Inhibitory action of Persistent Organic Pollutant on Mitochondrial complex III of Zebrafish

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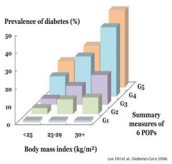
ABSTRACT

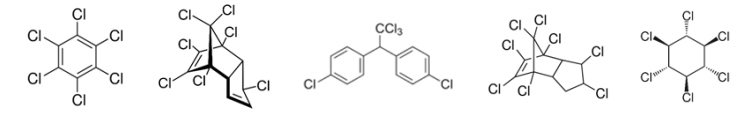
Synthetic environmental toxins are widely distributed with adverse effect on human health. Persistent organic pollutants(POPs) are synthetic chemicals that are resistant to degradation through chemical, biological and photolytic process. Meanwhile, several animal models like mouse or rats are used to study the effect of toxic materials. Zebrafish, well-known vertebrate organism, has strength as novel animal model in research of disease or toxicology due to its rapid ontogeny process and easier genetic and proteinic manipulation than other animal models. Also, it is beneficial to examine several exposure-effect from its water-survival nature. Here, 5 kinds of persistent organic pollutants mixture (*β*-HCH, Heptachlor, Hexachlorobenzene, 4, 4'-DDT and Chlordane) are directly treated to isolated mitochondria of zebrafish's liver to analyze its effect on mitochondrial activity. After adding POPs mixture, activity of each mitochondrial complex I, II, III and IV was measured immediately to examine the short-term exposing effect by spectrophotometric method. As a result, higher concentration of POPs treated mitochondria exhibited much more enhanced enzymatic activity than non-treated one except complex III. In the case of complex III, conversion of hexachlorobenzene contained in POPs into redox-active form of tetrachloro-1,4-benzoquinone in oxidative condition is competitively able to inhibit the Q binding site of mitochondrial complex III resulting in decreased enzymatic activity. Furthermore, competitive inhibitory action was confirmed by steady-state kinetic analysis of complex III with different concentrations of substrate and POPs mixture. This study suggests that brief in-vitro effects of POPs on mitochondrial OXPHOS system and suitability of zebrafish as toxicological risk-assessment model to analyze function of mitochondria with toxins.

INTRODUCTION

- Persistent organic pollutants(POPs) are synthetic chemicals that are related to metabolic diseases including type 2 diabetes
- Metabolic disease is commonly characterized by abnormal function of mitochondria
- Experimental evidence of their toxic action is limited

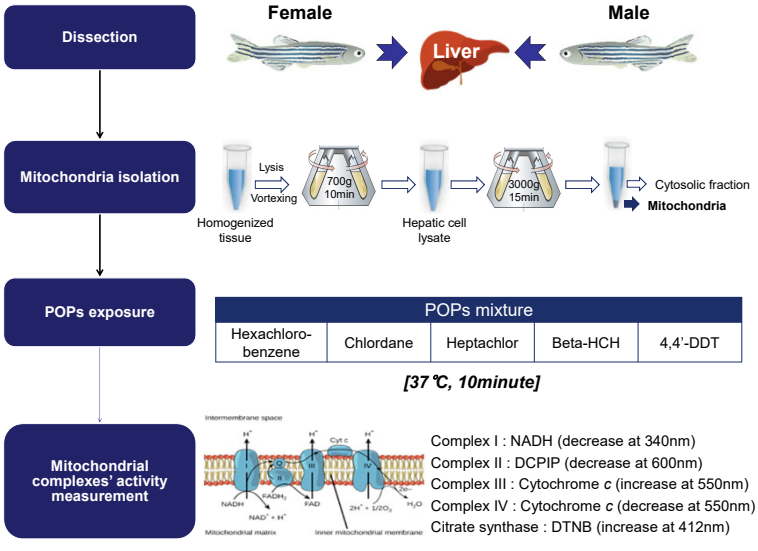


- Effect of POPs on mitochondrial function was analyzed using POPs mixture using isolated mitochondria of zebrafish
- Liver is selected as target organ, since it is representative organ with abnormal function with metabolic disease
- POPs were exposed as a mixture composed of 5 well-known POPs and
- Mitochondrial OXPHOS complexes' activity were measured in the presence of POPs



