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APPLICATION OF TOXICITY TESTING TECHNIQUE: DETERMINATION OF LC50 OF ARSENIC ON SOME MARINE ORGANISMS

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ABSTRACT Results obtained from toxicity testing experiments on Artemia (1998), Tiger prawn (Penaeus monodon) (1998-1999) and Seabass (Lates carcarifer) (1998-1999) led to determine the LC50 values of Arsenic on them with 24-h, 48-h, 72-h, and 96-h exposure. LC50-s tended to decrease by the exposure duration. In the same exposure duration, LC50 value of Seabass showed smaller than that of Tiger prawn and both values were smaller than that of Artemia. It means that the sensitivity to lethal toxic effect of Arsenic showed decreasing by following order: Seabass > Tiger prawn > Artemia.

ÖNG DUNG KY THUAT KIEM ÑÒNH ÑOC TO: XAC ÑÒNH NONG ÑO GAY CHET 50% (LC50) CUA ASENIC ÑOI VÔI MOT SO SINH VAT BIEN

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TOM TAT Cac ket qua thu ñöôc tö nhöng lo kiem ñònh ñoc to tren Artemia (1998), tom Su (Penaeus monodon) (1998-1999) va ca Chem (Lates carcarifer) (1999) ña xac ñònh ñöôc gia trò LC50 cua Arsenic ô 24h, 48h, 72h va 96h. Cac gia trò LC50 giam dan theo thôi gian sinh vat ñöôc kiem ñònh qua tiep xuc vôi Arsenic. LC50 cua ca Chem nho hôn cua tom Su va cung nho hôn cua Artemia ô cung thôi ñiem kiem ñònh, co nghóa la möc nhay cam ñoc to gay tö vong Arsenic cung giam dan theo thö tö ca Chem > tom Su > Artemia.

INTRODUCTION

Aquatic Toxicology is a scientific branch of Ecotoxicology [4, 5, 6] and has been created around more than 20 years ago due to environment pressing issues caused by human activities. It has been developed and utilized effectively in many developed countries in different aspects, especially as the role in assessing ecological risks caused by environmental impacts. Aquatic Toxicology is the one of important establishments that can assist environmental managers giving appropriate decision makers in environmental management. It also helps approaching criteria concentrations of environmental pollutants. Preeminent characteristic of this technique is "desirable in water pollution evaluation because chemical and physical tests alone are not sufficient to assess potential effects on aquatic biota" [2]. Aquatic Toxicology can help us to determine more correctly and clearly responses of aquatic organisms against toxic pollutants, meaning it can help in ecological risk estimation of bioavailable toxins to natural living resources. Furthermore, this technique does not require as big expense as using physical and chemical methods.

LC50 is already known as one of important toxicity testing endpoints that is used to estimate lethal effect on organisms. In period of 1992 – 1995, scientists at the

Institute of Oceanography – Nhatrang (ION) developed Toxicity Testing Technique as to estimate toxic effect of several compounds such as potassium dichromate, copper sulphate and some drilling fluids, dispersant on sea urchin fertilization, Artemia, Tiger prawn juvenile, bivalves...[8, 9]. From 1997 to 1999, under the support by Canadian International Development Agency (CIDA) in framework of Cooperative Programme on Marine Science phase II (CPMS-II), Laboratory of Toxicity Testing at ION has been created and assigned as group for conducting experiments to test lethal effect of Arsenic on marine organisms. First results were presented in the Fourth ASEAN - Canada Technical Conference on Marine Science, Langkawi, Malaysia 1999 [7,12]. This paper is summarizing LC50 values of Arsenic tested on some marine organisms such as Artemia, Tiger prawn, and Seabass have been done mainly in 1999 with expectation to inform that application of Toxicity Testing Technique in Vietnam has successfully carried out. Furthermore, this is hoped to be practical and scientific bases in order to contribute to develop Environmental Criteria as well as to assess ecological risks for marine environment in Vietnam as well as in SE Asian region.

MATERIALS AND METHODS

Tested organisms

-Artemia (Artemia sp.): Artemia nauplii were used to test after 20-24h hatching. Artemia source was purchased from Aquatic Lifeline Inc. U.S. by importing.

-Tiger prawn (Penaeus monodon): PL 5-26 stage Tiger prawns were used to test and they were supplied by Cuabe Hatchery Station which belongs to the University of Fishery.

-Seabass (Lates carcarifer): samples of 19 - 30 days old were selected from Lig Hatchery Company from Cangio district - Hochiminh City.

