Bisphenol A impairs brain formation and neurodevelopment by enhancing *HOX* genes

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Plastic pollution and exposure to synthetic chemicals are becoming a serious problem. There has been accumulating evidence that exposure to endocrine disrupting chemicals (EDCs) during development can lead to various adverse effects in humans. Brain and neuronal development is highly dependent on a combination of multiple signaling molecules and considered to be susceptible to EDCs. In fact, there are an increasing number of incidents of neurodevelopmental impairments such as autism spectrum disorder (ASD) and attention deficit hyperactivity disorder (ADHD). However, the causal link between these neurodevelopmental disorders and EDCs is not clear. One of the reasons for this is due to the lack of a suitable testing method.

Here, we used iPSC and zebrafish embryos to investigate the effect of Bisphenol A (BPA) on brain and neuronal development. BPA is the most highly studied EDC and has been widely used in manufacturers of polycarbonate plastics and epoxy resins including food and drink packages, baby bottles, dental sealants, and thermal receipt paper. We found that BPA enhanced *HOX* gene expression in the presence of exogenous retinoic acid (RA), but not when BPA was used alone in iPSC. This effect was abolished with RAR antagonists, but not with estrogen receptor antagonists. To verify their physiological relevance, we also applied BPA to hindbrain formation in zebrafish. Spatial expression analysis of *hoxb1a, fgf8*, and *otx2* revealed that BPA enhanced RA-induced posteriorization of the brain region and resulted in the duplication of Mauthner cells and the abnormal craniofacial cartilage formation, but not when BPA was used by itself. Transcriptome analysis showed that 3'*HOX* genes and genes related to brain formation exhibit a similar expression profile by the co-exposure to BPA and RA in iPSC and zebrafish. We compared these transcriptomes using GO-based gene set enrichment analysis and revealed characteristic clusters, which consists of GO terms related anterior/posterior pattern specification, brain formation, and skeletal system development. These GO terms were highly related to the anomalies we observed, suggesting that the chemical treatment and anomalies could be linked with transcriptional change. This result also implies that iPSC can be a useful model for predicting adverse effects in zebrafish and possibly provide a new testing method in environmental sciences.

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