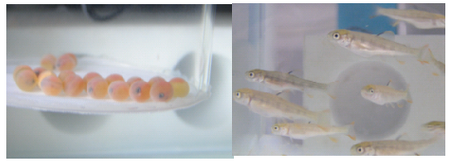

Some EcoTox and PK Data For Ammonium Perfluorohexanoate (APFHx)

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Fish, Early Life Stage Toxicity Test to Oncorhynchus mykiss (Rainbow Trout)

Introduction

The objective of the study was to establish the effect of PFH Ammonium Salt on the growth and development of embryos and larvae of the freshwater fish species Oncorhynchus mykiss (Rainbow trout) in a Fish Early-Life Stage (ELS) Toxicity Test. The study was performed in accordance with the OPPTS Biological Effect Test Guideline No. 850.1400, Fish Early-Life Stage Toxicity Test (1996).

The test was conducted with a flow through test design.

Concentrated stock solutions and test media were analysed during the ELS test. Measured concentrations of PFH active moiety in the concentrated stock solutions ranged between 98 and 100% of the nominal concentrations, test media concentrations ranged between 94 and 108% of nominal corresponding to geometric mean measured concentrations of 0.103, 0.310, 0.916, 3.14 and 10.1 mg/L.

Table 1. Hatching success and fish larval survival during the fish early-life stage test

concentration (mg/L)	hatched larvae (%)	Larvae surviving at 28days (%)
0	74	93
0.1	81	96
0.3	68	100
1.0	63	96
3.0	66	100
10.0	65	99

Table 2. Total Length of and Dry Weight of Fish - 28 Days Post-Hatch

concentration (mg/L)	Total Length of Fish (cm)	Dry Weight of Fish (g)
0	3.4 ± 0.23	0.0564 ± 0.0126
0.1	3.5 ± 0.17	0.0583 ± 0.0129
0.3	3.4 ± 0.20	0.0558 ± 0.0110
1.0	3.4 ± 0.19	0.0571 ± 0.0097
3.0	3.5 ± 0.15	0.0578 ± 0.0087
10.0	3.4 ± 0.19	0.0588 ± 0.0102

RESULTS

Hatching Success

Hatching success in the control group was 74%. As this exceeded 66%, the validity criterion for hatching success was satisfied. First egg hatch in treatment and control vessels occurred in the 24-hour period between the Day 25 and Day 26 pre-hatch observation timepoints. This indicated no difference in time to first hatch across all treatments when compared to the control group.
NOEC and LOEC were determined as nominal concentrations of 9.96 and >9.96 mg/L as PFHx active moiety.

Survival

Larval survival until Day 28 post-hatch in the control group exceeded 70% (93%) thereby satisfying the validity criterion for hatching success.
Post-hatch larval survival across all remaining treatments ranged between 96 and 100%.
In terms of measured concentrations, the NOEC and LOEC for post-hatch larval survival until Day 28 were both considered to be equal to or greater than 10.1 mg/L (highest mean measured concentration).

Fish Total Lengths and Wet Weights

The NOEC and LOEC for both total fish length and fish weight were determined on Day 28 post-hatch and were considered to be 9.96 and >9.96 mg/L respectively.

Abnormalities

There were no dose related abnormalities recorded during the test.

Water Quality and Environmental Conditions

All water quality parameters were within the specified ranges stated in the Guidelines.

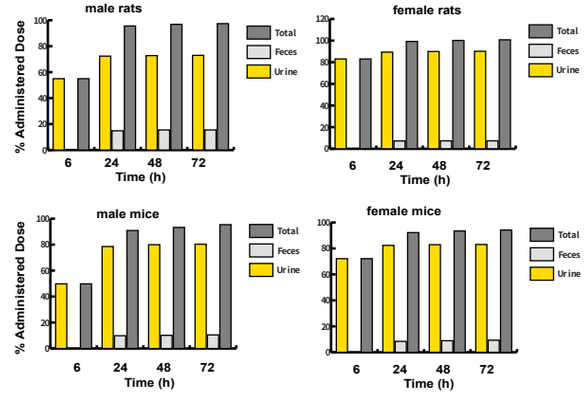
(This fish study is currently under review at EPA.)

& The Excretion and Tissue Distribution of [14C]- APFHx in the Mouse and the Rat Following a Single and Multiple Oral Administration at 50 mg/kg.

