AACL BIOFLUX
Aquaculture, Aquarium, Conservation & Legislation
International Journal of the Bioflux Society

Physiological response of the intertidal copepod *Tigriopus japonicus* experimentally exposed to cadmium

Emadeldeen H. Mohammed, Guizhong Wang, Zhihuan Xu, Ziming Liu, and Lisheng Wu

College of Oceanography and Environmental Science, State Key Laboratory of Marine Environmental Science, Xiamen University, People's Republic of China.
Corresponding author: G. Wang, gzwang@xmu.edu.cn

Abstract. The intertidal harpacticoid copepod *Tigriopus japonicus* is a benthic copepod and has been commonly used in ecotoxicology and environmental genomics studies as a marine model species. In this study, Laboratory experiments were conducted to investigate the effects of cadmium (Cd) on survival, development, growth and reproductive performance of *T. japonicus*. Our results indicated that Cd was significantly affected adult survival and development, but not those of nauplii. Despite the reduction in adult female total body length, Cd was not significantly affected copepod growth. Concerning *T. japonicus* reproduction response, Cd was significantly reduced the number of nauplii produced at 10 µg L^{-1}. Thus, survival, development and reproduction in *T. japonicus* as a model test species could be effective physiological markers to monitor marine metal pollution and to assess population response.
Keywords: metals, cadmium, copepods, *Tigriopus japonicus*, ecotoxicology studies.

Introduction. Heavy metals are common environmental pollutants and their toxicity is a problem of increasing significance for ecological, evolutionary, nutritional and environmental reasons. They are considered hazardous to aquatic life because of their extreme persistence, high toxicity, tendency to bioaccumulate, and because they are available through many and diverse anthropogenic sources (Fairbrother et al 2007; Nicula et al 2010; Rahman et al 2010). Thus, environmental monitoring is essential to restore and resolve metal pollution. Major emissions of Cd to aquatic environment are atmospheric fallout and effluents from smelting, metal mining, refining industries, sewage, and runoff from agricultural fertilizers (WHO 1992; CCME 1999; Jarup 2003). In the marine environment, river runoff is another major pathway of Cd (CCME 1999). Although Cd occurs in very low concentrations in open ocean waters, however, its concentrations in coastal and estuarine waters are higher (Chiffoleau et al 2001).

There are several copepod species used in marine ecotoxicity testing of chemical pollution as a sensitive test species and in adopting standard protocols for ecotoxicity testing, such as *Acartia tonsa*, *Nitocra spinipes*, *Tisbe battagliai*, and *Amphiascus tenuiremi*s (Andersen et al 1999, 2001; Breitholtz & Wollenberger 2003; ASTM 2004; OECD 2006; Templeton et al 2006; Kusk & Wollenberger 2007; Raisuddin et al 2007). There is an increase in the knowledge base of the copepods genus *Tigriopus*, particularly in the areas of their ecology, geophylogeny, genomics, and their behavioural, biochemical, and molecular responses following exposure to environmental stressors and chemicals (Raisuddin et al 2007). The intertidal harpacticoid copepod *Tigriopus japonicus* has a wide geographical distribution along the coast in the Western Pacific including Japan, South Korea, China, Taiwan, and Hong Kong. It has been commonly used in ecotoxicology and environmental genomics studies as a marine model species (Raisuddin et al 2007). *T. japonicus* has several advantages as a model test species. Hence, its distinct developmental stages, dimorphic sexes, high fecundity, short life-cycle, and easy to culture permit detailed ecotoxicological studies on the effects of toxicants (Raisuddin et al 2007). Moreover, *Tigriopus* spp. have shown sensitivity towards exposure to

chemical pollutants, however, metals and organic pollutants have shown reproducible biological responses when tested using *Tigriopus* spp. in ecological risk assessment associated with water pollution (Lee et al 2007, 2008; Raisuddin et al 2007; Kwok et al 2008, 2009).

In order to develop the copepod *T. japonicus* as a model species, information on its sensitivity to different chemical pollutants is, therefore, highly desirable. Moreover, laboratory studies addressing acute and chronic effects of heavy metals on physiological response of copepods, particularly life-cycle and reproductive tests, are considered important to understand how copepod populations cope with metal pollution and to conclude a holistic risk assessment of metal pollution. However, recent studies indicated that Cd pollution in Xiamen waters and Jiulongjiang estuary, where *T. japonicus* was collected, is the most hazardous with respect to other metals. Hence, Jian-qing et al (2007) reported dissolved and particulate Cd concentrations in Jiulongjiang estuary of 0.34 and 0.0064 μg L^{-1}, respectively. They also estimated the average flux of dissolved and particulate Cd from Jiulong river to Jiulongjiang estuary of about 1.47 and 0.027 kg d^{-1}, respectively. Weili et al (2009) found that Cd content in the surface sediments of Jiulongjiang river estuary is beyond marine quality standard in some stations. Thus, the present study primarily aimed to: (1) determine the 48h-LC50 value of Cd to the copepod *T. japonicus* (2) assess the sublethal toxicity of Cd in a full life-cycle toxicity test on survival, development, and growth of *T. japonicus*. (3) assess the sublethal toxicity of Cd on offspring production of *T. japonicus*.

