrations of TCDD for organisms in different media. Such data are summarized in this report.

INTRODUCTION

There is a need for more accurate techniques for hazard evaluation of 2,3,7,8-TCDD (2,3,7,8-tetrachlorodibenzo-p-dioxin) and other chemicals in the environment. Important data, often missing, are the toxicity to the organism, the concurrent related concentration in the organism, or specific tissue, and the concentration in the media in which the organism lives or through which it is exposed (food, soil, water, air). LC_{50} data are frequently the only data available. These concentrations in the media which result in 50 percent lethality to the organisms do not reveal the concentrations in the organisms which are lethal. Equally, the lethal concentration measured in the organism or tissue does not necessarily reveal the concentration at the specific site in the organism responsible for lethality. However, such concentrations as the latter are rarely known because of lack of our knowledge of the mode of toxic action of TCDD and most other chemicals and the sequences leading to effects such as death.

There appears to be more data available in the aquatic environment than in the terrestrial or aerial environment. Due to the toxicity of TCDD and extreme care needed to be used by humans handling it, toxicity tests for other organisms are limited. The test methods have often been different than standard tests, especially for aquatic organisms. Because of these difficulties the test results often have no uniform basis for comparative toxicity with other chemicals. Nevertheless, we believe that the information in this paper is summarized in a way to help use bioconcentration factors (BCF), environmental concentrations in different media, and concentrations toxic to aquatic and terrestrial organisms in their respective media in the hazard assessment process. Important data from field and laboratory toxicity and environmental fate tests (including bioconcentr- ation factors) are correlated for use in predicting results based on one or the other piece of information. While acute toxicity may be important, chronic toxicity is even more important with persistent chemicals because of delayed toxicity. Likewise, the no-observable-effect concentration (NOEC) is more important than the LC_{50} or LD_{50}. Consequently, in this paper more attention is given to the chronic no-observable-effect concentrations. Due to lack of information on environmental residues and toxicity of other isomers of the variously chlorinated dibenzo-p-dioxins and dibenzofurans only 2,3,7,8-TCDD is considered in this evaluation. Norris (1981) has provided a comprehensive review of the movement, persistence and fate of TCDD in the forest. Bovey and Young (1980) reviewed the presence of TCDD in other environments.

LABORATORY TOXICITY TESTS ON ANIMAL AND PLANT ORGANISMS

Toxicity to Mammals

Kociba et al, (1978) fed rats a diet containing 22 parts-per-trillion (ppt) TCDD for two years and produced no irreversible toxic effects. Rats fed 210 ppt showed several symptoms of toxicity, including liver and lung lesions. Rats fed 2200 ppt TCDD suffered mortality, carcinomas and acute effects. These diets were equivalent to 0.001, 0.01, and 0.1 µg TCDD/kg/day, respectively. Using a maximum food bioconcentration factor (BCF) of about 25 (based on measured fat or liver residues) the body burden of TCDD would be 0.55 to a maximum of 5.2 µg/kg at steady state.

Murray et al, (1979) studied the three-generation reproduction of rats receiving 0.001, 0.01, 0.1 µg TCDD/kg body weight/day. The 0.1 dosage produced no effect on the adults, but neonatal survival was poor. The 0.01 dosage significantly decreased fertility in the second and third generation, but not the first. The 0.001 dosage caused no-effect on any of the three generations. The total dosage per generation was 0.1 µg/kg/day x 130 days = 13 µg/kg, 0.01 µg/kg/day x 130 days = 1.3 µg/kg and 0.001 µg/kg/day x 130 days = 0.13 µg/kg TCDD, respectively.

Beach mice, <u>Peromyscus polionotus</u> having alumina dust containing 2.5 ppb TCDD applied to their pellage 10 times in 28 days in laboratory studies suffered no mortality, nor cellular changes, but there were statistically significant differences in liver to body weight ratios, (Cockerham et al, 1980).

The toxicity of TCDD to organisms is highly influenced by its solvent or adsorbent carriers, either dermally or by intestinal absorption. When TCDD was administered orally to rats in ethanol the amount adsorbed was twice that when the TCDD was administered in soil particles. Adsorption onto activated charcoal almost completely prevented uptake of the compound. Similar results were obtained dermally. Threshold levels for the induction of chloracne lesions on the skin of rats were 160 times greater when TCDD was adsorbed on charcoal than when administered as pure TCDD (Poiger and Schlatter, 1980). Such results indicated that TCDD is strongly adsorbed on soil and organic matter and is not easily removed by contact or through ingestion.

