

Comparative Toxicity in Developmental Stages of Fish from 2,3,7,8-Tetrachlorodibenzo-*p*-dioxin (TCDD)

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1. Introduction

A number of studies have shown that the toxic effect of TCDD depends on the fishes life stage and species. In order to properly assess the impact of TCDD in the environment it is important to know the TCDD body burden. Little is known about the relationship between the TCDD body burden and the toxic effects of TCDD in fish^{1,2,3)}.

This paper compares the sensitivity and toxicity of TCDD in early life stages of the fathead minnow (*Pimephales promelas*) with other fish species. In addition, TCDD toxicity studies were carried out using the fathead minnow embryo and larvae as an animal models which emphasize on the relationship between the tissue dose and the biological effects.

2. Methodology

Embryos and larvae of the fathead minnow (*Pimephales promelas*, Rafinesque) were waterborne exposed to [¹H]2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) (40 Ci/mmol, radiochemical purity of 98%). Fish embryos and larvae were divided into a no treatment control, a solvent vehicle control (acetone < 0.05%), and 5 concentrations of TCDD. The TCDD concentrations in water and in tissue were measured by a scintillation counter (Tracor Analytic Mark III) with a detection limit of 0.001 ng/g. Fathead minnow embryos were exposed at the blastula stage in a static non-renewal system. The concentrations of TCDD at the time of embryo exposure were 0.37, 0.59, 1.2, 2.83 and 10.16 ng/L. The embryos were examined daily through development for the appearance of lesions and up to 48 hs after hatching. Larvae of one month old (0.0017-0.0094) were exposed for 24 hr to TCDD at concentrations of 3.8, 40.0, 68.6, 78.48 and 90.0 ng/L. Then, larvae were transferred into clean water tanks and sacrificed at day 64 after exposure. Growth, appearance of a wasting-type syndrome, and death were noted.

3. Results and Discussion

The lowest adverse effect level (LOAEL) in the embryos of the fathead minnow was shown at a tissue dose of 0.04 ng/g and the LD50 (the measured TCDD tissue dose in eggs or embryos that caused 50% of death) was at a tissue dose of 25.71 ng/g TCDD (wet weight)⁴⁾. When compared these findings with the ones in lake trout, the sac fry stage had 23% mortality at a TCDD tissue dose of 0.040 ng/g, an ER50 (residual concentration that produces effect on

50 % of the population) of 0.065 ng/g, and a LOAEL of 0.055 ng/g (48 hr of exposure)¹⁾. Gross lesions such as multifocal hemorrhages, edema, and deformation of the lower jaw were observed in the fathead minnow. These lesions were classified as mild and severe due to their onset and their pathogenesis. Other toxicity studies have reported similar types of lesions in the rainbow trout, lake trout, northern pike, mummichog and medaka embryos³⁾.

Fathead minnow larvae exposed to TCDD had a non-observable adverse effect level (NOAEL) at a tissue dose of 4 ng/g. Significant differences in growth and wasting-type syndrome were shown at a tissue dose of 20 ng/g, and 100% mortality at 163 ng/g (wet weight)⁵⁾. The tissue doses are in a similar range to earlier reports on fathead minnows of 0.5-1.5 g, in which 100% mortality was found with 2.4-1,280 ng/g TCDD (dry weight)⁶⁾. Signs of toxicity such as fin necrosis, body weight loss and lethargy in fathead minnow larvae are similar to other studies in which the same or different species, systems and/or experimental designs were applied³⁾.

The comparison of the results in the present study with other acute exposures of embryo and larvae < 8 g are summarized in Table 1. Table 1 is a modification of Table 4-1 that appeared in the EPA 1993 Interim Report³⁾. It has been updated with the latest studies on the effects of TCDD in early life stages of fathead minnows.

