P151

Transcriptional Regulation of the Human CMP-NeuAc:GM3 α2,8 Sialyltransferase (GD3 synthase) Gene in Human Melanoma Cells

Nam-Young Kang[1], Hyun-Mi Dae[1], Kyoung-Sook Kim[1],
Cheorl-Ho Kim[3] and Young-Choon Lee[1,2]

[1]Brain Korea 21 Center for Silver-Bio Industrialization and [2]Department of Biotechnology, Dong-A University, Busan 604-714, Korea, [3]Department of Biological Sciences, SungKyunKwan University, Kyunggi-Do 440-746, Korea

Promoter analyses of the 5′-flanking region of the human GD3 synthase gene using luciferase gene reporter system showed the strong promoter activity in SK-MEL-2 cells. Deletion study revealed that the region as the core promoter from -1146 to -646 (A of the translational start ATG as position +1) was indispensable for endogenous expression of human GD3 synthase gene. This region lacks apparent TATA and CAAT boxes, but contains putative binding sites for transcription factors c-Ets-1, CREB, AP-1 and NF-κB. Electrophoretic mobility shift assays using specific competitors, chromatin immunoprecipitation assay and site-directed mutagenesis demonstrated that only NF-κB element in this region is required for the promoter activity in SK-MEL-2 cells. These results indicate that NF-κB plays an essential role in the transcriptional activity of human GD3 synthase gene essential for GD3 synthesis in SK-MEL-2 cells.

Key words: GD3 synthase, promoter, human melanoma, NF-κB, transcription

P152

Transcriptional Regulation of Human GM3 Synthase Gene induced by Valproic acid in SK-N-BE(2)-C Cells

Hyun-Mi Dae[1], Nam-Young Kang[1], Hwa-Young Kwon[2],
Kyoung-Sook Kim[1] and Young-Choon Lee[1,2]

[1]Brain Korea 21 Center for Silver-Bio Industrialization and [2]Department of Biotechnology, Dong-A University, Busan 604-714, South Korea

We investigated how valproic acid (VPA) mediates enhanced expression of ganglioside GM3 synthase gene in this study. The result showed that VPA up-regulates expression of GM3 synthase gene through the activation of mitogen-activated protein (MAP) kinase and phosphatidylinositol 3-kinase (PI-3K)/AKT signal cascade controlling survival and apoptosis. In addition, PI-3K/AKT activation induced by VPA in SK-N-BE(2) cells led to the phosphorylation of cAMP-responsive element binding protein (CREB) as a transcription factor. Functional analysis of the 5′-flanking region of GM3 synthase gene by the transient expression method showed that the -177 to -83 region, which contains a CREB binding site at -143, functions as the VPA-inducible promoter in SK-N-BE(2) cells. In addition, site-directed mutagenesis indicated that the CREB binding site at -143 is crucial for the VPA-induced expression of the GM3 synthase gene in SK-N-BE(2) cells. These results indicate that transcription factor CREB plays an essential role in the transcriptional regulation of human GM3 synthase through the activation of PI-3K/AKT pathway in VPA-induced SK-N-BE(2) cells.

Key words: GM3 synthase, valproic acid, promoter, CREB, transcription