Toxicity to Birds

<u>Acute and subacute oral toxicity</u>. The acute oral toxicity of 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) to mallards was determined by Tucker and Hudson (1970). No mortality occurred at dosages of 2000 mg/kg 17 to 19 days after treatment. Assuming a TCDD impurity of <0.1 ppm in the 2,4,5-T, the TCDD dosage would be <0.2 µg/kg causing essentially no mortality in this test. Seventeen to nineteen days seems a sufficient

period of time to observe delayed symptoms or mortality typical of TCDD. Other tests with 2,4,5-T on chickens are shown by Kenaga (1975) with similar low levels of toxicity.

TCDD fed as one dose orally each day for 21 days to 3-day old Leghorn chicks produced no-observable-effect at 0.1 µg/kg/day (2.1 µg/kg total), 80 percent mortality and chick edema at 1 µg/kg/day for 21, days and 100 percent mortality at 10 µg/kg/day (in 15 days) (Schwetz et al, 1973).

These two pieces of information suggest that one would not expect mortality to occur with a total dose of less than 2.1 µg/kg, whether from a single or multiple dose. This is not meant to imply a "cumulative effect" from multiple doses, only to recognize that TCDD excretion is slower than the excretion of 2,4,5-T.

The results of tests on the chick edema effects of TCDD have been summarized by the National Research Council of Canada (1981) but are not reported here.

Bird/egg contact tests. A review of the effects of 2,4,5-T, (and TCDD impurities) by injection of pheasant and grouse eggs indicates that this method is a poor and unreliable index of toxicity relative to the practical use of the chemical. Spraying with heavy concentrations of 2,4,5-T on eggs of chickens and pheasants does not cause greater mortality or malformed embryos greater than occurs in control eggs. Concentrations of TCDD in the eggs, if present, have not been identified, (Kenaga, 1975).

Dietary feeding tests. The main oral intake of birds is via their diet, and thus dietary tests represent a more realistic route of exposure for birds than acute oral, or egg injection tests. The following tests (See Table 1) indicate a rather low order of toxicity from dietary feeding of 2,4,5-T to mallards and gallinaceous birds. 2,4,5-T assumed to contain 0.1 or less ppm TCDD caused no observable effect on chickens or turkeys at calculated concentrations of about 200-300 ppt TCDD in the diet in 11 to 21-day feeding studies (Roberts and Rogers, 1957; Whitehead and Pettigrew, 1972).

Reproduction tests using bobwhite in an 18-week dietary study using 50 ppm 2,4,5-T containing 0.06 ppm TCDD (equals 3 ppt TCDD) caused no observable effects at the highest concentration tested (Kenaga, 1975).

Table 1. Effects of TCDD Administered in 2,4,5-T on Various Species of Birds in Dietary Feeding Studies

Species	Concentration in the diet 2,4,5-T (ppm)	Concentration in the diet TCDD (ppt)	Duration of Test[a]	Effect[b]	References
Bobwhite *Colinus virginianus*	2776[c]	167	5 day/8 day	LC50	Kenaga, 1975
Bobwhite	50[c] 5[c]	3 0.3	18 wk/18 wk	No effect on reproduction	Kenaga, 1975
Mallard *Anas platyrhynchus*	4640[c]	278	5 day/8 day	LC10	Kenaga, 1975
Turkey *Malleagris gallopavo*	2500[d]	>259	11 day/11 day	EC0	Roberts & Rogers, 1957
Chicken *Gallus domesticus*	5000[d] 2000[d] 1000[d]	>500 >200 >100	21 day/21 day	LC90[e] LC0[e] LC0	Whitehead & Pettigrew, 1972

[a] Duration of exposure/Duration of observation.

[b] LC50 = Concentration causing 50% lethality, EC0 = no effect.

[c] 2,4,5-T containing 0.06 ppm TCDD.

[d] 2,4,5-T samples of this vintage usually contained >0.1 to 40 ppm TCDD.

[e] Also reduced feeding and growth.

Toxicity to Fish

The toxic response of fish to TCDD is characteristically slow with a long latent period for lethality after a short exposure. Death may be delayed from a few days to a few weeks or months. Other characteristic responses are edema, and reduced growth. These responses occur in water containing TCDD in the ppt to ppb range (Table 2).

Fish Dietary Treatment Tests

Rainbow trout consuming a diet containing 2.3 ppt or 2.3 ppb TCDD for 105 days showed no effect on food consumption, growth or survival, however, 50 to 88 percent mortality occurred in 61 and 77, days respectively, in fish exposed to 2.3 ppm TCDD (Hawkes and Norris, 1978). These authors expressed fish exposure data on a dryweight basis. We calculate these food concentration levels are equivalent to 0.0000064, 0.0072 and 4.2 µg TCDD/kg/day, respectively, on a fresh weight basis, assuming live weight is 5 times dry weight. The approximate no-effect oral intake level then would be 0.0072 µg/kg/day x 105 days of feeding or a total of 0.76 µg TCDD/kg live weight.

