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EFFECTS OF DIETARY PBDE-47 EXPOSURE ON THE MARINE GASTROPOD CREPIDULA ONYX

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Introduction

Polybrominated diphenyl ethers (PBDEs) are widely used brominated flame-retardants and are persistent in the environments. It has become ubiquitous in the ocean and have accumulated in marine sediment [1] and biota [2-4] to very high levels. Concerns have thus been raised on the biological effects of PBDEs on animals. PBDEs have been found to disrupt endocrine system and affect neurobehavioural development [5,6], they are also genotoxic and teratogenic [3,7] in mammals and fishes. In comparison, very little is known concerning the accumulation and effects of PBDEs on marine invertebrates. Except in female mussels which ovarian follicles and oocytes were damaged [8], and in Daphnia magna which molting was inhibited [9].

Dietary exposure, or trophic transfer, is known to be a major transport route of organic pollutants from organisms at lower trophic level to their consumers. Compared with passive uptake, POPs are delivered more effectively to higher trophic level via assimilation [10]. It is believed that plankton at the lower trophic level play a major role as in transferring organic pollutants into the marine food web [11], because of their high ability to uptake organic pollutants from water [12,13]. Moreover, studies have evident the bioavailability of PBDEs to offspring through parental transfer in marine organisms, leading to exposure of offspring during early developmental stages [14-16]. This early exposure may have a significant impact on the fitness and survival of the organism. Therefore, it is interesting to find out whether dietary exposure of PBDEs with microalgae as vehicle would have negative impacts on marine invertebrate, and whether parental exposure could affect offspring of later generations.

Materials and methods

The effects of long-term dietary exposure to environmental level of BDE-47 on marine invertebrate were systematically studied using Crepidula onyx (Gastropod, Calyptraidae). BDE-47 had been encapsulated into microalgae (Isochrysis galbana) prior to feeding to the C. onyx. The impacts on Darwinian fitness traits (i.e. growth, development, and reproduction) of maturing C. onyx and their offspring were determined by measurement of shell length, settlement assays and observation of embryonic development. Transcriptomic analysis by RNA sequencing of exposed C. onyx was conducted to unravel mechanisms underlying the observed phenotypic changes.

The body burden of PBDEs, including a few common PBDE analogs (BDE-28, methoxylated- and hydroxylated- PBDEs), were measured using the protocol of chemical analysis described in Wang et al. [17] with modifications. In brief, lipids in samples were extracted by chloroform/methanol (1:1) with sonication. After phase separation, the neutral and the phenolic fractions were cleaned up by columns with acidified silica gel and fluorosil, respectively. Target analysts of PBDEs were quantified by gas chromatography interfaced to a mass spectrometry equipped with VF-5MS capillary column.

Results and discussion

Bioaccumulation was evident from the measured body burden of BDE-47 in C. onyx from larval stage to adult after chronic dietary exposure. Although BDE-28 was formed in I. galbana, the assimilation rate of that into C. onyx was insignificant as none of the PBDE analogs were detected in the gastropods. Reduction of juvenile survivorship and delay of time to emerge as male in both treatment groups showed physiological impairments of C. onyx upon BDE-47 exposure despite that shell growth and larval settlement percentage were unaffected. Further, reproductive impairment of F0 adults was clearly evident by the reduction in number of oviposition per female and proportion of well-developed F1 larvae. The maternally transferred BDE-47 in F1 offspring had embryotoxic effect as demonstrated by the higher number of broods with non-viable F1 embryos which failed to develop normally through early cleavage stages (see Figure 1). Lastly, the transcriptomic analysis provided potential mechanistic explanations on some of the phenotypic changes observed above. Namely, the malformed velar lobes might be cause be dysregulation in genes related to cilia; the delay in male emergence might be the result of dysregulation of steroid hormones; failure of embryos to perform cleavage at early stage could be due to the suppression of RHO protein, and potentially related to the suppression of Wnt signaling pathway.

Finding from the present study has highlighted the need for long-term exposure study of toxicants because the parental transfer effect of pollutants may cause serious and subtle impact to the offspring at their early stage.

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References

1 Mai, B. X., Chen, S. J., Luo, X. J., Chen, L. G., Yang, Q. S., and Sheng, G. Y. D. (2005) Environ Sci Technol 39, 3521-3527.
2 Wang, Y., Jiang, G., Lam, P. K. S., and Li, A. (2007) Environ Int 33, 963-973.
3 Lee, H., and Kim, G. (2015) Ocean Sci J 50, 119-142.
4 Law, R. J., Allchin, C. R., de Boer, J., Covaci, A., Herzke, D., Lepom, P., Morris, S., Tronczynski, J., and de Wit, C. A. (2006) Chemosphere 64, 187-208.
5 McDonald, T. A. (2002) Chemosphere 46, 745-755.
6 Legler, J. (2008) Chemosphere 73, 216-222.
7 Yu, L., Han, Z., and Liu, C. (2015) Gen Comp Endocr 219, 64-73.
8 Aarab, N., Lemaire-Gony, S., Unruh, E., Hansen, P. D., Larsen, B. K., Andersen, O. K., and Narbonne, J. F. (2006) Aquat Toxicol 78, S86-S92.
9 Davies, R., and Zou, E. (2012) Ecotoxicology 21, 1371-1380.

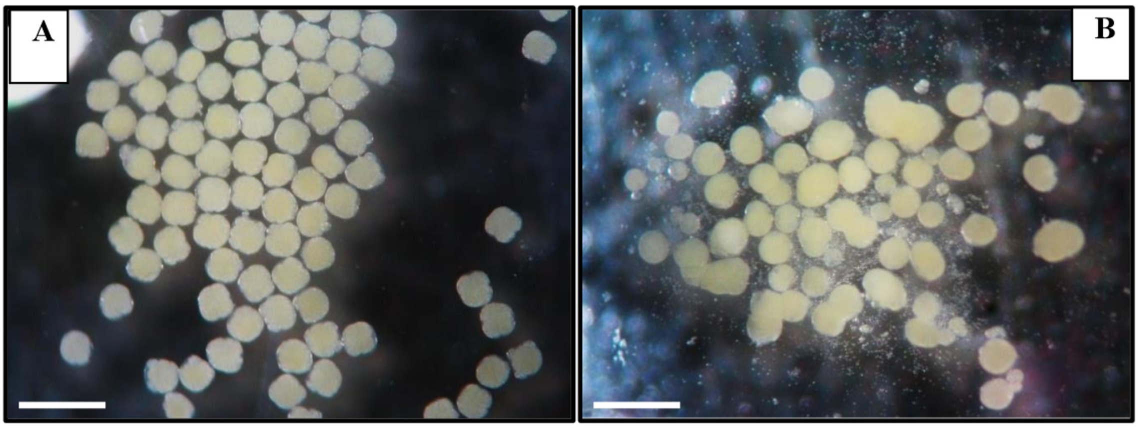

Figure 1. Some broods of F1 *Crepidula onyx* embryos developed abnormally during blastula to gastrula stage and became non-viable. (A) Normal embryos at blastula stage and (B) non-viable embryos with yolky materials of irregular shapes and sizes. Bar represents 0.5 mm.