4. Conclusions

- a) The relative toxicity to the early life stages in fish based on body burden are: embryo>larvae>juveniles. Adult fish are the least sensitive stage to TCDD.
- b) In a number of fish species, the toxicity in embryos is associated with vascular lesions which may determine the death of the organism, while the toxicity in larvae is associated with a decrease in growth and the occurrence of a wasting-type syndrome.
- c) The fathead minnow embryos and larvae (0.0017-0.0094 g) are one of the most sensitive fish species to TCDD based on body burden.
- d) The LOAEL in fathead minnows embryos was 0.04 ng/g, and the NOAEL in larvae was 4 ng/g TCDD. These concentrations in feral fish have only been observed in highly contaminated areas. Therefore, it is unlikely that in less contaminated areas, fathead minnow body burdens would reach toxic levels.

Table 1. Literature summary of the toxicological effects during the acute exposures on fish embryos and larvae to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin. (Modified from EPA 1993³⁾)

| Test Species | Test Method | Water Conc. (ng/L) ^a | Organism Conc. (ng/g) ^b | Exposure | Duration | Observation | Effect | Reference ^c |
|--|-------------|---------------------------------|------------------------------------|-----------|----------|----------------|-------------------|--------------------------|
| Rainbow trout (<i>Oncorhynchus mykiss</i>) | | | | | | | | |
| eggs | egg | | 0.230 | single | | Fertilized egg | LR50 ^d | Walker and Peterson 1991 |
| McConaughy strain | inject. | | (in eggs) | injection | | to swim-up | inc fry | |
| eggs | egg | | 0.240 | single | | Fertilized egg | LR50 ^d | Walker and Peterson 1991 |
| Erwin strain | inject. | | (in eggs) | injection | | to swim-up | inc fry | |
| eggs | egg | | 0.374 | single | | Fertilized egg | LR50 ^d | Walker and Peterson 1991 |
| Arlee strain | (injection) | | (in eggs) | injection | | to swim-up | inc fry | |
| eggs | egg | | 0.488 | single | | Fertilized egg | LR50 ^d | Walker and Peterson 1991 |
| Eagle Lake strain | (injection) | | (in eggs) | injection | | to swim-up | inc fry | |
| eggs | egg | | 0.421 | single | | >48h to post | LR50 ^d | Walker et al. 1992 |
| Fish Lake strain | (injection) | | (in eggs) | injection | | swim-up | inc fry | |