Aquatic Exposures

Hiltibran (1967) exposed small bluegills for 12 days to 50 ppm of the sodium salt of 2,4,5-T (45.9 ppm acid equivalent) causing no mortality. Assuming the 2,4,5-T contained 0.1 ppm TCDD, the concentration in the water would be 4.6 ppt TCDD. Miller et al, (1979) reported young coho salmon were more sensitive, with approximately 50 percent mortality 60 days after 96 hours exposure to 5.4 ppt TCDD in water, but 0.54 ppt for 96 hours had no effect in 114 days. A fish bioconcentration factor of 6400 for TCDD (Isensee, 1978) can be used to convert TCDD concentrations in water to concentrations in fish.

All of the water treatment tests in Table 2 on TCDD were conducted under static water conditions. Concentrations as low as 0.001 - 0.056 ppt caused some mortality (Miller et al, 1973; Helder, 1981). Yockim et al, (1978) measured tissues of dead mosquito fish and catfish which contained 7.2 (ppb) µg/kg and 4.4 (ppb) µg/kg, respectively. Thus, it appears that effect levels between 0 and 100 percent mortality for fish based on the body burden is approximately between 0.006 - 29.4 µg/kg.

Toxicity to Amphibians

Intraperitoneal injections of 1 mg TCDD/kg of body weight in bullfrog tadpoles and adults showed no effect indicating

Table 2. Effects of TCDD on Aquatic Organisms

Species	Test Duration[a/]	Effect	Concentration in Water(ppb) (µg/1)	Reference
Snail, Physa sp.	48 d/48 d	EC35, reduced reproduction	0.200	Miller et al, 1973
Oligochaete worm, Paranais sp.	49 d/55 d	EC35, reduced reproduction	0.200	" "
Mosquito (larvae) Aedes aegypti	17 d/39 d	No effect on pupation	0.200	" "
Guppy, Poecilia reticulata	5 d/37 d	Feeding decline, skin discoloration, fin necrosis, mortality	10	" "
Coho Salmon Oncorhynchus kisutch	24-96 hr/40 d	LC100	0.056	" "
Coho Salmon	24-96 hr/60 d	LC55	0.0056	" "
Guppy	5 d/5 d	LC8	0.100	Norris and Miller, 1974
Guppy	5 d/21.7 d	LC50	0.100	" "
Guppy	5 d/37 d	LC100	0.100	" "
Guppy	5 d/11.6 d	LC50	1.0	" "
Guppy	5 d/18.2 d	LC50	10.0	" "
Alga Oedogonium cardiacum	32 d/32 d	None	1.33	Isensee, 1978

(Continued)

Table 2. Effects of TCDD on Aquatic Organisms (Continued)

Species	Test Duration[a]	Effect	Concentration in Water(ppb) (μg/l)	Reference
Pond weeds, *Elodea nuttali* and *Certaophyllum emersum*	Months	none	0.0537	Tsushimoto et al., 1981
Snail	32 d/32 d	none	1.33	Isensee, 1978
Daphnia magna	32 d/32 d	none	1.33	" "
Mosquito fish	14 d/14 d	LC100	0.0025-0.0042	" "
Catfish	6 d/6 d	none	0.24	" "
Channel catfish *Ictalurus punctatus*	15 d/15 d	LC100	0.0042	Yockim et al., 1978
Mosquito fish *Gambusia affinis*	15 d/15 d	LC100	0.0028	" "
Rainbow Trout	61-71d/61-71 d	50-88% mortality feeding decline, weight loss, fin erosion, fungal growth liver degeneration	2,300[b] (in food)	Hawkes & Norris, 1977
Rainbow Trout	15 wk/15 wk	No effect	2.3[b] (in food)	" "
Rainbow Trout	15 wk/15 wk	No effect	0.0023[b] (in food)	" "