Materials and Methods. The copepod *T. japonicus* was obtained from continuous stock culture in our laboratory and maintained under static-renewal conditions in 0.45μm millipore filtered seawater with 7 to 7.9 mg L^{-1} dissolved oxygen, 7.90 to 8.25 pH, 18 to 22° C and 24 to 26 ppt salinity. The original stock culture of *T. japonicus* was obtained from rocky intertidal zone pools in Xiamen Bay, China. Copepods were fed a mixed algal diet of *Isochrysis galbana* and *Platymonas subcordiformis*. The algae were cultured in filtered seawater contain f/2 enriched media at 20° C.

Cadmium was provided as a chloride salt ($CdCl_2 \cdot 2.5H_2O$) from Sinopharm Chemical Reagent Co., Ltd, China (≥ 99.0% pure). The stock solutions were prepared (at concentrations high enough to prevent weighing errors (= 1000 mg L^{-1}) in double distilled water. Selected test Cd concentrations were prepared by dilution of the stock solution in 0.45 μm filtered seawater. Reference seawater used in the experiments was obtained 20 km offshore in Xiamen Bay. All Cd concentrations reported in this study were nominal concentrations.

Acute toxicity tests. Semi-static 48-h acute toxicity tests were performed using standard methods outlined by Verriopoulos & Dimas (1988) and ASTM (2004) with certain modifications. Experimental conditions were the same as copepod maintenance, except for temperature and salinity, which were 20° C and 25 ppt salinity, respectively. Only active adult copepods (females) were subjected to the acute tests. Copepods were exposed to different Cd concentrations in a semi-static exposure system bioassays, during which the test solutions were replaced every 24 h. Six toxicant Cd concentrations as nominal concentrations and control were used. Each treatment was run in triplicate, with 10 active individuals for each replicate. Acute tests of *T. japonicus* were done in six well culture plates, each with 10 mL of solution and five female copepods. Culture plates were provided from Corning Incorporated, USA. Animals were monitored and dead animals removed at 24 and 48 h. The criteria for mortality are total lack of movement and lack of response after repeat touches with a probe during two minutes. Because of the shorter time, copepods were not fed during the test period.

Life-cycle toxicity tests. Complete life-cycle toxicity tests of Cd to the copepod *T. japonicus* were conducted using a modified ASTM protocol (ASTM 2004). Nauplii (<24h after hatching) were obtained by sorting females with egg sac and allocating them to six-well tissue culture plates (Corning, USA) with seawater from the stock culture. After 24 h, females were taken back to the stock culture and 20 newly-hatched nauplii (<24h old) per test concentration were randomly transferred to six-well tissue culture plates, with 10 mL working volume and 10 nauplii in three replicates (total 60 nauplii). Experimental

conditions were the same as acute toxicity test. Cd test concentrations used were 0, 1, 10, 100 μg L^{-1}. Experiments were carried out as renewal bioassay, with daily renewal of half of the exposure solution. After the daily renewal of half of the test solutions, copepods were fed with *Isochrysis galbana* at a density of 5×10^5 cells mL^{-1} during whole life-cycle exposure. Survival and development were monitored everyday until animals reached females with egg sac. Observations of copepods were performed under a stereomicroscope (Motic, series SMZ140). Nauplii survival rate calculated as percentage of surviving nauplii which reached copepodid stage on day 8. Adult survival rate calculated as percentage of surviving copepod (male and female) on day 21. Mean developmental time defined as the time (days) required for development of 50% of individuals into a specific stage (Vidal 1980) and was estimated from the graphical representation (regression analysis) of cumulative percentage of the specific stage against time. Nauplii developmental time (the duration of nauplii stage) was expressed as the time taken for 50% of nauplii (<24h old) to reach the first copepodid stage. Time to first reproduction (the duration to reach adult female stage with egg sac) was expressed as the time taken for 50% of nauplii (<24h old) to reach the adult female stage with egg sac. Individuals that became males were considered in the calculations of adult survival rate, but not in adult growth and developmental time. At the end of the tests females were preserved in 5% formalin to measure their growth as total body length. Total body length measurements of 15 individuals per treatment were obtained on a microscope (Olympus BX51, Japan) slide using Image-pro Express software version 6.0.0.319, Media Cybernetics Inc.

