【P-38】

Repression of the TGF-β1 Gene by PPARγ-RXRα Heterodimer: Transrepression Without Direct Interactions with the Promoter DNA Binding Elements

Seung Jin Lee and Sang Geon Kim

National Research Laboratory, College of Pharmacy and Research Institute of Pharmaceutical Sciences, Seoul National University, Seoul 151-742, Korea

Peroxisome proliferator-activated receptor-γ (PPARγ) and retinoic acid X receptor (RXR) heterodimer regulates cell growth and differentiation. This study examined whether activation of PPARγ and RXR affects the transforming growth factor- β1 (TGFβ1) gene expression, and if so, what the molecular basis is for the gene regulation. Treatment of L929 fibroblasts with either 15-deoxy-d(12,14)-prostaglandin J_2 (PGJ_2) or 9-cis-retinoic acid (RA) decreased the TGFβ1 gene expression. When compared to PGJ_2 or RA alone, combination treatment with PGJ_2+RA synergistically repressed constitutive and TGFβ1-inducible TGFβ 1 expression, which was abrogated by PPARγ antagonists. Also, PGJ_2+RA or ectopic expression of the PPARγ-RXRα heterodimer decreased the luciferase reporter gene activity from the TGFβ1 gene promoter. The pGL3-323 that comprises the -323 bp TGFβ1 promoter, but lacks PPAR-responsive elements (PPREs), allowed PGJ_2+RA to repress luciferase expression, indicating that the PPREs present in the upstream region are nonfunctional. The band intensities of protein binding to the NF-1, ZF-9 or SP-1 binding sites (-323 bp to -175 bp) were unchanged by PGJ_2+RA treatment. Deletion of the upstream region comprising the AP-1 binding sites (-453 bp to -323 bp) markedly decreased the basal TGFβ1 expression. Although AP-1-DNA binding and AP-1 reporter activities were both unaffected by PGJ_2+RA, specific mutation of the proximal AP-1 site decreased luciferase expression from pGL3-453, resulting in the loss of its responsiveness to PGJ_2+RA. Our data provide evidence that PGJ_2+RA represses TGFβ1 expression via PPARγ-RXRα without its interactions with the promoter DNA elements, and that the constitutive AP-1 activity plays a crucial role in PPARγ-RXR responsiveness.

Keyword: TGFβ1, PPARγ, RXRα