Function Analysis of Tilapia(Oreochromis mossambicus) Hepatocyte Nuclear Factor-3 β Promoter Fragments

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ABSTRACT

The hepatocyte nuclear factors-3 (HNF-3) family members HNF-3 α , HNF-3 β and HNF-3 γ are hepatocyte-enriched transcription factors, they play important roles in controlling development, differentiation, metabolism and organogenesis. In our previous study, the expression of insulin-like growth factor-I/II (IGF-I/II), HNF-1 α , -1 β and -3 β were detected in the liver and gonads of tilapia, and expreession level of HNF-3 β was higher than others and it could be regulated by 17 β -estradiol. In this study, four fragments (0.5, 1.0, 1.5 and 2.0 kb) of tilapia HNF-3 β promoter were constructed with green fluorescent protein (GFP) gene for biological activity assay by performing transfection into tilapia ovarian cell line (TO-2) and human hepatoma cell line (Hep3B) for western blot analysis and luciferase assay or microinjection into zebrafish eggs and assay. The GFP was mainly expressed in yolk and somites 24 h after injection, in notochord and floor plate 96 h after injection. The 0.5 kb fragment was expressed in notochord, yolk, eye and head, the expression rates were 18.3%, 1.3%, 35.6% and 26.0%, respectively; the 1.0 kb fragment was expressed in notochord, yolk, and head, the expression rates were 44.4%, 44.4% and 2.0%, respectively; the 1.5 kb fragment was expressed in notochord and yolk, the expression rates were 33.7% and 50.5% respectively, and the 2.0 kb fragment was expressed in notochord, yolk and head, the expression rates were 61.2%, 26.1% and 2.7% respectively. The results were similar to our previous studies. The supplement of add 17 β -estradiol enhanced western blot analysis of eGFP expression and luciferase assay in TO-2 and Hep3B cells. Based on the present results, hypothesizing that estrogen response element (ERE) in tilapia HNF-3 β promoter could promote the expression of HNF-3 β in the gonads of tilapia through the action of steroids.

Keywords : tilapia、gonad、hepatocyte nuclear factors-3 (HNF-3)、promoter、steroid hormones

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