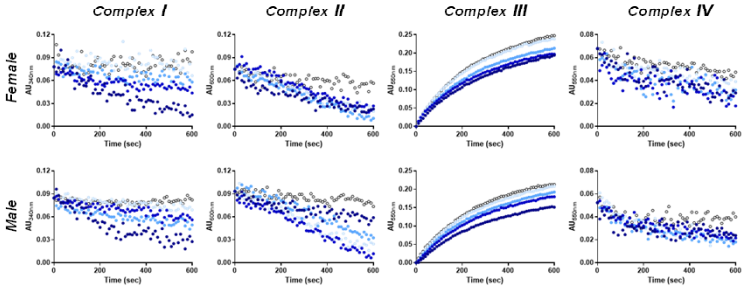
Hexachlorobenzene | Heptachlor | 4, 4'-DDT | Chlordane | *β*-HCH

MATERIALS AND METHODS



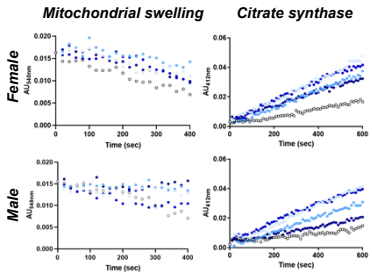
RESULT

[1] Activities of mitochondrial complexes were changed by POPs mixture



- Activity of complex I, II and IV were enhanced whereas activity of complex III was inhibited
- Female and male exhibited same tendency

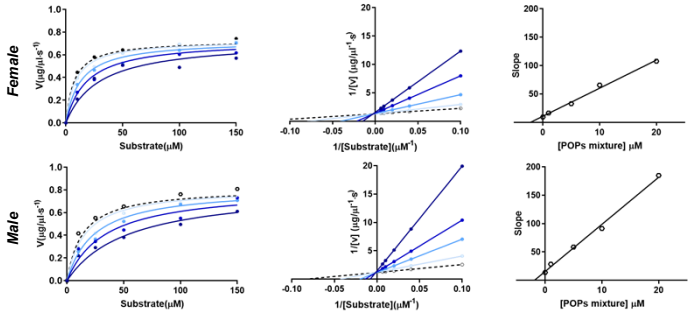
[2] Effect of POPs mixture on mitochondrial ion channel



- POPs exposed mitochondria exhibited **uncontrolled ion regulation**
- Activity of citrate synthase was enhanced with POPs exposure
- **POPs have an effect on opening of calcium channel in mitochondria**

[3] Competitively inhibited activity of mitochondrial complex III

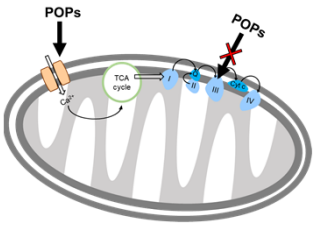
- **Activity of complex III was inhibited** depending on the concentration of POPs mixture
- Hexachlorobenzene might be converted into redox active form of tetrachloro-1,4-benzoquinone in oxidative condition by other POPs



Steady-state kinetic analysis show POPs as competitive inhibitor of complex III

CONCLUSION

The results of this study show *in vitro* exposure of POPs influences hepatic mitochondrial function of zebrafish. Firstly, POPs enhances activity of complex I, II, IV and citrate synthase by opening of mitochondrial calcium channel and boosting of mitochondrial OXPHOS [2]. In contrast, POPs mixture competitively inhibits activity of complex III probably due to oxidation of hexachlorobenzene to tetrachloro-1,4-benzoquinone functioning as a redox-active compound [3].



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