以 AMPK 活化爲分子標靶的第二型糖尿病治療藥之研發

Development of therapeutic agents for type II DM by using AMPK as molecular target

中文摘要

AMP-activated protein kinase (AMPK)是一個感應細胞內能量代謝的一個關鍵的調控者，細胞藉著 AMPK 的活化而減少內臟脂肪及膽固醇的合成，並且抑制肝臟糖質新生，因此 AMPK 已被廣泛的認爲是治療第二型糖尿病的分子標靶。爲了找到一個可治療第二型糖尿病的潛力藥物，我們篩選了八十四種由中草藥分離出來的純化物。我們發現了一個編號爲 TMU023 的純化物，可以刺激 NRK52E 細胞中 AMPK 的磷酸化，活化的 AMPK 也可以增加其下游受質 acetyl CoA carboxylase (ACC)的磷酸化和 NRK52E 細胞的脂肪酸 b-氧化作用。以 PKA 抑制劑(H-89)處理細胞後，可以有效的抑制 TMU023 所誘發的 AMPK 磷酸化、ACC 磷酸化及 b-氧化作用，TMU023 活化 AMPK 可能是透過 PKA 的訊息傳遞路徑。另外，由於高度醣化最終產物(Advanced glycosylation end products, 簡稱 AGE)已知和各種糖尿病併發症的發生關係密切，所以我們發展了一項高通量篩檢法，進行糖尿病併發症治療藥物的研發，發現 TMU023 對過度糖化最終產物(AGE)的形成有抑制的作用。總而言之，我們的結果證實了，TMU023 除了是 AMPK 的活化劑外，在體外的試驗中也可以有效的抑制 AGE 的形成，因此 TMU023 可能具當作治療第二型糖尿病的潛力。

英文摘要

AMPK activated protein kinase (AMPK) is a key sensor and regulator of intracellular and whole body energy metabolism. Activation of AMPK has been shown to reduce visceral fat content, cholesterol synthesis and increase hepatic glucose disposal. Thus, AMPK has been considered as a molecular target for type II DM. In order to develop a potential drug for type II DM, we have screened 84 compounds isolated form Chinese herbal medicine for AMPK activator. We found that compound TMU023 stimulated a dose dependent AMPK phosphorylation in NRK52E cells. The activation of AMPK is associated with increased phosphorylation of its downstream substrate, acetyl CoA carboxylase (ACC), and increased fatty acid b-oxidation in NRK52E cells. Treatment of H89, a pharmacological inhibitor specific for PKA, inhibited TMU023-activated AMPK phosphorylation, ACC phosphorylation, and fatty acid b-oxidation, suggesting TMU023 mediated AMPK activation through PKA dependent signaling pathway. Advanced glycosylation end products (AGE) have been linked to the pathogenesis of diabetic complications. To explore whether TMU023 exerts other beneficial effects to attenuate AGE formation, a high throughput screening assay

using Amadori products as substrate has been developed. We demonstrated that in addition to AMPK activation, TMU023 was able to suppress AGE formation in vitro. Taken together, our results suggest that TMU023 may serve as a potential therapeutic agent for type II DM.