Species	Test Duration[a/]	Effect	Concentration in Water (ppb) (μg/1)	Reference
Coho salmon	96 hr/114 d	LC50	.0056 (5.4[c/])	Miller et al., 1979
Coho salmon	96 hr/114 d	No effect (Feeding growth, survival)	.00056 (0.54[c/])	" "
Northern pike (eggs) <u>Esox lucius</u>	96 hr/23 d	LC11	0.0001	Helder, 1980
Northern pike (eggs)	96 hr/23 d	LC42	0.001	" "
Northern pike (eggs)	96 hr/23 d	LC98	0.01	" "
Rainbow trout (juvenile)	96 hr/72 d	LC12	0.01	Helder, 1981
Rainbow trout (juvenile)	96 hr/21 d	LC50	0.100	" "
Rainbow trout (juvenile)	96 hr/27 d	LC100	0.100	" "
Rainbow trout (juvenile)	96 hr/73 d	EC54 (growth)	0.01	" "
Rainbow trout (eggs)	96 hr/24 wk	EC45 (growth)	0.01	" "
	96 hr/24 wk	EC8 (growth)	0.001	" "
	96 hr/24 wk	No effect (growth)	0.0001	" "
American bullfrog <u>Rana catesbeiana</u>	35 d	No mortality for larva & adults	1,000[b/] (IP)	Beatty et al., 1976

[a/]Test durations are expressed as follows: duration of exposure/duration of test; e.g., 36 day/48 day = 36 days exposure, 48 day test duration (12 day post-exposure observation). LC = lethal concentration, EC = some type of effect concentration other than lethal. IP = Interperitoneal injection.

[b/]Dosage expressed in μg/kg of organism or food (i.e., not concentration in water).

[c/] ng TCDD in water per gram of wet body weight of fish (i.e., not concentration in water).

that bullfrogs are relatively insensitive to TCDD compared to fish (Beatty et al, 1976) (Table 2.)

Toxicity to Aquatic Invertebrates

The species of snails, worms, cladocerans, and insects tested seem to be considerably less sensitive to TCDD than fish, although some effect was seen on reproduction on snails and worms at high levels of exposure (Table 2). No observable effect was seen on mosquito larvae at 0.2 ppb in water, the highest concentration tested (Table 2).

Toxicity to Aquatic Plants

Plants are relatively insensitive to TCDD compared to animals. No attempt has been made to determine the maximum no-effect levels. Plants tested at low concentrations of TCDD were not affected (Table 2).

Toxicity to Algae

The algae tested were not affected by the low concentrations (ppt) of TCDD (Table 2).

Toxicity to Soil Microorganisms

Bollen and Norris (1979) tested the respiratory effect of 2,4,5-T containing < 0.1 ppm TCDD on the microbial population of a forest floor and soil. No effect on CO_2 evolution was noted at concentrations of TCDD of 131 ppt in the forest floor or 52 ppt in the forest soil, the highest concentrations tested.

NO-EFFECT DOSAGE FROM DIETARY FEEDING IN MAMMALS, BIRDS, AND FISH

The total body intake of TCDD in daily dietary food giving a semi-chronic no-effect level for TCDD appears to be 0.55 - 5.2 μg/kg in mammals; 2.1 μg/kg in birds; and 0.76 μg/kg in fish. These values are remarkably similar.

AQUATIC AND TERRESTRIAL FIELD TOXICITY AND BIOCONCENTRATION TESTS ON ANIMAL AND PLANT ORGANISMS

Young et al, (1975) conducted field investigations on rodents, insects, aquatic organisms, and plant species associated with a unique 3 km^2 military test site (Test

Area C-52A, Eglin Air Force Base, Florida) that was sprayed with 73,000 kg 2,4,5-T and 77,000 kg 2,4-D between 1962 - 1970. Although neither 2,4-D nor 2,4,5-T residues could be detected in the soils in 1973 or 1974, significant levels (10-710 parts-per-trillion - ppt) of TCDD were found within the top 15 cm of test site soils although in some instances 10 years had elapsed since the last aerial application of 2,4,5-T.

A detailed study of the field effects of the herbicide and TCDD was conducted on populations of beach mice, *Peromyscus polionotus* and hispid cotton rats, *Sigmodon hispidus*. Liver tissue from rodents inhabiting the test site contained 210-1,300 ppt TCDD. However, no gross or histological evidence of teratogenesis or toxicity was found in 122 adults and 87 fetuses. An analysis of variance of liver and spleen weights for the beach mouse indicated significant differences between control and TCDD-exposed animals. Analysis of plant seeds revealed no detectable levels of TCDD (minimum detection limit of 1 ppt TCDD). TCDD accumulation in liver tissue was thought to be associated with pelt contamination from burrowing and subsequent ingestion of soil particles via grooming. Species diversities and food chain studies were conducted in two aquatic ecosystems draining the test area.

Deposition of soil form runoff occurred into a pond on the test area and into a stream immediately adjacent to the area. TCDD levels of 10 to 35 ppt were found in silt of the aquatic systems but only at the point where eroded soil entered in the water. Species diversity studies of the stream were conducted in 1969, 1970, 1973, and 1974. Insect larvae, snails, diving beetles, crayfish, tadpoles, and major fish species (by body parts) from both aquatic systems were analyzed for TCDD.