Reproductive toxicity test. Reproductive tests were conducted using a modified ASTM protocol (ASTM 2004). Experimental conditions and Cd concentrations used were the same as life-cycle toxicity tests. Copepods were fed daily with 5×10^5 cells/mL of the algae *Isochrysis galbana*. For each test concentration, 20 females of *T. japonicus*, bearing an egg sac, were individually transferred to a new six well culture plates (Corning, USA), each with 10 mL of solution. These females were cultured for 10 days. The fecundity (offspring production) was assessed as the number of nauplii per female. Every day females were transferred to a new culture plate with fresh solutions and the resulting nauplii were counted under a stereomicroscope (Motic, series SMZ140).

Two statistical programs (Microsoft excel 2003 package; SPSS 17, Chicago, IL, USA) were used to analyze the data.LC50 value was computed using probit analysis (Finney 1971). The data were expressed as mean ± standard deviation (S.D.). Percentages for survival rate were arcsine-transformed prior to statistical analysis. Statistical analysis for survival rate, developmental time, body length, and offspring production comparisons was carried out using one-way ANOVA and the Fisher least significant difference test to evaluate whether the means were significantly different among cadmium treatments. The level of significance was accepted as $p < 0.05$.

Results

Acute toxicity tests. There was no control mortality during the experimental period. The probit analysis of acute toxicity data showed that 48h-LC50 value of Cd for *T. japonicus* was 12.1 mg L^{-1} (Fig. 1).

Life-cycle toxicity tests. Despite nauplii survival decreased with time extended and Cd concentration increased, there was no significant difference between the control group and the treated groups (Fig. 2, $p < 0.05$). *T. japonicus* adult survival was decreased with increasing Cd concentrations and was significantly ($p < 0.05$) affected at 10 μg L^{-1} Cd, however, there was no significant difference between adult survival rate at Cd concentrations 1, 10 and 100 μg L^{-1} Cd (Fig. 2, $p < 0.05$). Compare to control treatment, nauplii developmental time (days) increased when exposure to other Cd treatments, but the difference between control and all treated groups was insignificant (Fig. 3, $p < 0.05$). Thus, there was no significant delay in nauplii developmental time when exposure to the tested Cd concentrations. However, time to first reproduction (days) increased when exposure to Cd treatments, and it was significantly different from control at 100 μg L^{-1} Cd (Fig. 3, $p < 0.05$). But, there was no significant difference between time to first reproduction (days) at Cd concentrations 1, 10 and 100 μg L^{-1} Cd (Fig. 3, $p < 0.05$).

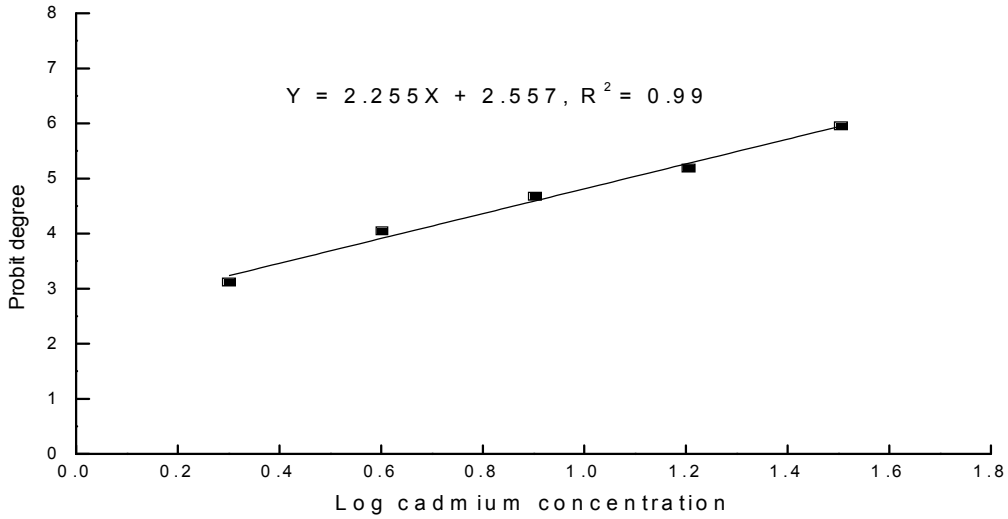

Fig. 1. 48h-LC50 value of Cd for *T. japonicus* on probit scale was calculated as the return logarithmic value of x, when y = 5.

