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제2형 수분통로 세포 내수송에서 Rab GTPase 활성 단백질인 AS160의 역할

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The Rab GTPase-activating Protein AS160 as a Negative Regulator for AQP2 Trafficking in Collecting Duct Cells

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Background : PI3K and Akt are known to be activated in response to vasopressin in the collecting duct. However, it is unclear whether the PI3K/Akt pathways regulate AQP2 trafficking. AS160 is a novel Akt substrate of 160 kDa containing a Rab GTPase-activating protein (GAP) domain. The present study examined whether Akt and AS160 play a role in the AQP2 trafficking.

Methods : The strategy was to examine whether siRNA-mediated AS160 knockdown in mouse cortical collecting duct cells (M-1 cells) affects AQP2 trafficking.

Results : Short-term dDAVP treatment in M-1 cells stimulated phosphorylation of Akt (S473) and AS160. Conversely, the PI3K inhibitor LY 294002 diminished phosphorylation of Akt (S473) and AS160. siRNA-mediated Akt1 knockdown was associated with unchanged total AS160 but decreased phospho-AS160 expression, indicating that phosphorylation of AS160 is mediated by PI3K/Akt pathways. Moreover, siRNA-mediated AS160 knockdown significantly decreased total AS160 and phospho-AS160 expression. Semiquantitative immunocytochemistry revealed that AS160 knockdown was associated with significantly increased AQP2 expression of the plasma membrane (123±2%, $p<0.05$, $n=52$) despite the absence of dDAVP stimulation, compared with the control M-1 cells transfected with non-targeting siRNA.

Conclusions : Taken together, PI3K/Akt pathways mediate AS160 phosphorylation, which is likely to play a role as a negative regulator for AQP2 trafficking. Since phosphorylation of AS160 is known to inhibit its GAP activity leading to an increase in the active GTP-bound form of the AS160 target Rab proteins, this supports that vasopressin-stimulated phosphorylation of AS160 plays a role in the AQP2 trafficking.

Key Words : 수분통로, 집합관, 항이뇨호르몬
Aquaporin, Collecting duct, Vasopressin