Species diversity studies indicated no significant change in the composition of ichthyofauna between these dates relative to a control stream. Concentrations of TCDD (12 ppt) were found in only two species of fish from the stream, *Notropis hypselopterus* (sailfin shiner) and *Gambusia affinis* (mosquito fish). Samples of skin, muscle, gonads, and gut were obtained from *Lepomis punctatus* (spotted sunfish) from the test grid pond. Levels of TCDD in those body parts were 4, 4, 18, and 85 ppt, respectively. Gross pathological observations of the sunfish revealed no significant lesions or abnormalities.

BIOCONCENTRATION

Bioconcentration factors are important because they provide the link between residues of the chemical in organisms compared to concentrations in other media such as water, soil, plants etc. These concentrations are linked to toxicity.

Norris (1981) reviewed environmental residues of TCDD in the field and concluded that bioaccumulation of TCDD in excess of 10 ppt in the majority of the animal population is not occurring in or near forest areas treated with 2,4,5-T or silvex.

Bioconcentration in Terrestrial Mammals from Food

Kociba et al, (1978) in two-year feeding studies with rats consuming dietary concentrations of TCDD at 2,200, 210, and 22 ppt found liver bioconcentration factors (BCF's) of 10.9 - 24.5, and fat BCF's of 3.7 - 24.5.

Kenaga (1980) reported a BCF of 3.5 based on a fat accumulation of 84 ppt of TCDD in cows after feeding a diet containing 24 ppt for 28 days. Bioconcentration factors in animals, based on dietary intake are of several different orders of magnitude less than those from water. Kenaga (1980) provides an equation for converting between terrestrial and aquatic bioconcentration factors.

Bioconcentration in Aquatic Animals from Water

Matsumura and Benezet (1973) calculated BCF's in organisms from simulated aquatic-terrestrial ecosystems exposed in different ways. The main source of uptake appears to be from water. Only these data are given in Table 3. The highest BCF figure for fish was 2850.

Isensee and Jones (1975), Isensee (1978), and Yockim et al, (1978) conducted static water tests in which soil was treated and organisms accumulated TCDD. Wet weight BCF's in several organisms (Table 3) ranged form 2200 - 6400, the latter for mosquito fish.

Isensee (1978) reported 6-14% degradation of ^{14}C-TCDD in his BCF tests. This infers that most but not all of the ^{14}C count found in animals used in calculating BCF data is based on TCDD.

Bioconcentration in Aquatic Plants from Water

It is unlikely that the TCDD accumulation for algae and duckweed shown in Isensee and Jones (1975) and Yockim et al, (1978) represent much more than adsorption of TCDD onto the plant. Since the surface area ratio of foliage and algae are

Table 3. TCDD Bioconcentration Factors for Water for Aquatic Organisms.

Species	Concentration in Water	Bioconcentration Factor[a/]	Exposure Duration (days)	Reference
Algae, *Oedogonium cardiacum*	2.42 ppt	2,075	7	Isensee, 1978[d/] (Table 2)
Algae	0.05 ppt - 239 ppt	3,268[b/] (aver.)	31	Isensee, 1978 (Table 1)
Pondweeds, *Elodea nuttali* & *Ceratophyllum emersum*	53.7 ppt	130 (max. 5 d)	5 - >60	Tsushimoto et al., 1981
Snail, *Physa sp.*	2.42 ppt	2,095	7	Isensee, 1978 (Table 2)
Snail	0.05 ppt - 239 ppt	6,106[b/] (aver.)	31	Isensee, 1978 (Table 1)
Daphnia magna	2.42 ppt 0.05 ppt - 239 ppt	7,070 4,438[b/] (aver.)	7 31	Isensee, 1978 (Table 2) Isensee, 1978 (Table 1)
Channel catfish, *Ictalurus punctatus*	4.2 ppt 2.6 ppt	1,048 2,269	32 15	Yockim et al., 1978 " "
Mosquito fish, *Gambusia affinis*	2.4 ppt 0.05 ppt - 239 ppt 2.42 ppt	4,875 6,970[b/] (aver.) 4,850	7 3 7	" " Isensee, 1978 (Table 1) Isensee, 1978 (Table 2)
Silversides *Labidesthes sicculus*	1300 ppt	545[e/]	4 - 7	Matsumura & Benezet, 1973
Mosquito larvae *Aedes aegypti*	1300 ppt	2,846[e/]	4 - 7	Matsumura & Benezet, 1973

a/ Based on ^{14}C count as ^{14}C TCDD, whole body, average values, wet weight.
b/ Dry wt/water of Isensee & Jones (1975), converted to wet weight.
c/ Calculated.
d/ Soil treated with TCDD and put in aquatic ecosystem.
e/ Values above 0.2 ppb exceed water solubility of TCDD and are probably low and not included in summary table.

great compared to their weight, the residue accumulated from adsorption from water is several thousand fold. The bioconcentration data concerning organisms in water are in Table 3.

