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Identification of housekeeping basal promoter 5'pcmah-2 of pig CMP-N-acetylneuraminic acid hydroxylase gene

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Sialic acid is a generic term used for neuraminic acid and its many derivatives. N-glycolylneuraminic acid (NeuGc) is one of the most common sialic acid types found in most mammals except normal humans. NeuGc is closely linked with various diseases including cancer, infections and immune rejection response during pig-to-human xenotransplantation. Biosynthesis of NeuGc is mediated by a specific hydroxylase, cytidine-5'-monophospho-N-acetylneuraminic acid hydroxylase (CMAH), that converts CMP-NeuAc to CMP-NeuGc. In the present study, we isolated pCMAH house-keeping promoter regions (Ph), which are responsible for transcriptional regulation and which are located upstream of the alternative transcript pcmah-2. Luciferase reporter assays using serial construction of each deleted promoter demonstrated that the Pc promoter was highly active in pig-derived kidney PK15. Ph promoter of pcmah lacked a TATA box, but contained three putative Sp1 binding sites. Mutations of these Sp1 binding sites always resulted in the reduction of luciferase activities in Ph-334. In addition, treatment with mithramycin A (25–100 nM) decreased the luciferase activities of the Ph promoters in a dose-dependent manner. Electrophoretic mobility shift assay analysis revealed that the probes containing each Sp1 binding site bound to Sp1. Taken together, the results indicate that Sp1 bind to their putative binding sites on the Ph promoter regions of pcmah and positively regulate the promoter activity in pig kidney cells.

Biography

Choong-Hwan Kwak graduated from Food and Biotechnology Department, KyungSung University, Busan, Korea in 2010. He obtained Master of Science from the Molecular and Cellular Glycobiology Lab, Department of Biological Science, Sungkyunkwan University, Suwon, Korea in 2012 and presently, he is a PhD candidate at the same laboratory under supervisor Prof Cheorl-Ho Kim, Sungkyunkwan University. His research interest is glycan structure and biosynthesis of glycoproteins, glycoconjugates and glycolipids and NeuGc synthesis by CMP-N-acetylneuraminic acid hydroxylase.

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