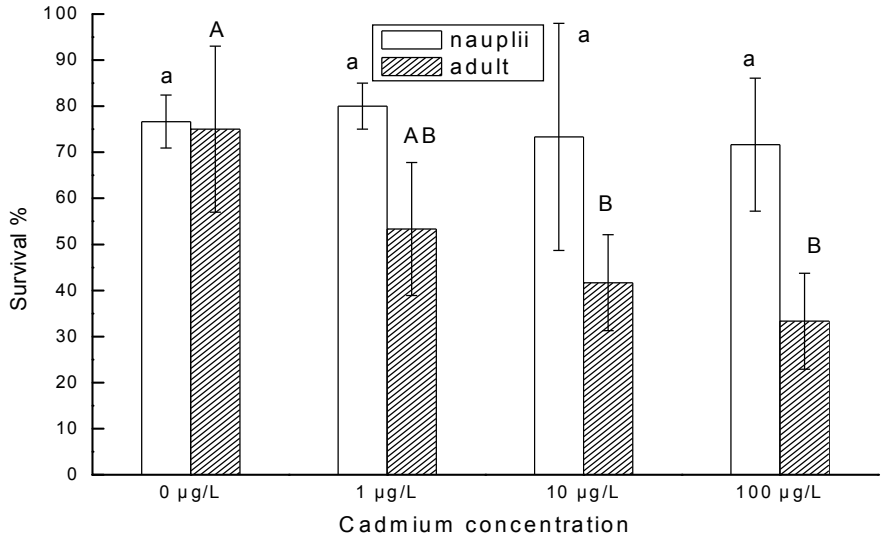

Fig. 2. Effects of cadmim on nauplii and adult survival of *T. japonicus*. Data are described as mean ± standard deviation. Different letters indicate a significant difference among different Cd concentrations at $p < 0.05$.

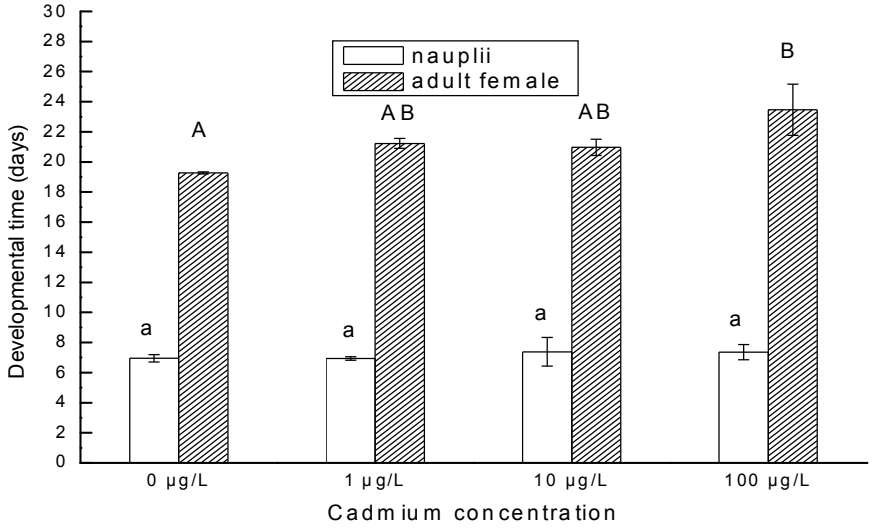

Fig. 3. Effects of cadmium on nauplii and adult developmental time of *T. japonicus*. Data are described as mean ± standard deviation. Different letters indicate a significant difference among different Cd concentrations at $p < 0.05$.

Figure 4 shows the effect of Cd on the growth of *T. japonicus* adult female, measured as total body length (μm). There was no statistically significant difference between control treatment and all Cd treated groups 1, 10 and 100 μg L^{-1} Cd (Fig. 4, $p< 0.05$). However, female total body length decreased when exposure to Cd treated groups.