Bioconcentration in Aquatic Animals from Sediment

Most tests of this type of exposure have not involved organisms that "live" in or on sediment. Fish or other free swimming types of organisms have been most commonly used. In laboratory tests TCDD has usually been adsorbed on sand or soil which was then placed in an aquarium and water and organisms added. Exposure probably involved desorption of TCDD from the sand or soil into the water and subsequent uptake from the water by the organism. In tests like this the BCF has usually been based on the level of TCDD in the water. Based on the level of TCDD in the soil or the sand, BCF values of less than 1 to 10 for snails, Daphnia, mosquito fish and catfish were found by Isensee and Jones (1975) and for brine shrimp, mosquito larvae, and fish by Matsumura and Benezet (1973). In these same tests the BCF values for transfer from water to organisms were much higher as discussed in a previous section. In a field test, TCDD residues of 10 to 35 ppt in sediment were associated with residues of 4 to 85 ppt in fish from the same site (BCF of less than 3).

Bioconcentration in/on Terrestrial Plants from Soil

TCDD apparently is not translocated in appreciable quantities in terrestrial plants. The amount found could be accounted for by volatility from the soil, or by rain splashed soil particles. This view is bolstered by the work of Kearney et al, (1973) who found that plants in soil fortified with high levels of TCDD do not readily take up TCDD and translocate it to aerial plant parts. Young et al, (1975) and Homberger et al, (1979) also found negligible BCF from plants on heavily treated areas.

Bioconcentration in Mammals from Soil and/or Food

Young et al, (1975) reported little bioconcentration by mammals from treated soil containing TCDD at the Eglin Air Force test site. Fanelli et al, (1980) reported 4.5 ppb TCDD in mice collected in an area where the concentration in soil was 3.5 ppb near Seveso (BCF about 1).

Conclusions from Bioconcentration Data

Bioconcentration of chemicals in organisms is highly variable and dependent upon the metabolism of the organism, molecular stability of the chemical, surface area to weight ratio of the organism exposed, food intake rate and the medium in which the organism lives, etc. The accuracy of BCF's are also dependent on the reliability of the analytical information. For organisms in water the BCF of TCDD is moderately high because of TCDD's hydrophobic nature (Tables 3 and 4). TCDD is also highly adsorbed on soil and particles.

BCF's of TCDD from soil to mammals, sediment to aquatic organisms, food to mammals and soil to plants are very low. Such low BCF's in organisms exposed to TCDD from soil is related to high soil adsorption and consequently to the low availability of TCDD and consequent low mammalian exposure to TCDD.

CORRELATION OF THE BCF AND THE NO-EFFECT CONCENTRATION OF TCDD IN ORGANISMS AND IN THE MEDIA CONTACTED BY THE ORGANISMS

The purpose of this correlation is to relate the no-observable-effect concentrations in water, soil, and food to the approximate no-effect concentrations occurring in organisms and to BCF's. Too often one measurement is available and the others are not. Where all three kinds of concentration data are available they have been tabulated (Table 4). Since these values vary among different species and test methods the values are useful as estimation, not as exact figures.

From the data in the tables and text, as summarized in Table 5, it is shown that no-observable-effect concentrations of TCDD in aquatic organisms are several hundred or thousand times greater than those in water. These differences correspond to the BCF values in whole body, liver or fat tissues as designated. It is also found that the TCDD no-observable-effect concentrations in organisms are no higher than 25 times that in their food from dietary uptake, whether aquatic or terrestrial organism, or form soil contacted by the soil exposed organism. BCF values for a given chemical vary greatly depending on the media to which the organism is exposed. Thus, by selection of the BCF for a specific organism (o) category (aquatic or terrestrial) and its specific media (m) category (water, food, or soil), the following equations are applicable:

Table 4. Media - Organism - Concentration - Toxicity Relationships with TCDD.

