MOLAR ACUTE TOXICITY OF ELEVEN HEAVY METALS ON TWO MARINE CLAMS: *Tapes decussata* and *Venerupis aurea* (Bivalve Mollusk Phylum Veneridae)

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ABSTRACT

The following eleven heavy metal ions aluminum; tin (stannous), cadmium; lead; mercury; dichromate; manganese, nickel; cobalt; zinc; and copper were tested on the two bivalve clams Tapes decussata; and Venerupis aurea specimens taken from Temsah Lake at Ismailia. The acute 96 hr. LC₅₀ values were deduced fron dose probit regression lines. Tapes decussata proved to be more susceptible for toxicity by these eleven heavy metals. Molar LC50 values were calculated as a more precise criterion to compare the ions which widely vary in the ionic weight. Comparison showed that aluminum, tin, and mercury were the most hazardous ions to the two clam species, followed by cadmium, lead and cobalt as medium toxic; while nickel, zinc, cobalt, copper and manganese were relatively the least toxic. Generally, the highly acute toxic metals were also known to be of high capacity to accumulate and cause chronic toxicity to both the marine biota and the consumers. This confers importance of the acute toxicity assessment as a preliminary screening criteria to detect and predict the acute and chronic hazardous chemicals in the environment.

INTRODUCTION

Determining acute toxicity is the first requirement when assessing the toxicological hazards associated with a potential pollutant in the aquatic environment. (Le Blanc, 1984). Any comprehensive hazard assessment will require acute toxicity data using a variety of species occupying several trophic levels. USEPA (1975) lists several species recommended for acute toxicity tests and subdivides them into four basic categories: freshwater vertebrates, freshwater invertebrates, marine and estuarine vertebrates, and marine and estuarine invertebrates.

Water pollution with heavy metals is globally increasing mainly due to the discharge of industrial chemicals, run-off of agricultural chemicals, and the dissolving effect of acid rain on soil minerals. Most of the published data deal with the effect of mineral cations on fresh water fish and biota. On the other hand, few studies are reported dealing with the marine fish. On marine mollusca, scarcely little data are available concerning acute toxicity of heavy metals on marine snails such as the edible short necked bivalve clams.

Saliba et al., (1978) studied the lethal and sublethal effects of mercury salts on the marine mollusca Monodonta articulata. He reported that several auto toxicity studies on molluscs have been performed and reached the conclusion that the molluscs phylum members proved to be particularly highly susceptible to mercuric compounds.

Eisler (1985) described a thorough study on environmental hazards of cadmium to fish, wildlife, and invertebrates in both marine and fresh water biota. The data showed that cadmium is preferably accumulated in molluscs clams in both marine oyster and squid particularly in the soft parts where the Cd⁺² concentration was accumulated up to 782.mg/kg of dry weight of squid liver.

Bioconcentration factor of cadmium from ambient medium by selected species of marine molluscs Aquipecten irradians after 3 weeks of exposure to 10 mg/kg was 131 (Eisler 1971); while it reached 2720 after 40 weeks expasure to ambient Cd concentration of 5 mg/kg. In marine mollusc Crassostrea virginica (Zaroogian and Cheer 1976). Eisler (1985) concluded that a substantial toxicological data base demonstrates that ambient cadmium water concentration exceeding 10 ppb (ug/kg) are associated with high mortality, reduced growth, inhibited reproduction and other adverse effects to aquatic biota.

Stromgren and Bongard (1987), reported that leakage of organotin compounds from hull coating may expose organisms to incuased levels of these toxic substances, mainly tributyltin oxide (TBTO) and tributyltin fluoride (TBTF). Growth rate and shell development are affected in mussel larvae and in Oyster larvae and adults at concentrations down to 0.1 ug l⁻¹, and lethal effects occur at even lower concentrations (Thain, 1983); Waldock and Thain (1983); Beaumont & Budd (1984).