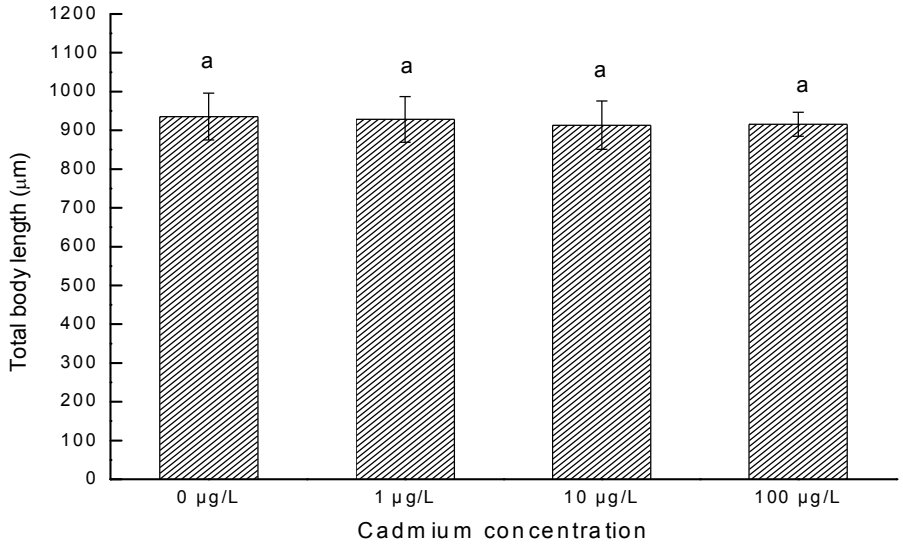

Fig. 4. Effects of cadmium on adult female body length of *T. japonicus*. Data are described as mean ± standard deviation. Different letters indicate a significant difference among different Cd concentrations at $p< 0.05$.

Reproductive toxicity test. During the reproductive toxicity tests of *T. japonicus*, there was no mortality in all treatments. In summary, a 10 day exposure of *T. japonicus* to Cd resulted in a significant reduction of its reproductive output. Offspring production (i.e. number of nauplii produced) of *T. japonicus* decreased with increasing Cd concentrations and was significantly ($p< 0.05$) reduced by 24.25% at 10 μg L^{-1} Cd (Fig. 5).

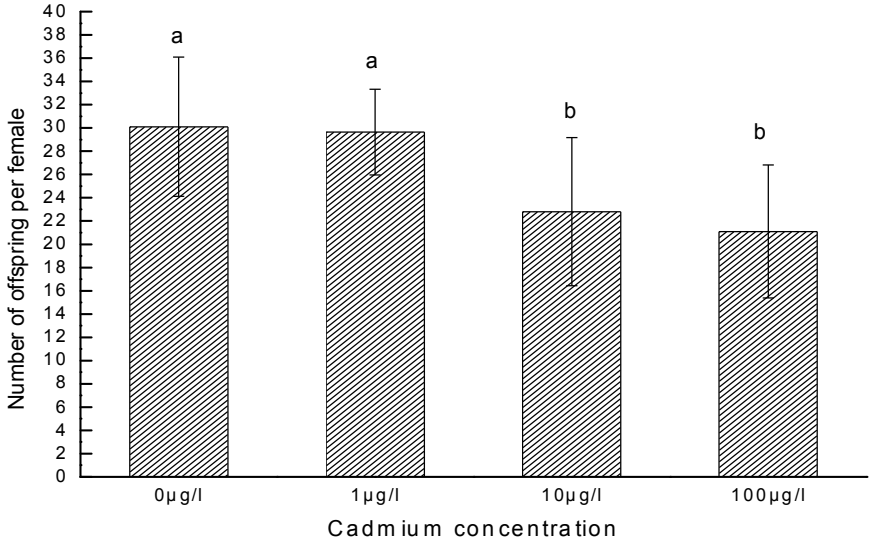

Fig. 5. Effect of Cd on offspring production of *T. japonicus*. Data are described as mean ± standard deviation. Different letters indicate a significant difference among different Cd concentrations at $p< 0.05$.

Discussion. Previous studies have clearly demonstrated the potential deleterious effects of metal pollutants on copepod biology (Raisuddin et al 2007; Kwok et al 2008, 2009). Generally, the results of this study indicated that Cd was negatively affected the survival, development, growth, and reproduction of harpacticoid copepod *T. japonicus*.

48h-LC50 value of Cd for *T. japonicus* was 12.1 mg L^{-1} (Fig. 1). A study by Lee et al (2007) reported a 96-h LC50 value of 25.2 mg L^{-1} Cd for the same species. The difference may be due to different experimental conditions, especially salinity. The experimental salinity of this study is 25 ppt while theirs is 32 ppt. However, there was a significant decrease in LC50 values as salinity decrease and was most likely related to increase in cadmium free ion (Hall Jr et al 1995). Previously we found a 48-h LC50 value of 17.7 mg L^{-1} Ni for *T. japonicus* (Mohammed et al 2010). Thus, Cd was more acutely toxic to *T. japonicus* than Ni. Verriopoulos & Dimas (1988) reported a 48-h LC50 value of 0.9 mg L^{-1} Cd for the copepod *Tisbe holothuriae*. Thus, *T. holothuriae* was more sensitive to Cd than *T. japonicus* (12.1 mg L^{-1}). Forget et al (1998) found a 96-h LC50 value of 47.9 µg L^{-1} Cd for the copepod *Tigriopus brevicornis*. Exact comparisons are difficult because of difference in LC50 time interval.

