

속수질 집합관 상피세포에서 dDAVP에 의한 제 2형 수분통로단백 (aquaporin-2)의 세포막 수송에 70 kDa Heat Shock Protein의 역할

경북대학교 의학전문대학원

박의정, 임정숙, 정현준, 최효정, 권태환

The Role of 70-kDa Heat Shock Protein in dDAVP-induced AQP2 Translocation in Kidney Collecting Duct Cells

Eui-Jung Park, Jung-Suk Lim, Hyun Jun Jung, Hyo-Jung Choi, Tae-Hwan Kwon

Kyungpook National University School of Medicine

The aquaporin-2 (AQP2) is a vasopressin-regulated water channel in the kidney collecting duct principal cells. Several proteins including heat shock protein 70-kDa (Hsp70 and Hsc70) have been identified to be able to interact with C-terminus of AQP2, where phosphorylation at Ser 256 residue occurs. In this study, we examined the role of Hsp70 in regulation of AQP2 trafficking and expression. Semiquantitative immunoblotting revealed decreased expression of AQP2 and Hsp70 in the kidneys of rats with hypokalemia-induced nephrogenic diabetes insipidus. In contrast, increased expression of AQP2 and Hsp70 was seen in rats with long-term dDAVP infusion via osmotic minipumps (40 ng/h for 5 d, s.c.). This was further revealed by immunohistochemistry. In silico analysis of 5'-flanking regions of AQP2, Hsp70-1 and Hsp70-2 genes revealed that transcriptional regulator binding elements associated with cAMP response are present at the promoter regions of Hsp70-1 and Hsp70-2, in addition to AQP2. Luciferase reporter assay demonstrated that MDCK cells transfected with 1 kb of 5'-flanking region in Hsp70-2 construct revealed a significant increase of luminescence after dDAVP stimulation (10⁻⁸M, 3 h). Moreover, cell surface biotinylation analysis of the mpkCCDc14 cells with siRNA-mediated knockdown of Hsp70-2 demonstrated significantly decreased forskolin-induced AQP2 targeting to the apical plasma membrane. Consistent with this, forskolin-induced AQP2 phosphorylation (Ser 256) was significantly reduced in the mpkCCDc14 cells with siRNA-mediated Hsp70-2 knockdown. Taken together, Hsp70-2 is likely to play a role in AQP2 trafficking to the plasma membrane by affecting AQP2 phosphorylation at Ser 256 residue (PKA phosphorylation consensus site).

Key Words: 제 2형 수분통로단백, 샤페론, 수송

Aquaporin-2, Heat shock protein, Trafficking