Genotoxic Effects of PCBs and Heavy Metals on Marine Mussels

Results from in vitro experiment

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INTRODUCTION

- Rapidly increased industrialization causing serious environmental pollutions

- Organic pollutants, such as Polychlorinated biphenyls (PCBs) widely used in commercial products and industrial processes, can be toxic and persist in the environment

- Inorganic contaminants, such as heavy metals, at higher concentration are toxic

- Bioaccumulation of various pollutants through the food web can cause adverse effects to human health and environment
PCBs
HOW DO PCBs GET TO AQUATIC ENVIRONMENT?
Major Concerns about PCBs:

- Rapidly accumulating in humans and animals
- Hormonal disruption
  - Effects on thyroid, estrogen and testosterone
- Developmental effects
  - Irreversible learning/behavioral effects in young animals
  - Decreased ovarian follicles, sperm counts
  - Recent Scandinavian study – maternal PCBs associated with genital birth defect in baby boys
- Cancer
  - Structures similar to known carcinogens (PBDEs, PBBs)
  - Environmental conversion to known carcinogens (dioxins and furans)
EXAMPLES OF BIOINDICATORS AQUATIC ORGANISMS

BIOMARKERS:
- fish
- crustaceans
- mollusks
- plankton

- Strongly accumulate both organic and inorganic pollutants
- Organism biochemical and other biological processes
- Wide geographical distribution, their abundance
- Sessile life style feeding mechanism
- Accessibility (Ease to collect)

PICES 2015 Annual Meeting Change and Sustainability of the North Pacific 2015-10-15
RESEARCH QUESTIONS

- Can PCBs, Cu and Zn induce adverse effect at cellular level in vitro?
- Can DNA strand breaks detected by COMET assay efficiently assess the stress in mussel haemolymph after exposure?
- Can in vitro COMET assay be used as a routine potential tool for ecotoxicology testing and pollution monitoring of PCBs and heavy metals together in marine organisms?

AIM OF STUDY

This study is aimed to apply marine mussels as model to investigate the genotoxic effects after exposed to PCBs, Cu and Zn concentrations either singly or in a combination.
**MATERIALS AND METHODS**

1. **Extraction of haemolymph from mussels**
2. **Exposure**
3. **Gel Immobilization**
4. **Lysing**
5. **Alkali unwinding and Electrophoresis**
6. **Neutralization**
7. **DNA staining and COMET visualization**
8. **Scoring**
RESULTS AND DISCUSSIONS

Cell viability and Tail DNA % with different concentrations of Hydrogen peroxide exposure.

- CTRL
- 10 μM
- 100 μM
- 500 μM
- 1000 μM

Hydrogen peroxide exposure
RESULTS AND DISCUSSIONS-PCBs exposure

![Bar chart showing TAIL DNA % against PCB concentration (µg/ml).]
RESULTS AND DISCUSSIONS-Zn exposure

![Graph showing tail DNA percentage for different concentrations of Zn exposure.](image-url)

- **Con.** (Control)
- **Positive Con.**
- **Conc. 150**
- **Conc. 500**
- **Conc. 1500**
RESULTS AND DISCUSSIONS-Cu exposure

![Graph showing tail DNA% for different concentrations of Cu exposure]
RESULTS AND DISCUSSIONS-Exposure in combination

No significant different (p<0.05) except for Cu\(^{2+}\) to PCB+Cu\(^{2+}\), and PCB+Zn\(^{2+}\)+Cu\(^{2+}\). Also PCB+Zn\(^{2+}\) and PCB+Zn\(^{2+}\)+Cu\(^{2+}\).
This demonstrated the genotoxicity of PCBs, Cu and Zn in the sentinel species.

The irreversible DNA damage seems to be mainly caused by PCBs or a combination with Cu and Zn, also the concentrations may be easily reached in natural ecosystem.

The in vitro cellular response to exposure may be a rapid approach to indicate environment stress in marine organisms.

Further in-depth studies will carry out to elucidate the relationship among these chemicals using other biomarkers such as micronucleus test and red neutral assay.

Analysis of bioaccumulation of pollutant will also be carried to explain the actually in vivo dose-response in mussels haemolymph.
ACKNOWLEDGEMENTS

Prof. Ying YE, Ocean College, Zhejiang University

Prof. Jianfang CHEN, Second Institute of Oceanography

Dr. Yanan DI, Ocean College, Zhejiang University

Directors, Nigerian Institute for Oceanography and Marine Research, Lagos Nigeria
THANK YOU FOR LISTENING