Lau (1991) described the acute toxicity threat of tributyltin antifoulings to the Hong Kong marine environment. He stated that a concentration of as low as 20 parts per trillion (20 ng 1⁻¹) was effective in controlling fouling. Nevertheless, recent research has demonstrated acute toxicity of the tin compounds to the non-target organisms., even down to very minute concentrations in the order of parts per trillion. Its non specificity can cause damage to marine and fresh water organisms ranging from microscopic phytoplankton to crah, shrimp, oyster and fish.

Recently, Ruiz et al., (1995) investigated the acute and chromic toxicity of tributyltin (TBT) to pediveliger larvae of the bivalve clam Scrobicularia plana.

Wong and Dixon (1995) set the criteria for bioassessment of water quality using fish or other sensitive biota to measure the levels of pollutants in water systems.

Therefore, the present study was planned to evaluate the acute toxicity of a selected number of heavy metal cations on the two sensitive clam Snails:- *Tapes decussata*, and *Venerupis aurea* (Bivalve mollusk) which are abundant in the suez canal salt water, and in marine waters. These two clam species are edible and of high economic importance. They are produced in big acqua-forms at Ismailia for exportation.

MATERIALS AND METHODS

Tested Chemicals :-

The following chemicals of pure grade were used:- aluminum chloride, cadmium choride, mercuric chloride, stannous chloride, cobaltous chlorde, lead nitrate, nickel nitrate, zinc sulphate, copper

sulphate, potassium dichromate and manganese phosphate (dibasic), as representatives of the heavy metals.

Bioassay Organisms:-

The benthic bivalves of molluses fauna of marine Vaneridae snails: Venerupis aurea and Tapes decussata were used.

The shell length was measured to the nearest 0.1 cm. This length was considered as an index of size and age of the animals. The average shell length of <u>Venerupis aurea</u> was 2,5 - 3 cm for <u>Tapes decussata</u> was 2,8 - 3,1 cm. At , these measurements, it was easy to detect and collect them acuretly in pelagic areas. These clams were made available from lake Timsah at Ismailia.

Acute toxicity testing:-

The marine benthic bivalves (2 species) were transferred with sea water, in big containers saturated with oxygen until arrival to the Lab., at Alareesh; clams were transferred to a new sea water in a 25 litter glass aquaria and adaptation took place to the lab. conditions for at least 10 days before testing. In a glassbeaker (1L capacity), 10 clams were used in each replicate and 2 replicates were used for each concentration. The sea water was also used for dilutions to one litter in each replicate. That work was done for each tested species. The tests were repeated and the average of mortality counts were recorded after 96 hrs. By using the concentration/mortality data, logarithmic probits relationships were expressed in log/probit regression lines.

The corresponding 96 hrs LC₅₀'s with their confidense limits and regression line slopes were computed according to Finney (1971).

The bioassay technique adopted in this study complies with the standard procedure specified by the American Society for testing and Materials (ASTM)(Anonymous, (1980). The procedure was previously applied by Elsabae (1999).

RESULTS AND DISCUSSION

Comparative acute toxicity to the tested two Marine Clams

The acute 96hr LC₅₀ values of the compared eleven heavy metals to the two bivalves *Tapes decussata* and *Venurupis aurea*, are shown in Tables (1) and (2) respectively. The 96hr LC₅₀ values in terms of mg/l

with the corresponding slopes and confidence limits deduced from the log probit regression lines are tabulated. It can be generally noticed that the clam *Tapes decussata* was shown to be more susceptible to the tested heavy metals than the clam *Venurupis aurea*. However, the two species are known as bottom sediment biota, where they are in continuos exposure to the adsorbed and slowly released heavy metals and other pollutants. This might result in certain degree of tolerance of the living biota on the sediments. Therefore, the 96 hour LC₅₀ values are relatively higher than the more sensitive biota especially the highly susceptible as the shrimps and the sensitive marine fish living at the water surface (Elsabae, 1994).

Table (1): Acute 96 hours LD₅₀'s of Tested Heavy Metals On The Marine clam *Tapes decussata*.