Tier 1

a Single Oral Dose of [14C] APFHx to Rats and Mice at a Target Dose level of 50 mg/kg

Fig 1. Recovery of Total Radioactivity Following a Single Oral Administration



Results expressed as % administered dose
Total includes cagewash and expired air

RESULTS

Tier.1 a single oral dose

Irrespective of sex or species, following a single oral administration, total radioactivity excretion was rapid, with mean recoveries of over 90% of the dose at 24 h post dose. The major route of elimination was via the urine (means of 73.0-90.2% of the dose), followed by the feces (mean of 7.0-15.5%). Elimination via expired air was negligible.

At 72 hours post dose in rats, mean recoveries of total radioactivity were 97.4 and 100.8% in males and females respectively, with approximately 0.2% remaining in the gastrointestinal tract and carcass.

At 72 hours post dose in mice, mean recoveries of total radioactivity were 95.4 and 97.3% in males and females respectively, with approximately 0.6-0.9% remaining in the gastrointestinal tract and carcass.

Tier.2 a multiple (14daily doses) oral dose

Irrespective of sex or species, a multiple (13 daily doses) oral administration of APFHx followed by a single oral administration of [14C]-APFHx, total radioactivity excretion was rapid, with mean recoveries of over 90% of the dose administered (and with mean values >95% of the ultimately recovered material) at 24 h post dose. The major route of elimination was via the urine (means of 77.8-83.4% of the dose), followed by the feces (mean of 9.6-12.9%), indicating that the majority of the administered dose had been absorbed.

At 168 h post dose in rats, mean recoveries of total radioactivity were 95.1 and 93.7% in males and females respectively, indicating that the dose was almost completely excreted, with only approximately 0.2% remaining in the gastrointestinal tract and carcass.

At 168 h post dose in mice, mean recoveries of total radioactivity were 97.0 and 96.4% in males and females respectively, indicating that the dose was almost completely excreted, with only approximately 0.1% remaining in the gastrointestinal tract and carcass.

At 168 h post dose in rats and mice, radioactivity was generally very low or below the limit of detection in most tissues. Tissue concentrations were below blood concentrations with the exception of liver, which was approximately 4-8 times higher than the circulating blood level. Elevated levels of measurable radioactivity in the liver are consistent with its role in metabolism and excretion.

At 12 h post dose in rats, mean plasma concentrations were to 0.8 and 0.4 mg/mL in males and females, respectively. By 24 h post dose, mean plasma values decreased to 0.5 and 0.3 mg/mL in males and females, respectively.

At 12 h post dose in mice, mean plasma concentrations were to 1.3 and 1.0 mg/mL in males and females, respectively. By 24 h post dose, mean plasma values decreased to 1.0 and 0.5mg/mL in males and females, respectively.

Tier 2

a Multiple (13 Daily Doses) Oral Administration of APFHx Followed by a Single Oral Administration of [14C] APFHx to Rats and Mice at a Daily Target Dose level of 50 mg/kg

Fig 2. Recovery of Total Radioactivity Following a Multiple Oral Administration

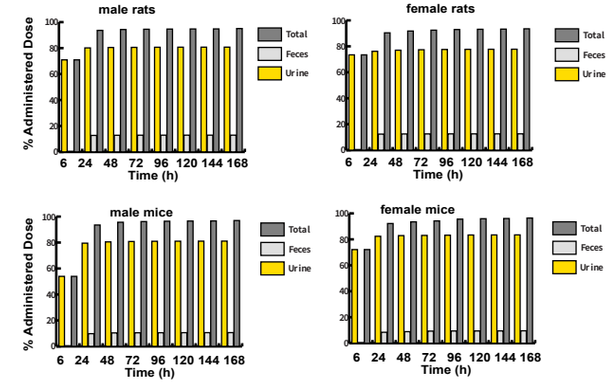


Table 4. Concentration of Total Radioactivity in Tissues at 168 h Post Dose

Sample	Male Rats	Female Rats	Male Mice	Female Mice
Fat-White	0.03	0.03	0.03	0.04
Kidneys	0.11	0.13	0.05	0.05
Liver	1.16	0.85	0.70	0.61
Spleen	0.03	0.04	0.02	0.02
G.I. Tract	0.03	0.03	0.04	0.03
Carcass	0.10	0.10	0.06	0.04
Wh Blood	0.15	0.16	0.17	0.17

Results expressed as % administered dose

Table 5. Plasma Concentrations of Total Radioactivity Following a Multiple Oral Administration

Timepoint (h)	Male Rats	Female Rats	Male Mice	Female Mice
12	0.8 ± 0.1	0.4 ± 0.1	1.3 ± 0.7	1.0 ± 0.5
24	0.5 ± 0.0	0.3 ± 0.1	1.0 ± 0.3	0.5 ± 0.1

Results expressed as % administered dose

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