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Effects of Bisphenol-S and Estrogen on p53 Expression in Ovarian Tissue of Zebrafish (Danio rerio)

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Background
- Bisphenol A (BPA) is one of the most widely used plasticizing compounds
  - BPA is an estrogen analogue - has the potential to affect development
- As BPA use has been decreased, a similar compound, bisphenol S (BPS), is being used in its place (Fig. 1)
  - Data available have shown BPS has a similar cytotoxicity as BPA at lower doses and the potential to induce DNA damage
- Apoptosis plays a key role in ovarian homeostasis
  - p53 is one of the genes that regulates apoptosis
  - Only in recent studies has the interaction between p53 and estrogen been shown

Fig. 1. Chemical structures of estradiol, bisphenol A, and bisphenol S (adapted from Environmental Estrogen – Bisphenol A by K. Zhang, 2014)

Objectives
- Observe the effects of BPS exposure on p53 gene expression in ovary tissue
- Observe the effects of estradiol (E2) on p53 gene expression in ovary tissue
- Compare BPS and E2 exposure to see if BPS is acting as an estrogen mimic
- Expand field of knowledge about possible health effects of BPS

Methods
- Zebrafish ovarian tissue samples were exposed to 0.01, 0.1 and 1.0 µM of BPS for two hours
- mRNA was extracted from ovarian cells and converted into cDNA
- qPCR was run using primers for p53 and β-actin as the reference gene. The relative amount of gene expression was compared to the amount of reference gene expression using CFX96 Real Time PCR Software
- Procedure the same for exposure to estradiol (E2)

Results & Conclusion
- Both estrogen and BPS have been known to have non-monotonic dose responses (Eladak et al., 2015)
  - Results confirm non-monotonic response in both BPS and E2 exposed ovarian tissue
    - At 1.0 µM-BPS, p53 expression increased compared with the control (Fig. 2)
    - At 1.0 µM-E2, p53 expression decreased, but at lower concentrations increased (Fig. 3)
  - BPS and E2 affect p53 expression in ovarian tissue, however in opposite trends. Higher concentrations of BPS suggest p53 expression will increase, while at lower concentrations, decreased or no change in expression. Higher concentrations of E2 suggest p53 expression will decrease, while at lower concentrations, increase in expression.

Fig. 2, 3. Real-time qPCR analysis of mRNAs for p53 in zebrafish ovarian tissue exposed to 0.01, 0.1 and 1.0 µM of BPS or an ethanol control for two hours. Exposure of ovarian tissue to 1.0 µM BPS increased p53 mRNA levels several fold over ovarian tissue exposed to 0.01, 0.1 µM BPS and control conditions. Values of p53 were normalized to β-actin.

Future Directions
- Continue in vitro studies of exposure of ovarian tissue to BPS and its effects on p53 expression
  - Protein content
- Study the role of p53 and other essential genes during development, such as Sonic Hedgehog (SHH)

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References

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