

Octamethylcyclotetrasiloxane (CAS no. 556-67-2)

Tier 1 *in vivo* Test

Fish Short Term Reproduction Assay (FSTRA, OECD TG 229) using Medaka (*Oryzias latipes*)

(1) Results

Fish were exposed to concentrations of 0.221, 2.18 and 22.0 µg/L (measured). No significant differences were observed in mortality, female total length, body weight, number of eggs, number of fertile eggs, gonadosomatic index, female hepatosomatic index and female secondary sex characteristics.

At 0.221 µg/L and higher, a significant decrease was observed in male total length, and a significant increase was observed in female hepatic vitellogenin level.

At 2.18 µg/L and higher, a significant decrease was observed in female total length.

At 22.0 µg/L, a significant decrease was observed in fertility rate, and significant increases were observed in male hepatic vitellogenin level, male hepatosomatic index and male secondary sex characteristics.

(2) Summary

Estrogenic activity of octamethylcyclotetrasiloxane has been indicated from literature and Tier 1 *in vitro* tests. In this study, increase in male hepatic vitellogenin level was observed at sublethal concentrations to suggest estrogenic effect.

On the other hand, it was not concluded that octamethylcyclotetrasiloxane is an antiestrogenic or androgenic compound.

Any adverse exposure level was not clearly determined for growth, since no significant decrease in body weight was observed.

Additionally, any adverse exposure level was not clearly determined for reproduction, since no significant decrease in fertility rate was observed between each exposure group and solvent control.

The no observed adverse effect level of 22.0 µg/L was ca. 270 times as high as the highest environmental water concentration of 0.082 µg/L for MOE's Environmental Survey and Monitoring of Chemicals in FY 2021.

Table 1-A Results

Measured concentration ($\mu\text{g/L}$)	Number of tested fish		Total length (mm)		Body weight (mg)		Total length (mm)	
	male	female	male	female	male	female	male	female
Control	12	12	0	0	32.6 \pm 1.5	31.8 \pm 0.8	383 \pm 49	408 \pm 66
Solvent Control	12	12	0	0	33.3 \pm 1.2	32.1 \pm 1.4	388 \pm 49	417 \pm 52
0.221	12	12	0	0	32.0 \pm 1.0*	31.7 \pm 0.9	386 \pm 34	424 \pm 42
2.18	9	9	11	0	32.6 \pm 1.6*	31.1 \pm 1.0*	387 \pm 60	400 \pm 38
22.0	12	12	0	0	31.9 \pm 1.2**	30.7 \pm 0.9*	384 \pm 48	401 \pm 24

Table 1-B Results (continued)

Measured concentration ($\mu\text{g/L}$)	Number of eggs (eggs/female/day)	Number of fertile eggs (eggs/female/day)	Fertility rate (%)	Gonadosomatic Index (%)	
				male	female
Control	1,600 \pm 154	1,562 \pm 163	97.4 \pm 1.5	0.774 \pm 0.16	9.16 \pm 3.1
Solvent Control	1,407 \pm 88	1,362 \pm 76	96.9 \pm 1.3	0.779 \pm 0.25	8.25 \pm 1.3
0.221	1,647 \pm 101	1,560 \pm 131	94.7 \pm 2.5	0.776 \pm 0.28	8.91 \pm 0.95
2.18	1,551 \pm 216	1,488 \pm 248	95.6 \pm 3.1	0.803 \pm 0.18	8.81 \pm 1.2
22.0	1,419 \pm 179	1,362 \pm 179	96.0 \pm 1.0(*)	0.695 \pm 0.17	8.69 \pm 1.0

Table 1-C Results (continued)

Measured concentration ($\mu\text{g/L}$)	Hepatosomatic Index (%)		Vitellogenin (ng/mg liver)		Secondary sex characteristics	
	male	female	male	female	male	female
Control	2.40 \pm 0.69	5.31 \pm 1.6	0.629 \pm 0.75	1,004 \pm 747	90 \pm 12	0 \pm 0
Solvent Control	2.01 \pm 0.53	5.89 \pm 0.71	ND	941 \pm 310	97 \pm 13	0 \pm 0
0.221	2.13 \pm 0.30	6.12 \pm 1.1	ND	1,303 \pm 222*	103 \pm 17	0 \pm 0
2.18	2.11 \pm 0.40	6.47 \pm 0.61	0.53 \pm 0.57	1,446 \pm 221*	98 \pm 13	0 \pm 0
22.0	2.80 \pm 0.48**	6.00 \pm 1.5	1,003 \pm 687*	1,576 \pm 321*	102 \pm 15(*)	0 \pm 0

Table 1-D Results (continued)

Measured concentration ($\mu\text{g/L}$)	Other observations
Control	Not found
Solvent Control	Not found
0.221	Not found
2.18	Abnormal swimming behaviors (lethargy and surface swimming) were observed in a fish on the 11 th day of exposure but were not found on the 12 th day and later.
22.0	Not found

Data show mean \pm SD (standard deviation)

Statistically significant differences from solvent control group (** p <0.01, * p <0.05)

ND: not detected (below detection limit of vitellogenin: 0.4 ng/mg liver)

Secondary sex characteristics: number of joint plates with papillary processes