Sodium Glucose Co-Transporter 2 Inhibitor

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Obesity is supposed to cause renal injury via autophagy deficiency. Recently, sodium glucose co-transporter 2 inhibitors (SGLT2i) were reported to protect renal injury. However, the mechanisms of SGLT2i for renal protection are unclear. Here, we investigated the effect of SGLT2i for autophagy in renal proximal tubular cells (PTCs) on obesity mice. We fed C57BL/6J mice with a normal diet (ND) or high-fat and -sugar diet (HFSD) for nine weeks, then administered SGLT2i, empagliflozin, or control compound for one week. Each group contained N = 5. The urinary N-acetyl-beta-d-glucosaminidase level in the HFSD group significantly increased compared to ND group. The tubular damage was suppressed in the SGLT2i–HFSD group. In electron microscopic analysis, multi lamellar bodies that increased in autophagy deficiency were increased in PTCs in the HFSD group but significantly suppressed in the SGLT2i group. The autophagosomes of damaged mitochondria in PTCs in the HFSD group frequently appeared in the SGLT2i group. p62 accumulations in PTCs were significantly increased in HFSD group but significantly suppressed by SGLT2i. In addition, the mammalian target of rapamycin was activated in the HFSD group but significantly suppressed in SGLT2i group. These data suggest that SGLT2i has renal protective effects against obesity via improving autophagy flux impairment in PTCs on a HFSD.

sodium glucose co-transporter 2 inhibitor | mammalian target of rapamycin (mTOR) | autophagy | obesity

1. Introduction

Much attention was focused on the associations between obesity/metabolic syndrome (MetS) and kidney injury recently. In large-cohort studies, high body mass index was shown to be a risk factor for end-stage renal diseases independently compared to hypertension or diabetes [1]. Many researches were conducted on the mechanisms of kidney injury by obesity/MetS. In addition, autophagy deficiency is reported to be one of the renal injury causes [2]. Autophagy contributes to cellular homeostasis by promoting degradation and recycling of intracellular defective macromolecules and organelles, while autophagy deficiency leads to cellular death. Autophagy deficiency is observed in renal proximal tubular cells (PTCs) of obesity patients [2]. In addition, autophagy flux impairment by lysosomal dysfunction, inflammasome activation, and macrophage invasion was observed in PTCs of obesity mice [3]. Thus, autophagy deficiency is believed to be involved in kidney injury by obesity/MetS.

Recently, sodium glucose co-transporter 2 inhibitor (SGLT2i), belonging to a class of medications used for the treatment of type II diabetes mellitus via inhibiting reabsorption of glucose in PTCs and, therefore, lowering blood sugar, was reported to have renal protective effects in chronic kidney disease patients [4]. SGLT2i was reported to

ameliorate glomerular hyperfiltration by normalization of tubuloglomerular feedback [5], as well as to improve the mitochondrial abnormalities in PTCs and to reduce the oxidative stress [6][7]. Effects of SGLT2i with respect to autophagy in the kidney were also researched; however, the results are controversial [8][9]. In this study, we investigated the therapeutic effect of SGLT2i on autophagy deficiency in PTCs of obesity mice.

2. Discussion

Several renal protective mechanisms of SGLT2i were reported; however, the effect of SGLT2i on autophagy was not elucidated. In this study, we found that SGLT2i has a therapeutic effect on autophagy flux impairment in PTCs on obesity mice. These findings might contribute to elucidating the renal protective mechanism of SGLT2i in obesity patients.