Media	TCDD Concentration in Media	Organism	TCDD Concentration in Organism[a]	BCF[b]	Duration[c]	Type of Test	Toxic Effect	Reference
1) Aquatic Habitat								
Water	2.42 ppt	*Daphnia*	17,100 ppt	7,070 (7d)	32d/32d	Static model ecosystem	LC0	Isensee, 1978
Water	2.42 ppt	Snail, *Physa*	5,100 ppt	2,095 (7d)	32d/32d	Static model ecosystem	LC0	" "
Water	2.42 ppt	Algae	5,000 ppt	2,075 (7d)	32d/32d	Static model ecosystem	LC0	" "
Water	53.7 ppt	Pondweeds	7,000 ppt	130	>60d	Outdoor pond	LC0	Tsushimoto et al., 1981
Food	2.3 ppb	Rainbow trout	1.57 ppb	≤ 1	105d	Dietary Feeding	None	Hawkes et al, 1977
Food	2.3 ppt	Rainbow trout	63 ppt	27(?)	105d	Dietary feeding	None	" "
Sediment	10-35 ppt	Mosquito fish	12-35	1-3	life	Field	None?	Young et al., 1975
2) Terrestrial Habitat								
Food	22 ppt	Rat	540[e,f]	24.5[e,f]	2 yr	Chronic & oncogenic	None	Kociba et al., 1978
Food	210 ppt	Rat	5,100[e] 1,700[f]	24.3[e] 8.1[f]	2 yr	Chronic & oncogenic	Decreased neonatal size, etc.	" "
Food	24 ppt	Cattle	84 ppt[f]	3.5	4 wk	Residue	None	Kenaga, 1980
Food & Soil	0.22-2.3ppt[d,g] plants (initially)	Mountain beaver	<3-17 ppt[e]	<1-14	45-59d	Field	None	Newton et al., 1979
Food & Soil	10-710 ppt (soil) <1 ppt (food)	Beachmouse & Cotton rat	210-1300 ppt[e]	2-21	multigeneration, teratogenesis	Field reproduction	None	Young et al., 1975
Soil	2-200 ppt	Various plants	<2.5 ppt	<0.01	several months	Field	None	Homberger et al., 1979
Soil	10-710 ppt	Plants (many species)	<1 ppt	<0.1	multigeneration	Field reproduction	None	Young et al., 1975
Explosion from factory into air	Variable (2-200 ppb)	Plants	up to 15 ppm	?	several weeks	Monitoring	Practically none	Homberger et al., 1979

[a] wet weight assumed. [b] Average figures. [c] Duration of exposure/duration of test (see Table 2 footnote[a].)
[d] in 2,4,5-T containing 0.1 ppm or less TCDD. [e] liver. [f] fat. [g] calculated.

Table 5. Summary of No-Effect Concentration in Media and Organisms, and Bioconcentration Factors of TCDD

Organism Tested	Approximate No Observable Effect Concentrations (From Tables and Text)		BCF[c/]
Aquatic Species	In Organism	In Medium	
		Water	
Fish	.1 ppb - 9.3 ppm[a,b/]	0.1 ppt - 1.33 ppb	1048 - 6970
Daphnia	5.9 - 9.4 ppm[b/]	1.33 ppb	4438 - 7070
Mosquito larvae	>569 ppb[b/]	>200 ppt	>2850
Snail	>0.88 ppb <1.22 ppm[b/]	>4.2, <200 ppt	2095 - 6106
Algae	>4.2 ppm	>1.3 ppb	2075 - 3268
Pondweed	>7 ppb[b/]	>53.7 ppt	130
		Sediment	
Fish	12-85 ppt	10-35 ppt	1 - 3
		Food	
Fish	63 ppt - 1.6 ppb	0.002-2.3 ppb	<1 - 27
Terrestrial Species			
	Organism	Food	
Mammals	>7.7-<5250 ppb[b,d/]	>22 - <210 ppt	3.5 - 25[d/]
	Organism	Soil	
Mammals	>1300 ppt[d/]	>710 ppt	2 - 21[d/]
Plants	>1 ppt	>710 ppt	<0.1

[a/] ppb = μg/kg body weight.

[b/] Calculated from BCF and amount in medium.

[c/] BCF = $\frac{\text{concentration in organism}}{\text{concentration in media}}$ (See data in Tables 3 and 4)

[d/] BCF based on fat and/or liver concentrations.

$$BCF = \frac{NOEC_o \text{ (no-observable-effect concentration in organism)}}{NOEC_m \text{ (no observable-effect-concentration in the media).}}$$

or $NOEC_o = BCF \times NOEC_m$

or $NOEC_m = \dfrac{NOEC_o}{BCF}$

The predictive value of these and other recently discovered relationships are considerable. Once the BCF has been determined or estimated only one no-observable-effect concentration for similar habitat and organism value is needed to estimate other unknown values. Thus, the no-effect concentration for the medium and for the organism are both easily estimated to within a reasonable and practical limit.