Tested Heavy Metal	96 hr LC ₅₀ mg/l ⁻¹ ppm	Slope	Confidence Limits (95%)
Áluminum Chloride	5.0	0.379	1.315x10 ⁻⁶ 1.9x10 ⁻⁵
Tin(Stannous)Chloride	9.0	0.387	2.94x10 ⁻⁶ 2.75x105 ⁴
Cadmium Chloride	16.0	0.608	5.234x10 ⁻⁶ —4.892x10 ⁻⁵
Lead Nitrate	34.0	0.472	1.112x10 ⁻⁵ 1.039x10 ⁻⁴
Mercuric Chloride	6.0	0.400	1.963x10 ⁻⁶ 1.834x10 ⁻⁵
Potasium Dichromate	40.0	0.525	1.308x10 ⁻⁵ 1.223x10 ⁻⁴
Manganese Phosphate	260.0	0.753	8.505x10 ⁻⁵ 7.948x10 ⁻⁴
Nickel Nitrate	80.0	0.677	2.617x10 ⁻⁵ 2.446x10 ⁻⁴
Cobalt Chloride	400.0	1.040	1.308x10 ⁻⁴ 1.223x10 ⁻³
Zinc Sulphate	409.0	0.722	2.139x10 ⁻⁴ 7.479x10 ⁻⁴
Copper sulphate	240.0	0.671	6.278x10 ⁻⁵ 9.12x10 ⁻⁴

Table (2): Acute 96 hours LD₅₀ Values of Tested Heavy Metals On The Marine clam *Venurupis aurea*

Tested Heavy Metal	96 hr LC ₅₀ mg/l ⁻ ¹ ppm	Slope	confidence Limits (95%)
Aluminum Chloride	8.00	0.268	3.69x10 ⁻⁶ 1.733x10 ⁻⁵
Tin(Stannous)Chloride	50.00	0.307	$2.413 \times 10^{-5} 1.036 \times 10^{4}$
Cadmium Chloride	160.00	0.448	$3.463 \times 10^{-5} - 7.393 \times 10^{4}$
Lead Nitrate	160.00	0.353	5.195×10^{-5} 4.928×10^{4}
Mercuric Chloride	85.00	0.300	$3.992 \times 10^{-5} - 1.842 \times 10^{4}$
Potasium Dichromate	300.00	0.339	6.496x10 ⁻⁵ 1.386x10 ⁻³
Manganese Phosphate	500.00	0.384	1.088×10^{-4} 2.297x10 ⁻³
Nickel Nitrate	470.00	0.284	1.023×10^{-4} 2.16×10 ⁻³
Cobalt Chloride	750.00	0.294	$1.624 \times 10^{-4} 3.464 \times 10^{-3}$
Zinc Sulphate	620.00	0.404	1.346×10^{-4} 2.865 \times 10 ⁻³
Copper Sulphate	500.00	0.375	1.397x10 ⁻⁴ 1.789x10 ⁻³

Table (3): Acute Molar 96 hours LC50 values of Tested Heavy Metals On The Marine Clam Tapes decussata and The

Order of Toxicity.

Tested Aquatic Metal Ion	96 hr LC ₅₀ ug/ ^{l-1} ppm	Order of Toxicity	Ionic weight	Molar LC ₅₀ 's	Order of Toxicit
Aluminum Al ³⁺	5.0	1 <u>st</u>	27.0	0.1851	3 <u>rd</u>
Tin(Stannous)Sn ⁺²	9.0	3 r <u>d</u>	112.4	0.0803	2 <u>nd</u>
Cadmium Cd ⁺²	16.0	4 <u>th</u>	58.0	0.2581	5 th
Lead Pb ⁺²	34.0	5 <u>th</u>	63.5	0.5354	6 <u>th</u>
Mercuric Hg ⁺²	6.0	2 <u>nd</u>	207.2	0.0289	1 <u>st</u>
Chromate Cr ₂ O ₇ ⁻²	40.0	6 <u>th</u>	200.59	0.1995	4 <u>th</u>
Manganese Mn ⁺²	260.0	9 <u>th</u>	55.0	4.7272	11 <u>t</u>
Nickel Ni ⁺²	80.0	7 <u>th</u>	58.8	1.3605	7 <u>th</u>
Cobalt Co ⁺²	400.0	10 <u>th</u>	116.0	3.4481	9 <u>th</u>
Zinc Zn ⁺²	400.0	10 <u>th</u>	118.69	3.3701	8 <u>t</u>
Copper Cu ⁺²	240.0	8 <u>th</u>	65.38	3.6708	10 <u>th</u>