We focused on lysosomal functions as a target of SGLT2i for improving autophagy flux impairment in PTCs. Lysosomes fuse with autophagosomes to form autolysosomes, and they play an important role in degradation process of autophagy. In obesity, lysosomal hydrolase is inactivated by the pH increase caused by lipid accumulation in the lysosome, resulting in impairment of lysosomal capacity and autophagy flux [3]. Autolysosomes, the degradation capacity of which is impaired by lipid accumulation, are reported to form MLBs in obesity mice [3]. Our findings, i.e., the decrease of autolysosomes and the increase of mitophagosomes after SGLT2i administration, suggest the therapeutic effect of SGLT2i on lysosomal dysfunction and autophagy flux impairment in PTCs. We considered that these effects are due to the suppression of the activated mTOR as the result of the PTC's decreased exposure of glucose. Activated mTOR forms mTORC1 and inhibits the transcription of lysosomal enzymes [10], as well as increases lipid deposition in lysosome, resulting in lysosomal dysfunction [3][11][12]. In this study, SGLT2i increased the amount of urinary glucose—thus decreasing the reabsorption of glucose into PTCs—and decreased the amount of serum glucose, suggesting the PTC's decreased exposure of glucose, both in apical and basal sides. Moreover, we demonstrated that SGLT2i suppressed mTOR activation in obesity mice. Furthermore, our in vitro study showed that SGLT2i significantly improved autophagy flux of PTCs exposed to high glucose. These data suggest that the decrease of glucose influx into PTCs by SGLT2i could directly affect mTOR activation in PTCs.

Our study has at least two limitations. One is that we could not definitively distinguish SGLT2i's effect for glucose and lipid metabolism in protecting PTCs; SGLT2i might have protected PTCs by also improving the lipid metabolism, resulting in amelioration of lipotoxicity in PTCs. In our in vivo study, SGLT2i administration at least showed no significant difference in bodyweight in obesity mice, indicating no significant difference in body fat. Furthermore, our in vitro study suggests that PTC's protective effect of SGLT2i by inhibiting glucose absorption is independent of lipid exposure. The other limitation is that SGLT2i might have effects on autophagy of PTCs via other mechanisms. SGLT2i is reported to have effects on various factors related to autophagy, such as serum insulin, insulin-like growth factor-1, and sirtuin 1 [13][14][15]. Further studies are needed to elucidate more deeply how SGLT2i affects the autophagy pathway in PTCs.

It is important to note that SGLT2i showed protective effects of PTCs in obesity mice showing no significant increase in serum glucose compared to control mice in this study. Similarly, there is a report which showed SGLT2i's protective effect of PTC's mitochondria of obesity mice, which showed no significant elevation of serum glucose [7]. These findings indicate that over-intake of sugar might cause glucotoxicity of PTCs in obesity mice even if there is no increase of serum glucose, and urinary glucose might have an important role in PTC's glucotoxicity. Unlike most types of cells which take in glucose only for their energy consumption, PTCs must reabsorb much urinary glucose for the whole body's survival and, thus, might be under stronger exposure than other types of cells. In fact, reabsorption of urinary glucose in proximal tubules plays an important role in the progression of tubular injuries in diabetic nephropathy [16]. Further studies are needed to elucidate deeply whether urinary glucose reabsorption could worsen PTC's injuries in obesity.

There is also a report which concluded that SGLT2i has no sufficient effect on autophagy in PTCs. Tanaka et al. reported that autophagy insufficiency in the tubular cells of diabetic mice was ameliorated by food-intake restriction, but not by SGLT2i [9]. They reported that the results were because the effect of SGLT2i on calorie balance is weaker than that of food-intake restriction; in their study, the amount of food intake and the bodyweight increase of the SGLT2i-treated group were greater than those in the control group. This might be because of an appetite increase via the glucose-lowering effect of SGLT2i. The change in the bodyweight by SGLT2i administration in their study was contrary to that in clinical trials, where SGLT2i significantly decreased the bodyweight in humans [17]. These reports suggest that free feeding of mice could mask the effect of SGLT2i on autophagy, and that the adjustment of food intake is needed to accurately evaluate the autophagy activity in in vivo experiments.

In conclusion, our study suggests that SGLT2i might ameliorate proximal tubular injuries in obesity via improving autophagy flux impairment.

References

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