DIFFERENTIAL REGULATION OF GONADOTROPIN RECEPTORS (fshr and lhcgr) BY ESTRADIOL IN THE ZEBRAFISH OVARY INVOLVES NUCLEAR RECEPTORS THAT ARE LIKELY LOCATED ON THE PLASMA MEMBRANE

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Introduction:

Follicle-stimulating hormone (FSH) and luteinizing hormone (LH) are gonadotropins that control all major events of gonadal function, including folliculogenesis, steroidogenesis and ovulation in females. FSH and LH signal through their cognate receptors, FSH receptor (FSHR) and LH/choriogonadotropin receptor (LHCGR), respectively, across vertebrates. Compared to the information in mammals, very little is known about these receptors in fish, especially the mechanisms that control their expression. Using zebrafish as the model, we have demonstrated that although both fshr and lhcgr increase their expression during follicle growth and maturation in the ovary, they exhibit significant temporal difference with lhcgr expression lagging behind that of fshr [1]. This raises an interesting question: how are these two gonadotropin receptors differentially controlled during folliculogenesis? We, therefore, have initiated this study to investigate the expression control of fshr and lhcgr in the zebrafish ovary.

Methods:

A primary follicle cell culture was used in the present study to investigate the regulation of fshr and lhcgr by various hormones and their action mechanisms. The expression levels of fshr and lhcgr were assessed by extraction of total RNA from the treated cells followed by reverse transcription and real-time qPCR. To elucidate the signaling mechanism, we collected the total protein from the treated cells to examine the phosphorylation level of certain signaling molecules with Western blot.

Results:

Estradiol (E2), but not testosterone (T), significantly and differentially up-regulated the expression of fshr and lhcgr within 1 hour. Although E2 stimulated the expression of both fshr and lhcgr, its effect on the steady-state level of lhcgr mRNA was much higher (>10-fold up-regulation) than that on fshr expression (~0.5-fold increase). This regulatory effect could be mimicked by E2 conjugated to bovine serum albumin (E2-BSA). Additionally, nuclear estrogen receptor (ER) antagonists (ICI 182,780) and transcription inhibitor (actinomycin D) abolished this E2-induced up-regulation while MEK inhibitor (U0126) greatly hindered the action of E2. Further immunoblot evidence confirmed that E2 rapidly induced phosphorylation of MAPK in less than 20 minutes and the activation of p38 MAPK could enhance the E2 action.

Conclusion:

We have provided the first evidence that E2 acts as a potent endocrine hormone involved in the differential expression of fshr and lhcgr in the zebrafish ovary. The regulation occurs at the transcription level via the nuclear ERs; however, these receptors appear to be located on the plasma membrane. This study has provided evidence for a novel action mechanism of E2 in the ovary of teleosts and the distinct mechanisms controlling lhcgr expression in mammals and teleosts.

Reference: