

# EMBRYOTOXIC AND BEHAVIORAL EFFECTS IN ZEBRAFISH EMBRYOS (*DANIO RERIO*) AFTER EXPOSURE TO BINARY MIXTURES OF ENVIRONMENTAL POLLUTANTS

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## Background

Environmental contamination is usually comprised of a complex mixture of pollutants, each of them bearing the potential of causing different toxic responses towards humans and wildlife. However, current risk assessment approaches are still focusing on single compound evaluation. In contrast, present research shows that mixture components can act together to cause a toxic effect. The concentration addition (CA) and independent action (IA) models have been developed to more easily predict mixture toxicity, and are based on dose-response curves of single compounds. Both models assume that there is no interaction between the components, resulting in an additive mixture effect. In comparison, a greater (synergistic) or lower (antagonistic) mixture effect than the predicted additive effect indicates mixture interactions.

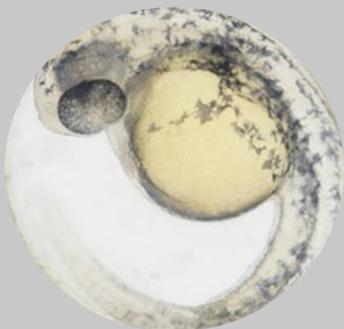
## Mixture toxicity



## Materials & Methods

### Test system:

Zebrafish embryo acute toxicity test (FET, OECD no. 236).



### Exposure:

- 96 hours
- Benzo[a]pyrene (BaP)+Perfluorooctanesulfonic acid (PFOS) or 3,3',4,4',5-pentachlorobiphenyl (PCB126)+PFOS
- $LC_{25} + LC_{25}$  = highest mixture concentration
- 1:2 dilution series

### Selection of endpoints:

- Lack of heartbeat
  - Formation of somites
  - Detachment of tail
  - Coagulation
  - Malformations
  - Edema
  - Behavioral alterations
- } Lethal effects

## Results

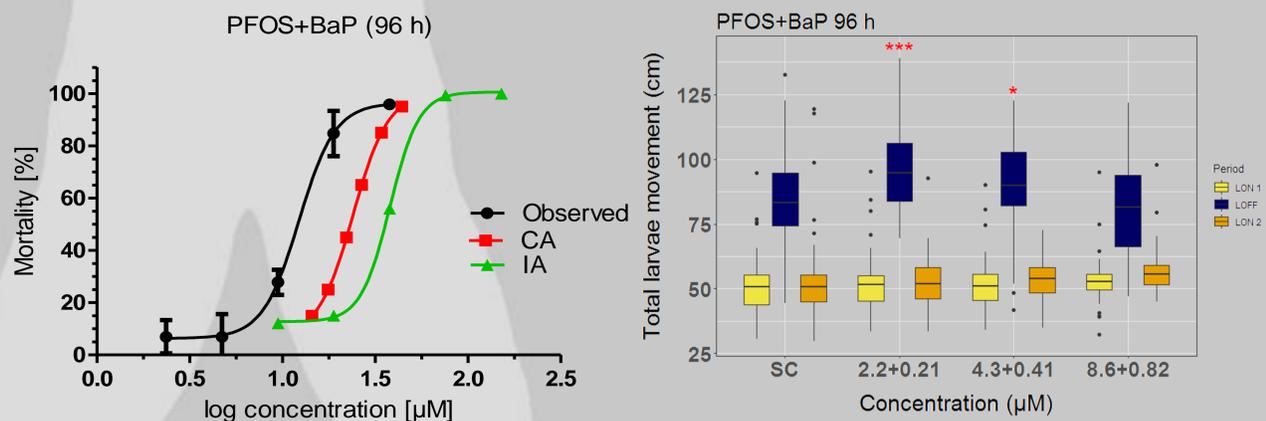


Figure 1. An equitoxic mixture ( $LC_{25} + LC_{25}$ ) containing PFOS (91 %) and BaP (9 %) caused a synergistic effect in zebrafish larvae (left figure). In addition, the mixture resulted in hyperactivity in darkness (right figure). CA = concentration addition, IA = independent action, SC = solvent control, LON 1/LON 2 = first (yellow)/second (orange) period of light, LOFF = period of darkness (blue).

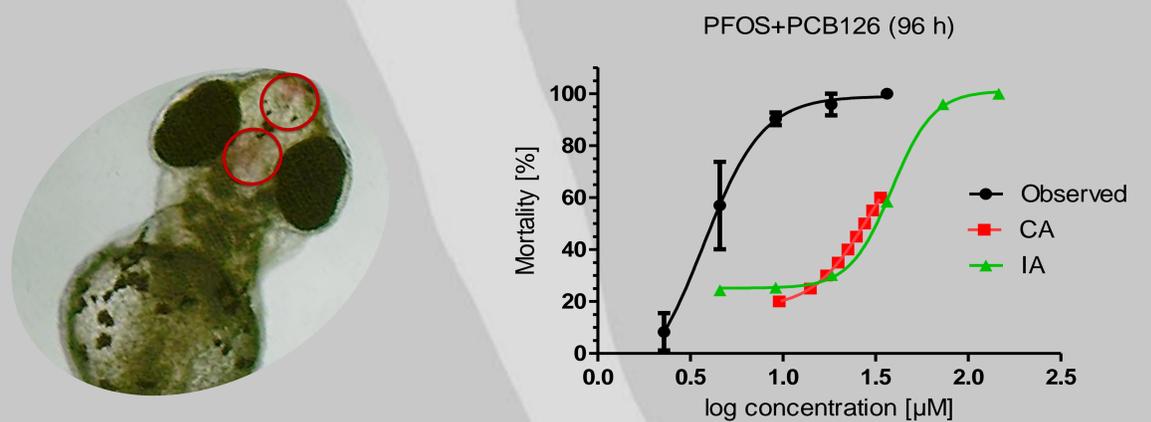


Figure 2. An equitoxic mixture ( $LC_{25} + LC_{25}$ ) containing PFOS (94.5 %) and PCB126 (5.5 %) resulted in pericardial edemas, lack of blood circulation and accumulation of red blood cells in the head of zebrafish larvae (left figure). In addition, PFOS and PCB126 interact to result in a synergistic effect (right figure). CA = concentration addition, IA = independent action.

## Conclusion & Outlook

The present study shows that models used to predict mixture toxicity tend to underestimate the risks that binary mixtures are posing. However, to better understand the mixture toxicity and the underlying mechanisms, metabolomics and qPCR analysis will be performed.