Lethal concentration 50 of Glutaraldehyde and Trichlorfon: differential concentration on zebrafish (*Danio rerio*)

Chaluch Limpasontipong ^a Tanwarat Choojit ^a, Tasneem Tosaman ^a, Janista Jamnong ^a, Thanakrit Ratanaprapaporn ^a, Thanakrit Manujantarat ^c, Suwaree Kitikiew^e, Sahabhop Dokkaew ^{a,b} and Phunsin Kantha ^{a,d*}

- ^aDepartment of Aquaculture, Faculty of Fisheries, Kasetsart University, Bangkok, Thailand
- bOrnamental Fish and Aquatic Plant Research and Technology Transfer Center, Faculty of Fisheries,
- Kasetsart University, Bangkok, Thailand
- ^cPCN Marketing & Distribution Co., Ltd, Samut Sakhon, Thailand
- ^dSamut Songkharm Fisheries Research Station, Kasetsart University, Samut Songkhram, Thailand
- ^eDepartment of Animal Production Technology and Fisheries, Faculty of Agricultural Technology, King Mongkut's Institute of Technology Ladkrabang, Bangkok, 10520, Thailand.

Abstract

- •Chemical agents are vital for quarantine and disinfection in ornamental fish management.
- •They ensure fish health and prevent disease spread.
- •Glutaraldehyde and Trichlorfon are commonly used due to their pathogen control efficacy.
- •The study focuses on these chemicals, examining LC50 values with zebrafish (Danio rerio).
- •Zebrafish is chosen for its genetic similarity to other vertebrates and transparent embryos.
- •LC50 indicates the lethal concentration for 50% of the organism under specified conditions.
- •Glutaraldehyde's LC50 is approximately 5-6 mg/L.
- Trichlorfon's LC50 is around 58 mg/L.
- •These values are crucial for determining safe and effective dosages in fish quarantine and disinfection.

Keywords: Glutaraldehyde, Lethal Concentration 50, Danio rerio, Trichlorfon, Toxicity

Introduction

The findings from the experiments conducted on glutaraldehyde toxicity revealed a diverse range of LC50 values across different fish species and exposure durations, such as *Cyprinodon variegatus*, *Lepomis macrochirus*, *Oncorhynchus kisutch*, *Oncorhynchus mykiss*, *Pimephales promelas* and *Takifugu bimaculatus* (Pereira et al., 2014).

The results of previous LC50 trials conducted to assess the effects of Trichlorfon on various fish species such as follows: *Poecilia latipinna*, *Poecilia reticulata*, *Gambusia holbrooki* and *Xiphophorus helleri*. These findings contribute to our understanding of the potential impact of Trichlorfon on diverse fish species across varying exposure durations, providing valuable insights for environmental risk assessments and aquatic ecosystem management (Vajargah & Hedayati, 2014).

Material and Methods

Quarantine Danio rerio for 1 week

↓Put 10 fish in each tank

Put chemical in tank

Collecting data of mortality

Reference

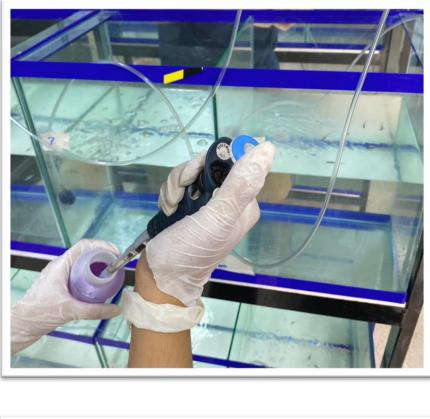
- Coelho, S., Oliveira, R., Pereira, S., Musso, C., Domingues, I., Bhujel, R. C., Soares, A. M., & Nogueira, A. J. 2011. Assessing lethal and sub-lethal effects of trichlorfon on different trophic levels. Aquatic toxicology, 103(3-4), 191-198.
- Duncan, W. P., Idalino, J. J. S., da Silva, A. G., Moda, R. F., da Silva, H. C. M., Matoso, D. A., & Gomes, A. L. S. 2020. Acute toxicity of the pesticide trichlorfon and inhibition of acetylcholinesterase in *Colossoma macropomum* (*Characiformes: Serrasalmidae*). Aquaculture International, 28, 815-830.
- Jian-Shao, W., Lei-Bin, L., Zhi-Huang, Z., Jian-Xing, Z., & Jian-Tao, Y. 2017.

