Transcriptional regulation of Rat Endothelial Nitric Oxide Promoter in Pulmonary Myofibroblasts cells and its implications in Pulmonary Fibrosis

Dominic Ochwang’i a,*, Charles Kimwele a, Stephen G. Kiama a, and Nancy Rice b
a Department of Veterinary Anatomy and Physiology, Faculty of Veterinary Medicine, University of Nairobi, Kenya;
b Department of Biology, Western Kentucky University, USA.

*Corresponding author: Department of Veterinary Anatomy and Physiology, Faculty of Veterinary Medicine, University of Nairobi, P.O. Box 30197, Nairobi 00100, Kenya; Tel: +254-72-3759997; Email: omosake@yahoo.com

Background: Nitric oxide (NO) levels may exert control on the persistence of pulmonary myofibroblast cells in pulmonary fibrosis.

Objective: This study set out to examine the regulation of NO levels by transcription factors that influence the expression of the endothelial nitric oxide synthase (eNOS3) gene.

Methods: Using a pGL3-Basic vector plasmid DNA, rat and human eNOS3 gene promoters were inserted upstream of a luciferase reporter gene and cloned in competent E. coli cells (DH5α). Transfection assays were performed and the cells treated with potential regulators of eNOS3 gene. Promoter activity of eNOS3 gene was assayed using the Dual Luciferase reporter gene assay.

Results: The results indicated that the rat NOS3 promoter was active in the cells, with the human NOS3 promoter showing little or no activity. The results demonstrated that transforming growth factor-β, EGTA and lipopolysaccharide up-regulated transcriptional activity while Phorbol 12-myristate-13-acetate, 23187 and S-nitroso-N-acetylpenicillamine, suppressed eNOS3 transcriptional activity. Treatment with Nω-Nitro-L-arginine methyl ester had no effect on the gene expression.

Discussion: The results of this study demonstrates that high concentrations of NO inhibit NOS3 gene activity, hence an enhanced expression of eNOS in response to pharmacological interventions using some transcriptional factors from these study could provide protection against interstitial pulmonary.

Key words: Nitric oxide, transcription, NOS3 gene promoter, pulmonary fibrosis

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1. Introduction

Myofibroblasts in idiopathic pulmonary fibrosis

Myofibroblasts are mesenchymal cells that possess both fibroblast and muscle-like characteristics and function in tissue development, remodeling and wound repair (Gabbiani, 1996). During normal wound healing myofibroblasts undergo apoptosis (Darby et al, 1990; Clark, 1993), but in certain circumstances, these cells persist and continue to secrete extracellular matrix. Persistent myofibroblasts have been implicated in African Journal of Pharmacology and Therapeutics Vol. 2 No. 1 Pages 1-8, 2013 Open Access to full text available at http://www.uonbi.ac.ke/journals/kesobap/ Research Article


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