If the BCF has not been established it can now be reasonably estimated for persistent organic chemicals by regression equations from water solubility, octanol-water partition coefficient (K_{ow}), soil-water adsorption coefficient (K_{oc}), or high pressure liquid chromatography (Kenaga and Goring, 1980; and Veith and Morris, 1979). Values for K_{ow} can be predicted from organic structure alone or from linear free energy (Leo et al, 1971). There may be limits to these correlations on polychloroaromatic compounds, such as biphenyls, dibenzofurans, and dibenzodioxins above the molecular weight of 450-500 and with limited water solubility resulting in decreased penetration and distribution of such chemicals in animal tissues, thus lower BCF's.

Values for water solubility not ascertained experimentally can be accurately predicted for organic chemicals from melting point and molecular surface area (Yalkowsky and Valvani, 1979).

If a BCF is known for aquatic species, BCF's can be calculated by regression equations for terrestrial species from their food (Kenaga, 1980), which are reasonably similar to those from soil.

COMPARISON OF EXCESSIVE CONCENTRATIONS OF TCDD IN VARIOUS MEDIA AND TOXIC EFFECTS WITH TCDD CONCENTRATIONS FROM RECOMMENDED USES OF 2,4,5-T

A comparison of different TCDD dosages per acre; residue concentrations in soil, vegetation, and animals; and toxicological effects are useful for practical consideration in hazard evaluation. Such measurements were made for the massive contamination by TCDD from the "Seveso Accident" (Homberger et al, 1979) and the Eglin Air Force Base test site from Agent Orange (Young et al, 1975) in contrast to maximum concentrations expected from normal EPA registered uses for control of vegetation by aerial application of 2,4,5-T to range (Table 6). It should be emphasized that no animal or plant mortality ascribed to TCDD alone has occurred at these sites. No long-term toxic effects such as teratogenesis or reproductive effects are known from animals living their entire life cycle on areas treated with among the highest concentrations of TCDD known to occur. The lack of effects at these sites illustrate the potential for substantially overestimating risk by the undue extrapolation of acute oral toxicity of TCDD to toxicity which might result from daily contact with TCDD in feed and soil in the natural environment of the animal.

The dosage difference between high concentrations of TCDD in the Seveso and Eglin Air Force Base sites, and the maximum concentrations expected from the EPA registered dosages is over 1000 fold. It appears that concentrations from 2,4,5-T applications registered by EPA result in TCDD concentrations which are far below those from which adverse effects on animals or plants could be expected from exposures to water, soil or food in the natural environment.

CONCLUSIONS

The data on TCDD summarized here establishes approximate no-observable-effect concentrations for the food and media of organisms and the bioconcentration factors to be expected between various media and organisms. With the use of the predictive relationships described, given one or two of these factors or no-effect levels the other no-effect level or BCF can be determined for practical purposes. None of the registered uses of 2,4,5-T will result in TCDD concentrations in the environment similar to those resulting from the Seveso, Italy explosion or the testing at the Eglin Air Force Base Agent Orange test site. Despite more than a 1000-fold greater concentration of TCDD at these sites, compared to those expected from the registered uses of 2,4,5-T, there appears to be little evidence of reduction of animal or plant organism populations due to TCDD. These findings are consistent with those predicted by the relationship between bioconcentration factor and no-observable-effect concentration of TCDD in the organism and in the medium through which it is exposed.

Table 6. Concentrations in Various Media and Toxic Effects of TCDD.

Location	lb. TCDD/A (average)	ppt TCDD Residues: Soil	ppt TCDD Residues: Vegetation[a]	ppt TCDD Residues: Animals	Toxicological Effects - Terrestrial Animals
Seveso, Italy	0.000026[b] 0.00056[b,c]	23[b] 10,000[b,c]	1 - 137[b] 15.8 (ppm)[b,c]	58,000 - 215,000[b]	Initial mortality of small animals, none after 2 mo.[a]
Eglin Air Force Base, Florida, U.S.A.	0.00025[d]	10 - 710[d]	<1[d]	210 - 1300[d]	No mortality or terato-genesis; livers enlarged[d]
EPA approved aerial application of 2,4,5-T to rangeland.	0.0000002[e]	0.18[c,e,f]	23[c,e,g]	4[c]	None known from many years of use

[a] No toxic effects seen at any concentrations due to TCDD. Acute effects ascribed to sodium trichlorophenate and alkalinity from explosion.

[b] Measured shortly after explosion in Zone A, area of highest contamination.

[c] Maximum values.

[d] Measured 1973, 4 up to 10 years after last application of 2,4,5-T totaling 160,948 lb/A; 2,4,5-T assumed to contain 1 ppm TCDD.

[e] Assume 2 lb. 2,4,5-T applied/A, containing 0.1 ppm TCDD.

[f] In top 20 cm of soil.

[g] See Hoerger & Kenaga (1972), 1 lb/A = 110 ppm[c] on grass, immediately after spraying.

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