Exposure to Cd was not significantly affected nauplii survival rate at Cd concentration tested, but in contrast it was significantly ($p< 0.05$) affected adult survival rate at 10 µg L^{-1} Cd (Fig. 2, $p< 0.05$). Conversely, Lee et al (2008) found that exposure to 0.1, 1, 10, 100 µg L^{-1} of Cu, As^{3+} and As^{5+} using *T. japonicus* in two generation life-cycle toxicity test were not significantly affected the adult survival (to reach maturity). The difference may be due to different experimental conditions, especially salinity. The experimental salinity of this study is 25 ppt while theirs is 32 ppt. Considering other copepod species, Medina et al (2008) observed that Cu was significantly affected the juvenile survival of the copepod *Tigriopus angulatus*.

Like all copepods, *T. japonicus* undergoes anamorphic development with 12 distinctive post-embryonic developmental stages, six naupliar stages, five copepodid stages, and an adult stage. Its distinct developmental stages permit detailed ecotoxicological studies on the effects of toxicants on their development (Raisuddin et al 2007). In this study the effect of Cd on *T. japonicus* development was evaluated. Although nauplii developmental time increased with the increasing Cd concentrations, it was not significantly affected when nauplii exposed to all Cd concentrations tested (Fig. 3, $p< 0.05$). However, Cd was significantly affected time to first reproduction at the highest Cd concentration tested, 100 µg L^{-1} (Fig. 3, $p< 0.05$). Thus, time to first reproduction of *T. japonicus* was more sensitive to Cd than nauplii developmental time. In agreement with our results, previous studies showed that metals were negatively affected copepod developmental time. For instance, Toudal & Riisgard (1987) observed that developmental time of copepodite and adult stages of the copepod *Acartia tonsa* was highly influence by Cd. Correspondingly, it was reported that Cu caused a prolongation of maturation time of the marine copepods *Tisbe holothuriae* (Moraïtou-Apostolopoulou et al 1983) and *T. anagulatus* (Medina et al 2008). Regarding *T. japonicus*, it was reported that time to develop into copepodid stage 1 (Kwok et al 2008, 2009) and time to first reproduction (Kwok et al 2008) slowed down significantly at 10 µg L^{-1} Cu in a full life-cyle test. Also, Lee et al (2008) found that exposure of *T. japonicus* to Cu, As^{3+} and As^{5+} were significantly increased the duration of nauplii phase at 1, 0.1, 0.1 µg L^{-1}, respectively, but not the generation time for adult.

Most copepods of the genus *Tigriopus spp*. are small in size, adult generally about 1.0 mm in length (Raisuddin et al 2007). Here we measured total body length of *T. japonicus* adult female in response to Cd. Despite the reduction in its total body length when exposure to Cd, copepod growth measured as total length was not significantly affected at all Cd concentrations tested, (Fig. 4 $p< 0.05$). Previously, Toudal & Riisgard (1987) found that the cephalothoracic length of adult copepod was significantly reduced at 40 µg L^{-1} Cd in life-cycle toxicity test with *A. tonsa*. Thus, the growth of *A. tonsa* was more sensitive to Cd than *T. japonicus*.

Our results indicate that Cd was significantly ($p< 0.05$) reduced offspring production from *T. japonicus* at 10 µg L^{-1} (Fig. 5). These results are consistent with previous findings that copepod offspring production is significantly impacted when

exposed to metals. For instance, Medina et al (2008) found that nauplii production of *T. angulatus* was significantly different from the control at Cu exposures to 103 and 180 µg L^{-1}. Regarding *T. japonicus*, it was reported that Cu and Ni were significantly reduced its offspring production at 10 µg L^{-1} (Kwok et al 2009; Mohammed et al 2010). Conversely, Lee et al (2008) found that Cu, As^{3+}, and As^{5+}, had no significant effects on offspring production of *T. japonicus* after 10 days exposure at the concentrations tested (0.1, 1, 10, and 100 µg L^{-1}). With the exception of salinity, they used natural seawater and the same experimental condition. Taken together, *T. japonicus* was more sensitive to Cd than Cu, As^{3+} and As^{5+}.

Conclusion. The present study showed that Cd was negatively affected the survival, development, growth and reproduction of harpacticoid copepod *T. japonicus*. Specifically, Cd was significantly affected adult survival and development at 10 and 100 µg L^{-1}, respectively. Moreover Cd was significantly reduced its offspring production at 10 µg L^{-1}. Consequently, Cd can have detrimental effects on copepod population dynamics. As a next step to understand the mechanistic basis, it is necessary to evaluate the potential impacts Cd on the biochemical and molecular components of different copepod stages in long-term toxicity test. Furthermore, the synergistic effects of Cd and other environmental factors, such as temperature, salinity, and pH should be investigated.

