Effects of Polycyclic Aromatic Hydrocarbons on Estrogen-Induced cAMP Responsive Element Binding Protein Expression and Activity in Developing Cortical Neurons

中文摘要

英文摘要

Environmental hormones such as polycyclic aromatic hydrocarbons (PAHs) are known to interrupt various physiological functions of estrogen via activation of arylhydrocarbon receptor (AhR). It has been shown previously that estrogen plays an important role in neuronal development. In this study, we primary cultured cortical neurons at 5 days in vitro to investigate the effects of 3-methylcholanthren (3-MC), a PAH and also an AhR agonist, on the activity of cAMP responsive element binding protein (CREB) mediated by estrogen. At first, we used Western blot analysis and luciferase activity assay to confirm that 3-MC can induced AhR ubiquitination and transcriptional activation of DRE promoter in the cultured developing cortical neurons. 17- -estradiol (E2) treatment increased CREB expression and phosphorylation, and was attenuated by pre-treatment but not post-treatment with 100 nM of 3-MC. We further found that E2 enhanced protein-protein interaction between ER and CREB, and this effect was decreased by 3-MC pretreatment. Since CREB is known as one of the major transcription factors for tyrosine hydroxylase (TH) gene expression, we further examined if cAMP responsive element (CRE), known as the CREB binding region, on Th gene promoter are affected by 3-MC. The developing cortical neurons were transfected with the Th-CRE construct with luciferase as the reporter gene, and showed high basal level of luciferase activity. Treatment with E2 significantly enhanced the Th-CRE-driven luciferase activity. The estradiol increased Th-CRE promoter activity was significantly reduced by estrogen receptor antagonist ICI-182,780, MAPK inhibitor PD98059 and CaMKII inhibitor KN-93. However, pretreatment with 3-MC showed further enhancement but not reduction of the estradiol-induced CRE activity. Lastly, Western blot analysis showed that TH expression enhanced by E2 was indeed reduced by 3-MC. These results suggest that estrogen-mediated CREB expression and function in developing cortical neurons are reduced by PAH. However, PAH decreased estradiol-elevated TH expression but enhanced estradiol-induced Th-CRE promoter activity implicates that PAH might have both anti-estrogenic and estrogenic effects on the TH gene expression.