| continuation | | | | | | | | |
|--|-------------------|---------------------------------|------------------------------------|--------------------|----------|---------------------------------|---|---------------------------------------|
| Test Species | Test Method | Water Conc. (ng/L) ^a | Organism Conc. (ng/g) ^b | Exposure | Duration | Observation | Effect | Reference ^c |
| Rainbow trout eggs | water (renewal) | | 0.279 (in eggs) | 48h | | >48h to post swim-up | Significant mortality in sac fry | Walker <i>et al.</i> , 1992 |
| eggs | water (renewal) | | 0.439 (in eggs) | 48h | | >48h to post swim-up | LD50 ^d sac fry | Walker <i>et al.</i> , 1992 |
| swim-up fry (0.38 g) | water (flow-thru) | 0.176 | 3.22 | 28d | | 28d | 95% mortality | Mehrlé <i>et al.</i> , 1988 |
| swim-up fry (0.38 g) | water (flow-thru) | 0.0011 | 0.021 ^e | 28d | | 28d | NOAEL ^d | Mehrlé <i>et al.</i> , 1988 |
| swim-up fry (0.38 g) | water (flow-thru) | 0.038 | 0.765 ^e | 28d | | 28d | LOAEL ^d 45% mortality | Mehrlé <i>et al.</i> , 1988 |
| fingerling (3-7g) | diet (0.494 ng/g) | | 0.25 | 13wk | | 13wk | No toxic effect | Kleeman <i>et al.</i> , 1986a |
| fingerling (8g) | i.p. injection | | 10 | single injection | | 2-4wk post exposure | Fin necrosis, no effect on immune suppression | Spitsbergen <i>et al.</i> , 1986 |
| Lake trout (<i>Salvelinus namaycush</i>) eggs | water (renewal) | | 0.034 (in eggs) | 48h | | >48h to post swim-up | NOAEL ^d | Walker <i>et al.</i> , 1991 |
| eggs | water (renewal) | | 0.040 (in eggs) | 48h | | >48h to post swim-up | 23% mortality in sac fry | Spitsbergen <i>et al.</i> , 1991 |
| eggs (renewal) | water (in eggs) | | 0.055 | 48h swim-up | | >48h to post mortality | LOAEL ^d sac fry | Walker <i>et al.</i> , 1991 |
| eggs (renewal) | water (in eggs) | | 0.065 | 48h swim-up | | >48h to post | LD50 ^d sac fry | Walker <i>et al.</i> , 1991 |
| eggs (injection) | eggs (in eggs) | | 0.047 injection | single swim-up fry | | Fertilized egg to | LD50 ^d sac fry | Walker <i>et al.</i> , 1992 |
| Fathead minnow (<i>Pimephales promelas</i>) eggs | water (static) | 0.37 | 0.04 | | | fertilized egg to 2d post hatch | LOAEL ^d embryo with lesions | Olivieri and Cooper 1995 ^h |
| eggs | water (static) | 1.2 | 0.16 | | | fertilized egg to 2d post hatch | ED50 ^d (0.14 ng/g) | Olivieri and Cooper 1995 ^h |
| eggs | water (static) | 0.59-10.16 | 2.46-37.01 | | | fertilized egg to 2d post hatch | LD50 ^d (25.71 ng/g) | Olivieri and Cooper 1995 ^h |
| larvae (0.0017-0.0094g) | water (static) | 3.8 | 4 | 24h | | 64d | NOAEL ^d | Olivieri and Cooper 1994 ^h |
| larvae (0.0017-0.0094g) | water (static) | 90 | 20 | 24h | | 64d | Decreased growth, wasting-type syndrome | Olivieri and Cooper 1994 ^h |
| larvae (0.0017-0.0094g) | water (static) | 40 | 26 | 24h | | 64d | 5.9% mortality, wasting-type syndrome | Olivieri and Cooper 1994 ^h |
| larvae (0.0017-0.0094g) | water (static) | 78.48 | 45 | 24h | | 64d | 32% mortality, decreased growth, wasting-type syndrome | Olivieri and Cooper 1994 ^h |
| larvae (0.0017-0.0094g) | water (static) | 68.6 | 163 | 24h | | 32d | 100% mortality, decreased growth, wasting-type syndrome | Olivieri and Cooper 1994 ^h |

| Test Species | Test Method | Water Conc. (ng/L) ^a | Organism Conc. (ng/g) ^b | Exposure Duration | Observation | Effect | Reference ^c |
|--|-----------------|---------------------------------|------------------------------------|---------------------------------|-------------|--|-----------------------------|
| Fathead minnow juvenile (0.5-1.5g) | water (renewal) | 83 | 2.4-1,280 ^d | 24h | 4kd | 100% mortality | Adams <i>et al.</i> 1986 |
| Japanese medaka (<i>Oryzias latipes</i>) eggs | water (static) | | 0.240 (in embryo) | fertilized egg to 3d post hatch | | ER50 ^e (embryos with lesions) | Wisk and Cooper 1990b |
| Yellow perch (<i>Perca flavescens</i>) juvenile (3-6g) | diet | | 0.143 | 13wk | 13wk | No toxic effect | Kleeman <i>et al.</i> 1986b |

^a Measured TCDD concentration in water.
^b Measured TCDD concentration in organism based on wet weight.
^c For literature citation referred to EPA 1993 Interim Report³.
^d LR50 (corrected for control mortality) The measured residue concentration in eggs that caused 50% mortality to sex. fry.
^e ER50 The measured residue concentration in eggs that caused 50% effect.
 LD50 The measured TCDD tissue dose in eggs or larvae that caused 50% death.
 ED50 The measured TCDD tissue dose in eggs or larvae that caused 50% effect.
^f NOAEL = No Observed Adverse Effect Level
 LOAEL = Lowest Observed Adverse Effect Level
^g NOAEL and LOAEL values calculated for the Interim Report³.
^h Measured TCDD concentration in organism based on dry weight.

5. References

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