 Acute toxicity of six drugs to *Fugu bimaculatus*. Journal of Fisheries Research, 39(6), 455.
- Lucini, C., D'Angelo, L., Cacialli, P., Palladino, A., & De Girolamo, P. 2018.

 BDNF, brain, and regeneration: insights from zebrafish. International journal of molecular sciences, 19(10), 3155.
- McDonnell, G., & Russell, A. D. 1999. Antiseptics and disinfectants: activity, action, and resistance. Clinical microbiology reviews, 12(1), 147-179.
- Miyawaki, I. 2020. Application of zebrafish to safety evaluation in drug discovery. Journal of toxicologic pathology, 33(4), 197-210.
- O'Brien, P. J., Siraki, A. G., & Shangari, N. 2005. Aldehyde sources, metabolism, molecular toxicity mechanisms, and possible effects on human health.

 Critical reviews in toxicology, 35(7), 609-662.







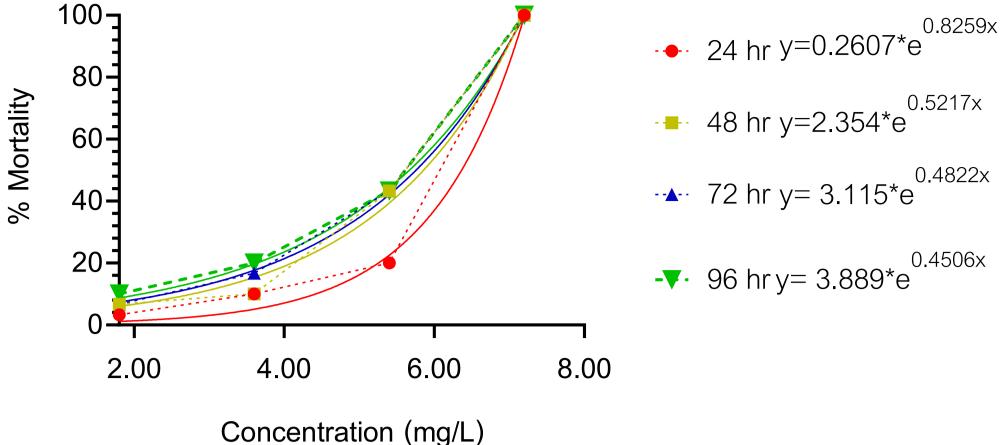


Fig. 1. Mortality curve of Zebra fish (*Danio rerio*) in Glutaraldehyde (24 hr,48 hr,72 hr and 96 hr)