Acknowledgments. This work was funded by National Natural Science Foundation of China (No. 40876060). We thank the colleagues of our lab for assistance in the laboratory.

References

Andersen H. R., Halling-Sørensen B., Kusk K. O., 1999 A parameter for detecting estrogenic exposure in the copepod *Acartia tonsa*. Ecotoxicology and Environmental Safety **44**:56–61.

Andersen H. R., Wollenberger L., Halling-Sørensen B., Kusk K. O., 2001 Development of copepod nauplii to copepodites – a parameter for chronic toxicity including endocrine disruption. Environmental Toxicology and Chemistry **20**:2821–2829.

ASTM, 2004 Standard guide for conducting renewal microplate-based life-cycle toxicity tests with a marine meiobenthic copepod. American Society for Testing and Materials, Philadelphia, PA, ASTM Standard No. E2317-04, pp. 1–16.

Breitholtz M., Wollenberger L., 2003 Effects of three PBDEs on development, reproduction and population growth rate of the harpacticoid copepod *Nitocra spinipes*. Aquatic Toxicology **64**:85–96.

CCME, Canadian Council of Ministers of the Environment, 1999 Canadian Water Quality Guidelines for the Protection of Aquatic Life: Cadmium. In Canadian environmental quality guidelines. Winnipeg.

Chiffoleau J.-F., Auger D., Chartier E., Michel P., Truquet I., Ficht A., Gonzalez J.-L., Romaña L.-A., 2001 Spatiotemporal changes in cadmium contamination in the Seine estuary (France). Estuaries **24**:1029–1040.

Fairbrother A., Wenstel R., Sappington K., Wood W., 2007 Framework for metals risk assessment. Ecotoxicology and Environmental Safety **68**:145–227.

Finney J., 1971 Probit analysis. 2nd ed. Cambridge, UK. Cambridge University Press.

Forget J., Pavillon J. F., Menasria M. R., Bocquene G., 1998 Mortality and LC50 values for several stages of the marine copepod *Tigriopus brevicornis* (Muller) exposed to the metals arsenic and cadmium and the pesticides atrazine, carbofuran, dichlorvos, and malathion. Ecotoxicology and Environmental Safety **40**:239–244.

Hall L. W., Jr., Ziegenfuss M. C., Anderson R. D., Lewis B. L., 1995 The effect of salinity on the acute toxicity of total and free cadmium to a Chesapeake Bay copepod and fish. Marine Pollution Bulletin **30**(6):376–384.

Jarup L., 2003 Hazards of heavy metal contamination. British Medical Bulletin **68**:167–182.

Jian-qing G., Bi-hua Q., Ai-cui D., Hong-ru Z., Deng-hui C., Jiong-hui S., Yun W., Shui-ying H., An-xiang Q., Xiao-yan L., Can-rong Q., Ming-gang C., 2007 [Elementary