Table (4): Acute Molar 96 hours LD₅₀ values of Tested Heavy Metals On The Marine Clam *Venurupis aurea* and The Order of Toxicity.

Tested Aquatic Metal Ion	96 hr LC ₅₀ mg/ ⁱ⁻¹ ppm	Order of Toxicity	Ionic weight	Molar LC ₅₀ 's	Order of Toxicit
Aluminum Al ³⁺	8.00	1 <u>st</u>	27.0	0.2962	1 <u>st</u>
Tin(Stannous)Sn ⁺²	50.00	2 <u>nd</u>	112.4	0.4448	3 <u>rd</u>
Cadmium Cd ⁺²	160.00	4 <u>th</u>	58.0	2.5855	5 <u>th</u>
Lead Pb ⁺²	160.00	4 <u>th</u>	63.5	2.5193	4 <u>th</u>
Mercuric Hg ⁺²	85.00	3 <u>rd</u>	207.2	0.4102	2 <u>nd</u>
Chromate Cr ₂ O ₇ ⁻²	300.00	5 <u>th</u>	200.59	1.495	3 <u>th</u>
Manganese Mn ⁺²	500.00	7 <u>th</u>	55.0	9.0909	11 <u>th</u>
Nickel Ni ⁺²	470.00	6 <u>th</u>	58.8	7.993	10 th
Cobalt Co+2	750.00	9 <u>th</u>	116.0	6.4655	8 <u>th</u>
Zinc Zn ⁺²	620.00	8 <u>th</u>	118.69	5.2236	7
Copper Cu ⁺²	500.00	7 <u>th</u>	65.38	7.6476	9 曲

Molar Comparison Of The Tested Heavy Metals:

For the precise comparison between the different heavy metals tested it was found that the ionic concentrations must be recalculated in terms of molar values, because the ions are the actual acting toxic forms in the marine water solutions. The new calculations are included in the Tables (3) and (4) respectively. The importance of such correction proved to be essential where the order of toxicity was changed. This is quite clear when the ions widely vary in their ionic weight, such as aluminum (atomic weight 27) when compared to mercury. (atomic weight 207). Therefore the corrected orders of toxicity as shown in tables 3, and 4 (Molar LC₅₀ values) will be more realistic and also more

precise in comparing the hazards of the different ions especially when there is a big variation in the ionic and atomic weights.

Against *Tapes decussata* bivalue, soluble mercuric cation was the most hazardous followed by tin, aluminum, dichromate, cadmium, lead, nickel, zinc, cobalt, copper and manganese in a descending order.

However, the order of toxicity was slightly changed with the second clam snail *Venurupis aurea*; where aluminum comes first, followed by mercury, tin, lead, cadmium, dichromate; zinc; cobalt, copper, nickel; and manganese in a descending order.

The present results coincide with the data published by Saliba et al., (1978) where they concluded that the phylum mollusks are highly susceptible to mercury. Similarly Stromgren and Bongard (1987) indicated the high toxicity of tin as tributyl tin oxide on oysters and mussels. Besides, Eisler (1985) reported the toxicity of cadmium to both marine fish and molluscs. Potte et al., (1991) stated that the acute toxicity of hexavalent Chromium to Rainbow trout was increased with decreasing the PH value. Thus the acidic medium favours more uptake and toxicity of Chromium to marine biota. Izmerov (1984) indicated that daphnia is very susceptible to sodium dichromate.

