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JOURNAL ARTICLE

E2F and GATA-1 are required for the Sertoli cell-specific promoter activity of the follicle-stimulating hormone receptor gene

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The follicle-stimulating hormone receptor (FSHR) gene is expressed in Sertoli cells in males and in granulosa cells in females. Cis-acting sequences and associated binding factors responsible for the transcription of the TATA-less FSHR gene in Sertoli cells were analyzed with dimethylsulfate (DMS) footprinting assays and electrophoretic mobility shift assay (EMSA). In vivo footprints in the core promoter using nuclear proteins from Sertoli cells identified several protected sequences, including an inverted GATA (TATC, -88/-85), and an E2F (TTTCGCG, -45/-39) motif. EMSA showed the presence of one or more sequence-specific proteins interacting with these potential regulatory elements. Antibody-supershift assays as well as competition assays further revealed that testis-specific GATA-1 recognized the inverted GATA element. The functional role of the potential cis-acting elements was analyzed by transient transfection assays with and without mutations of the putative elements. The mutational analysis indicated that the GATA and E2F elements were each required for optimal promoter activity. The effects of each of the promoter elements was examined in transfections in which mutations were made in each of the known regulatory sites, including the E box, GATA, and E2F sites in various combinations. All of these sites contribute to the maximum promoter activity such that mutations of the E box, GATA, and E2F sites eliminated nearly all promoter activity.

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