study on the geochemical behaviour of Cu, Pb, Cd in the Jiulong estuary, Xiamen]. Journal of Xiamen University (Natural Science) **46**:54-61. [In Chinese]
Kusk K. O., Wollenberger L., 2007 Towards an internationally harmonized test method for reproductive and developmental effects of endocrine disrupters in marine copepods. Ecotoxicology **16**:183–195.
Kwok K. W. H., Grist E. P. M., Leung K. M. Y., 2009 Acclimation effect and fitness cost of copper resistance in the marine copepod *Tigriopus japonicus*. Ecotoxicology and Environmental Safety **72**:358–364.
Kwok K. W. H., Leung K. M. Y., Bao V. W. W., Lee J.-S., 2008 Copper toxicity in the marine copepod *Tigropus japonicus*: Low variability and high reproducibility of repeated acute and life-cycle tests. Marine Pollution Bulletin **57**:632–636.
Lee K.-W., Raisuddin S., Hwang D. S., Park H. G., Lee J.-S., 2007 Acute toxicities of trace metals and common xenobiotics to the marine copepod *Tigriopus japonicus*: evaluation of its use as a benchmark species for routine ecotoxicity tests in Western Pacific Coastal Regions. Environmental Toxicology **22**:532–538.
Lee K.-W., Raisuddin S., Hwang D. S., Park H. G., Dahms H. U., Ahn I. Y., Lee J.-S., 2008 Two-generation toxicity study on the copepod model species *Tigriopus japonicus*. Chemosphere **72**:1359–1365.
Medina M. H., Morandi B., Correa J. A., 2008 Copper effects in the copepod *Tigriopus angulatus* Lang, 1933: natural broad tolerance allows maintenance of food webs in copper-enriched coastal areas. Marine and Freshwater Research **59**:1061–1066.
Mohammed E. H., Wang G., Jiang J., 2010 The effects of nickel on the reproductive ability of three different marine copepods. Ecotoxicology **19**:911–916.
Moraïtou-Apostolopoulou M., Kiortsis M., Verriopoulos V., Platanistioti S., 1983 Effects of copper sulphate on *Tisbe holothuriae* Humes (Copepoda) and development of tolerance to copper. Hydrobiologia **99**:145–150.
Nicula M., Bănăţean-Dunea I., Gergen I., Hărmănescu M., Simiz E., Pătruică S., Polen T., Marcu A., Lunca M., Szucs S., 2010 Effect of natural zeolite on reducing tissue bioaccumulation and cadmium antagonism related to some mineral micro- and macronutrients in Prussian carp (*Carassius gibelio*). AACL Bioflux **3**(3):171-179.
OECD, 2006 Detail review paper on aquatic arthropods in life cycle toxicity tests with an emphasis on developmental, reproductive and endocrine disruptive effects. OECD series on testing and assessment number 55, ENV/JM/MONO(2006)22, Environment Directorate, Organisation of Economic Corporation and Development, Paris, p. 125.
Rahman M. M., Rahman M. M., Chongling Y., Islam K. S., 2010 Changes in growth and antioxidant enzymes activities during cadmium stress in the mangrove plant *Kandelia candel* (L.) Druce. AES Bioflux **2**(1):15-24.
Raisuddin S., Kwok K. W. H., Leung K. M. Y., Schlenk D., Lee J.-S., 2007 The copepod *Tigriopus*: A promising marine model organism for ecotoxicology and environmental genomics. Aquatic Toxicology **83**:161–173.
Templeton R. C., Ferguson P. L., Washburn K. M., Scrivens W. A., Chandler G. T., 2006 Life-cycle effects of single-walled carbon nanotubes (SWNTs) on an estuarine meiobenthic copepod. Environmental Science and Technology **40**:7387–7393.
Toudal K., Riisgard H. U., 1987 Acute and sublethal effects of cadmium on ingestion, egg production and life-cycle development in the copepod *Acartia tonsa*. Marine Ecology **37**:141–146.
Verriopoulos G., Dimas S., 1988 Combined toxicity of copper, cadmium, zinc, lead, nickel, and chrome to the copepod *Tisbe holothuriae*. Bull Environ Contam Toxicol **41**:378–384.
Vidal J., 1980 Physioecology of zooplankton. II. Effects of phytoplankton concentration, temperature, and body size on the development and molting rates of *Calanus pacificus* and *Pseudocalanus sp*. Marine Biology **56**:135–146.
Weili W., Anchao G., Huatai L., Aiguo G., 2009 [Distribution and potential ecological risk evaluation of heavy metals in the surface sediments from the Juilongjiang river estuary]. Advances in Marine Science **27**:502-508. [In Chinese]
WHO, 1992 Cadmium-environmental aspects: Environmental Health Criteria 135. World Health Organization, Geneva.

Received: 22 December 2010. Accepted: 22 January 2011. Published online: 24 January 2011.
Authors:
Emadeldeen Hassan Mohammed, Department of Marine Biology, College of Oceanography and Environmental Science, Xiamen University, Xiamen, Postal Code 361005, Siming Area, Da Xue Street, People's Republic of China, Tel: +86-592-2188471, e–mail: emadeldeenelfahal@yahoo.com
Guizhong Wang, Professor of marine biology, Department of Marine Biology, College of Oceanography and Environmental Science, Xiamen University, Xiamen, Postal Code 361005, Siming Area, Da Xue Street, People's Republic of China, Tel: +86-0592-2188471, Fax: +86 0592-2186397, e-mail: gzwang@xmu.edu.cn
Zhihuan Xu, Department of Marine Biology, College of Oceanography and Environmental Science, Xiamen University, Xiamen, Postal Code 361005, Siming Area, Da Xue Street, People's Republic of China, Tel: +86-592-2188471, e-mail: zhihuanxu@gmail.com
Ziming Liu, Department of Marine Biology, College of Oceanography and Environmental Science, Xiamen University, Xiamen, Postal Code 361005, Siming Area, Da Xue Street, People's Republic of China, Tel: +86-592-2188471, e-mail: lziming1986@163.com
Lisheng Wu, Department of Marine Biology, College of Oceanography and Environmental Science, Xiamen University, Xiamen, Postal Code 361005, Siming Area, Da Xue Street, People's Republic of China, Tel: +86-592-2188471, e-mail: lisheng@xmu.edu.cn
How to cite this article:
Mohammed E. H., Wang G., Xu Z., Liu Z., Wu L., 2011 Physiological response of the intertidal copepod *Tigriopus japonicus* experimentally exposed to cadmium. AACL Bioflux **4**(1):99-107.