TRANSCRIPTIONAL REGULATION OF THE GP91-PHOX GENE

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ABSTRACT

The gp91-phox gene, which encodes a subunit of the NADPH oxidase, is nearly exclusively transcribed in terminally differentiating myeloid cells. The -450 to +12 base pair fragment of the gp91-phox promoter has been found to be sufficient to direct appropriate expression of linked reporter genes in a subset of monocyte/macrophages in transgenic mice, and multiple DNA binding proteins interact with the proximal gp91-phox promoter. Among them, CCAAT displacement protein (CDP) binds to four sites within this promoter region, and competes with transcriptional activators for binding to overlapping sites. CDP binding to each site is down-regulated during myeloid differentiation, coincident with induction of gp91-phox expression. Specific ablation of CDP binding sites leads to an increase in promoter activity in cells not expressing the endogenous gp91-phox gene. These results suggest that CDP functions as a transcriptional repressor of the gp91-phox promoter. Increased promoter activity is also exhibited by the -102 bp to +12 region of the gp91-phox promoter that lacks these four CDP binding sites. Derepressed promoter activity is dramatically decreased following a 16 bp truncation to -86 bp that deletes an interferon stimulated response element (ISRE) from the gp91-phox promoter. This ISRE is recognized by BID, a DNA-binding protein that interacts with three upstream sites within the gp91-phox promoter, as well as by IRF-1 and IRF-2. Both the IRF and BID binding sites are required to mediate activation of gp91-phox promoter activity in the absence of CDP binding. Overexpression of IRF-1 and IRF-2 leads to transactivation of gp91-phox promoter constructs. However, IRF-2 is predominant in the cell lines used in these studies. This suggests that IRF-2 acts as a transcriptional activator to direct derepressed gp91-phox promoter activity. Therefore, transcription of the gp91-phox gene is under the complex control of multiple repressors and activators, and down-regulation of CDP DNA binding activity is one necessary step for expression of the gp91-phox gene in mature myeloid cells.

TABLE OF CONTENTS