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Androgen Stimulation of GnRH Receptor Gene Expression Requires Multiple Transcription Factor Bindings in the GnRH Receptor Gene Promoter

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Introduction Gonadotropin-releasing hormone (GnRH), a hypothalamic decapeptide, is well known to be a master regulator in mammalian reproduction. The responsiveness of pituitary gonadotropes to GnRH is partially related to the number of cell surface GnRH receptor (GnRHR) and is very important in GnRH action and this is affected by steroid hormone melieu. Although it is reported that androgen can stimulate GnRHR gene expression, little is known about the molecular mechanisms of the androgen regulation of the GnRHR gene. According to the bioinformatic analysis, there is no possible androgen response element (ARE) in GnRHR gene promoter. In this study, therefore, we investigate involvement of other putative transcription factors rather than androgen receptor (AR) in androgen regulation of mouse GnRHR (mGnRHR) gene expression.

Methods αT3-1 cells were transfected with a 1.2 kb mGnRHR gene promoter-luciferase reporter construct (-1164/+62 GnRHR Luc). The luciferase activity was measured as an indicator for gene transcription after dihyroxytestosterone (DHT) treatment for 48 h. Functional transfection studies with serial 5'-deletion mutants of the mGnRHR gene promoter and mutants for binding sites of transcription factors, Oct-1, NF-Y and AP-1, were performed.

Results DHT treatment (10 nM) stimulated a dose-dependant increase in luciferase activity, maximal at 10^{-8} M. Functional transfection analysis indicated that deletion of sequences between -300 and -232

reduced the DHT-stimulated increase in luciferase activity. This region harbors Oct-1, NF-Y, and AP-1 binding elements. Combinatorial mutation of the Oct-1/NF-Y or Oct-1/AP-1 binding sites resulted in significant decrease in stimulation by DHT.

Conclusion Androgen stimulation of GnRHR gene expression requires intact Oct-1, NF-Y, and AP-1 binding sites in the GnRHR gene promoter, and suggest that the effects of androgens may be mediated by an interaction of AR with these transcription factors.