Testing materials

1.3248 gram Arsenic salt (As_2O_3) and 2gram of NaOH were both dissolved in 1 L distilled water to get stock solution of 1000 ppm As (1000 mg As/L). LC50 values were calculated at 24, 48, 72 and 96h exposure for Tiger prawn and Seabass, and 24-, and 48-h for Artemia. Testing concentration range of Tiger prawn and Seabass: 1.0, 1.8, 3.2, 5.6, and 10.0 mg As/L; Artemia: 3.125, 6.25, 12.5, 18.0, and 25.0 mgAs/L.

Procedure

Artemia, Tiger prawn and Seabass were tested with type of static acute test. [1, 3, 4, 10, 11]. 100mL volume beakers were used as testing containers for Artemia and Tiger prawn without feeding during the tests. Seabass were kept in 12-L volume aquarium, fed by Artemia larva. All experiments were conducted by 3 replicates at each concentration of solutions. During each experiment also daily measured water quality data as salinity (S‰), dissolved oxygen (DO), pH and temperature (T^0C) using professional meters. Those data should not exceed the acceptable range for culture (Table 1). Collected experimental data were statistically analyzed and calculated LC50 using the EFFL program.

RESULTS AND DISCUSSION

Survival rate of tested organisms at control replicates almost reached 100%, except at 48-h series of Artemia (Table 1) was 93.30%. Mean LC50-s of Artemia at 24-h and 48-h exposure was determined as 18.45 mg As/L and 9.90 mg As/L, respectively (Table 2).

In comparison with obtained results of Nguyen Tac An, et al (1992-1993) [8], bioavailibility of copper was noticeable higher than arsenic - 1.13 mg Cu/L compared to 18.45 mg As/L at 24-h exposure (Table 3).

From Table 2 it could be noted that LC50-s of Tiger prawn and Seabass all decreased temporally, in other words, the longer exposure the less resistant to toxic material, regularly. To combine the results published in 1999 [12] it showed unnoticeable differences

(Table 3). In other words, once again it affirmed relatively high stableness of tests done with Tiger prawn not depending on conducting experiments in different periods of the year.

Table 1: Water quality range at conducted experiments and Survival rates of Artemia, Tiger prawn and Seabass in control series

Species	Artemia	Tiger prawn	Seabass
	(mean values)		
Salinity	33.5-35.1	28.8 - 35.3	28.0 - 35.3
DO	-	4.0 - 7.1	5.4 - 7.2
Temp.	28.2 - 31.7	26.8 - 30.4	25.9 - 30.1
pH	6.9 - 9.1*	7.7 - 9.1*	6.6 - 8.8
24-h	100	100	100
48-h	93.30	100	100
72-h	-	100	100
96-h	-	100	100

Note: (*) high pH affected by Arsenic solution.

Table 2: LC50 values of Arsenic at 24-, 48-, 72-, and 96-h testing on Artemia, Tiger prawn, and Seabass

Test species	24-h LC50	48-h LC50	72-h LC50	96-h LC50
	(mg As/L)			
Artemia franciscana	18.45 ± 4.57 (13.89:23.67)	9.90 ± 3.26 (6.96 - 16.13)		
Penaeus monodon	9.32 ± 1.82 (7.66 -12.00)	6.85 ± 2.29 (4.03 - 9.13)	6.40 ± 2.55 (4.29 - 8.84)	5.89 ± 2.49 (3.44 - 8.84)
Lates calcarifer	8.68 ± 1.07 (7.68 - 10.00)	6.44 ± 1.82 (4.34 -7.48)	5.03 ± 1.25 (3.09 - 5.99)	4.50 ± 1.08 (2.95 - 5.58)

Table 3: LC50 values comparison to references

Testing duration	Artemia franciscana		Penaeus monodon	
	LC50 of As	Reference LC50 *	LC50 of As	Reference LC50 **
24-h	18.45 mg As/l	1.13 mg Cu/l	9.32 mg As/l	9.60 mg As/l
48-h	9.90 mg As/l	-	6.85 mg As/l	7.00 mg As/l
72-h			7.22 mg As/l	-
96-h			6.58 mg As/l	6.70 mg As/l

(*): According to Nguyen Tac An et al, 1996 [8]
(**): According to Vo Hai Thi and Le Hoai Huong 1999 [12]

The above mentioned results seem to reflect sensitivity of tested organisms different in term of evolution. With Arsenic it seemed that species placed higher of evolution was more sensitive to lethal toxic effect. For example, resistance to toxic effect of Seabass was worse than that of Tiger prawn and both of them were worse than that of Artemia (sensitive range: Seabass > Tiger prawn > Artemia). Furthermore, in first 24-h of exposure the LC50-s of Arsenic to tested organisms were significantly larger - 18.45 to 9.90 mg As/L with Artemia; 9.32 to 6.85, 6.40, and 5.89 mg As/L with Tiger prawn and 8.68 to 6.44, 5.03, and 4.50 mg As/L with Seabass.

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