고포도당으로 자극한 족세포 및 당뇨병성 사구체에서 알도스테론이 세포사멸에 미치는 영향

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Aldosterone Induces Apoptosis in High Glucose-stimulated Podocytes and in Diabetic Glomeruli

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Background : Even though aldosterone induces apoptosis in cardiac myocytes and skeletal muscle cells, and apoptosis is implicated as a potential mechanism of podocyte loss characterized in diabetic nephropathy, the effect of aldosterone on podocyte apoptosis has not been explored. Purpose: This study was performed to investigate whether local aldosterone system within podocyte is involved in the development of apoptosis in podocytes under diabetic conditions.

Methods : In vivo, 32 Sprague- Dawley rats were injected either with diluent (n=16, C) or with STZ intraperitoneally (n=16, DM) and 8 rats from each group were treated with spironolactone (SPR, 20 mg/kg/day) for 6 weeks. In vitro, immortalized mouse podocytes were exposed to media containing 5.6 mM glucose (NG), NG+24.4 mM mannitol, NG+10- 8M (SPR), NG+10- 8M aldosterone, 30 mM glucose (HG), or HG+10- 8M SPR for 24 hours. At the time of sacrifice, 24- hour urinary albumin excretion (UAE) was determined by ELISA. For the assessment of apoptosis, Western blotting for Bax, Bcl- 2, and active caspase- 3 were performed with sieved glomeruli and cell lysates. TUNEL assay and Hoechst 33258 staining and were also performed with renal tissue and cultured podocytes, respectively.

Results : UAE was significantly higher in DM (1.89+0.35 mg/day) compared to C rats (0.41+0.05 mg/day) ($p<0.05$) and this increase in UAE in DM rats was inhibited by SPR treatment (1.08+0.22 mg/day) ($p<0.05$). The ratios of Bax/Bcl- 2 protein expression were significantly increased in diabetic glomeruli and in podocytes exposed to HG by 124.0% and 113.0%, respectively ($p<0.05$), and these increases were ameliorated by the administration of SPR ($p<0.05$). Western blot for active caspase- 3 showed a similar pattern to the ratio of Bax/Bcl- 2 protein expression. TUNEL positive- stained nuclei were also significantly increased in DM (2.7+1.5) relative to C glomeruli (0.6+0.4) ($p<0.05$), which were inhibited by SPR (1.3+0.8) ($p<0.05$). In addition, apoptotic cells were significantly increased in HG compared to NG cells, and this increase was abrogated with SPR pretreatment. On the other hand, aldosterone significantly increased the ratio of Bax/Bcl- 2 and active caspase- 3 protein expression and induced apoptosis in cultured podocytes.

Conclusion : Local aldosterone system seems to be involved in podocyte apoptosis and aldosterone blockade may be beneficial in preventing podocyte loss under diabetic conditions.

Key Words : 고포도당, 족세포, 알도스테론
High glucose, Podocytes, Aldosterone