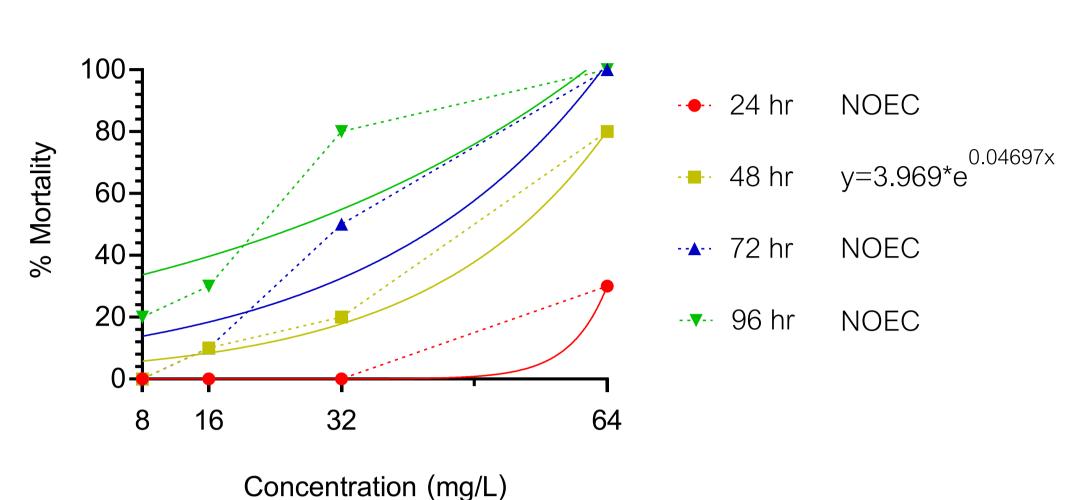


Fig. 2. Mortality curve of Zebra fish (Danio rerio) in Trichlorfon (24 hr,48 hr,72 hr and 96 hr)

 Table 1 acute toxicity of Glutaraldehyde and Trichlorfon to Zebra fish (Danio rerio)

	Concentration (mg/L)	% Mortality				LC50 (mg/L)			
		24 hr	48 hr	72 hr	96 hr	24 hr	48 hr	72 hr	96 hr
Glutaraldehyde	1.8	3.3	6.7	6.7	10.0				
	3.6	10.0	10.0	16.7	20.0	6.36	5.86	5.76	5.67
	5.4	20.0	43.3	43.3	43.3				
	7.2	100.0	100	100	100				
Trichlorfon	8	0.0	0.0	0.0	20.0				
	16	0.0	10.0	10.0	30.0	NOEC	53.94	NOEC	NOEC
	32	0.0	20.0	50.0	80.0				
	64	30.0	80.0	100.0	100				

NOEC: No Effective Concentrations

Result and Discussion

Glutaraldehyde LC₅₀

Upon introducing Glutaraldehyde into the water, an examination of water parameters including temperature, dissolved oxygen (DO), and pH revealed negligible alterations. Glutaraldehyde concentrations 1.8, 3.6, 5.4 and 7.2 mg/L were meticulously selected to investigate mortalities and ascertain the LC50 in *D. rerio*. Calculation of the LC50 values involved determining the percentage of mortalities for each dose, as illustrated in Fig 1. The LC50 values for Glutaraldehyde in D. rerio at 24, 48, 72, and 96 hours, accompanied by 95% confidence limits, were estimated at 6.36, 5.86, 5.76, and 5.67 mg/L, respectively, as detailed in Table 1.

Trichlorfon LC₅₀

Following the introduction of Trichlorfon into the water, an assessment of water parameters encompassing temperature, dissolved oxygen (DO), and pH revealed no discernible changes. Trichlorfon concentrations ranging from 8, 16, 32 and 64 mg/L to investigate mortalities and the LC50 in *D. rerio*. Computation of the LC50 values involved the utilization of the mortality percentage corresponding to each dosage (Fig 2). However, for Trichlorfon in *D. rerio*, the LC50 values, accompanied by 95% confidence limits is 58.94 mg/L at 48 hr, were solely estimated for the 48-hour time point, as the range of the results proved to be excessively broad, as outlined in Table

Conclusion

The LC50 results for Glutaraldehyde, measured at 24, 48, 72, and 96 hr, fall within the range of 5.67 to 6.36 mg/L, while Trichlorfon exhibits an LC50 of 58.94 mg/L at the 48 hr. These concentrations were determined as LC50 in adult zebrafish experiments. The obtained data serves as a valuable reference for the application of Glutaraldehyde and Trichlorfon in the treatment of parasites and pathogens affecting ornamental fish and in aquaculture fish. The results advance our comprehension of optimal dosage levels for addressing health concerns in aquatic environments, thereby improving the overall management of fish health and well-being.