Finally it can be concluded that regarding the acute toxicity, aluminum, tin, and mercury are the highly hazardous heavy metals to marine biota tested in this study; followed by lead, cadmium; and chromium, while the relatively less toxic were zinc, cobalt, copper, nickel and manganese. On the other havd most of these heavy metals are bioaccumulated and bioconcentrated in the marine biota where their reproductive capacity is mal affected. Besides, these marine biota because they are edible, it will be of high adverse health effect to humans Petrilli and Flora (1978), Bettin et al., (1996), and Shiber (1985).

The value of the acute toxicity assessment is important as a parameter for evaluating the potential hazard of such heavy metals in the environment.

It was also observed that the more acutely toxic heavy metals proved also to be of high potential to accumulate and thus are expected to cause chronic toxicity adverse health effects to the marine biota species and also to the consumers.

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REFERENCES

- Anonymous. Sandard Practice for conducting Acute Toxicity Tests with Fishes, macro invertebrate and amphibians. Amer Soc. For Testing and Materials E 729, March 3 (1980).
- Beaumont, A.R. and M.D. Budd. High Mortality of the larvae of the common mussel at low concentration of tributyltin. Marine pollut. Bull 15 (11): 402-405 (1984).
- Bettin, e., J. Ochlmann, and E. Stroben TBT-Induced Imposex in Marine neogastropods is mediated by an increasing androgen level. Helgolander Meeresunterusuch ungen; 50(3) 298-317 (1996).
- Eisler, R. Cadmium Poisoning in *Fundulus heterochlitus* (Pisces: *Cyprinosontidae*) and other marine organisms. J. Fish Res. Board Canada 28: 1226-1234 (1971).
- Eisler, R. Cadmium Hazards To Fish, Wildlife, and Invertebrates: A synoptic Review. <u>Biological Report</u> 85(1,2). U.S. Dept. of Interior (1985).
- Elsabae, A.A. Comparative susceptibilities of the Alareesh Marine Culture Center Shrimp *Penaeus Japonicus* and the Brine Shrimp *Artemia salina* to Different Insecticides and Heavy Metals. Alex. Sci. Exch. Vol. 15 (3): 425-435 (1994).
- Elsabae, A.A.H. Molecular Acute Toxicity of Eight Heavy Metals To Three Marine Fish Species. J. Pest Control and Environ. Sci. 7(1): 45-58 (1999).
- Finney, D.J. Probit Analysis 3rd Edition London and New York, Cambridge Univ. Press. (1971).
- Izmerov, N.F. editer, Chromium, 68, UNEP, IRPTC, USSR Commission For UNEP pp.50, Mscow (1984).

- Lau, M.M. Wong; Tributyltin Antifoulings: A Threat to the Hong Kong Marine Environment, Arch. Environ. Contamn. Toxicol. 20:299-304 (1991).
- Le Blanc; G.A. Interspecies Relationships In Acute Toxicity of Chemicals To Aquatic Organisms. Environmental Toxicology and Chemistry: 3: 47-60 (1984).
- Petrilli, F.L.; and S.D. Flora. Metabolic Deactivation of Hexavalent Chromium Mutagenecity. Mutation Research 54:139-147 (1978).
- Putte, Van Der; M.A. Brinkhorst; and J.H. Koeman. Effect of PH on The Acute Toxicity of Hexavalent Chromium To Rainbow Trout (Salmo Gairdneri). Aquatic Toxicology, 1:129-142 (1981).
- Ruiz J.M.; G.W. Bryan and P.E. Gibbs Acute and Chronic Toxicity of Tributyltin (TBT) to pediveliger larvae of the bivalve Scrobicularia plana. Marine Biology: 124: 1 (Nov) (1995).
- Saliba, L.J.; V. Schembri and M.J. Vella. Lethal and Sublethal Effects of Mercury Salts On Monodonta Articulata. XXV1 Congress and Plenary Assembly of ICSEM Antalya, Turky, 24 November (1978).
- Shiber, J.G. Trace Metals in edible Crustaceans from Lebanon. Heavy Metals in Water- Organisms. Solamki J.ed. Vol. 29: 285-298 (1985).
- Stromgren T.; and T. Bongard. The Effect of Tributyltin Oxide on Growth of *Mytilus edulis*. Marine Pollution Bulletin 18(1): 30-31(1987).
- Thain, J.E. The acute toxicity of bis (tributyltin) oxide to the adults, and larvae of some marine organisms. CM 1983 E:13, International Council for the Expiration of the Sea, Copenhagen, Deumark (1983).

