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Role of Specificity Protein-1 and Activating Protein-2 Transcription Factors in the Regulation of the Gap Junction Protein Beta-2 Gene in the Epididymis of the Rat¹

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Abstract

In prepubertal rats, connexin 26 (GJB2) is expressed between adjacent columnar cells of the epididymis. At 28 days of age, when columnar cells differentiate into adult epithelial cell types, Gjb2 mRNA levels decrease to barely detectable levels. There is no information on the regulation of GJB2 in the epididymis. The present study characterized regulation of the Gjb2 gene promoter in the epididymis. A single transcription start site at position –3829 bp relative to the ATG was identified. Computational analysis revealed several TFAP2A, SP1, and KLF4 putative binding sites. A 1.5-kb fragment of the Gjb2 promoter was cloned into a vector containing a luciferase reporter gene. Transfection of the construct into immortalized rat caput epididymal (RCE-1) cells indicated that the promoter contained sufficient information to drive expression of the reporter gene. Deletion constructs showed that the basal activity of the promoter resides in the first –230 bp of the transcriptional start site. Two response elements necessary for GJB2 expression were identified: an overlapping TFAP2A/SP1 site (–136 to –126 bp) and an SP1 site (–50 bp). Chromatin