J.Pest Cont. & Environ. Sci. 9 (1) (2001).

- U.S. Environmental Protection Agency (EPA) Methods for acute toxicity tested with fish, macroinvertebrate and anpiibians. Ecological Research series (EPA-660 (3-75-009) (1975).
- Waldock, M.J., and J.E. Thain. Shell thickening in *Crassostrea gigas*. Organotin antifouling or sediment induced. Marine pollut. Bull 14(11): 411-415 (1983).
- Wong P.T.S.; and G.Dixon. Bioassessment of Water Quality. Environ. Toxicology and Water Quality. By John Wiley sons. Inc. 10: 9-17 (1995).
- Zaroogian, G.E., and S. Cheer. Cadmium accumulation by the American Oyster *Crassostrea virginica*. Nature 261: 408-410 (1976).

الملخص العربي

الممية الحزينية الحادة لاحد عشر من المعادن الثقيلة على نوعين من فصيلة الجاتدوفليات (القواقع البحرية) Tapes decussata and Venerupis aurea

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تم مقارنة السمية الحادة لاحد عشر من المعادن التقيلة من ألا ملاح الذائبة في المسله وتشمل كالوريد الالومنيوم - كلوريد الكادميوم -كلوريد كلوريد الكوبلت، وكبريتات النحاس ونترات رصاص، وقومفات المنجنيز (ثنائي القاعدية) وكلوريد الزئبفيك، نيسترات النيك وثنائي كرومات البوتاسيوم وكلوريد القصديروز - وكبريتات الزنك وذلك على الافراد البالغة من نوعين من الجندوفلي (القراقع البحرية) وهما Tapes decussata and Venerupis من نوعين من الجندوفلي (القراقع البحرية) وهما aurea

والمأخوذة من بحيرة التمساح بالإسماعيلية .

وقد تم تقدير التركيزات المتوسطة للموت بعد ٩٦ ساعة LC_{50} 's لهذه الامسلاح من المعادن الثقيلة الذائبة في الماء وكلها على صورة الكاتيونات المتأنية فيما عدا الكروم فهو موجود على صورة شق حامضي هو ثنائي الكرومات (Cr_2O_7)حيث التكافؤ المسادس لعنصر الكروم •

وقد تبين أن المقارنة الدقيقة للسمية الجزئية تقتضى حساب التركيزات الجزيئية لكل من الايونات التي تم اختبار سميتها •

وفي ضوء قيم التركيزات الجزيئية المتوسطة للموت لايونات المعادن الثقيلة - تبين أن كلا من الالومنيوم والزئبق والقصدير كانت من المجموعة شديدة السمية يليها المجموعــة المتوسطة السمية وتضم الكاديموم والرصاص والكروم - والمجموعة الأقــل فــي ســميتها تشمل على المنجنيز والنيكل والكوبات والزنك والنحاس •

ومن المأفت النظر ان المعادن الثقيلة الشديدة في سميتها الحادة تتميز أيضا بقدرتها على التراكم مما يؤدى الى حدوث سميتها المزمنة وما يترتب عليها من تدهور تكاثر القواقسع البحرية ذات الاهمية الاقتصادية فضلا عن تلويثها كمصدر لغذاء الانسان, وهذا يبين قيمسة السمية الحادة في التنبؤ بالسمية المزمنة واخطارها السمية الحادة في التنبؤ بالسمية المزمنة واخطارها المدينة المزمنة المزمنة والمعارها المدينة المزمنة المزمنة المزمنة المزمنة المزمنة المزمنة المزمنة المؤلمة المؤلمة