Resveratrol 抑制血小板凝集作用之探討

Mechanisms Involved in the Antiplatelet Activity of Resveratrol

中文摘要

白藜蘆醇 (resveratrol) 是一種植物衍生的非生物黃酮多酚類二苯乙烯 (nonflavonoid polyphenol stilbene)，通常存在於蔬菜和水果裡，特別是葡萄的子跟葡萄皮。此一物質已被證實具有抗癌症，抗發炎，清除自由基以及預防心血管疾病的功能。本研究在於更深入討論 resveratrol 在血小板活化的過程中，對於訊息傳遞的抑制機轉。研究結果顯示 resveratrol 隨者濃度增加 (0.02-45 M)，能有效抑制 collagen (1 g/ml)、U46619 (1 M) 以及 arachidonic acid (AA) (60 M) 所引起的人類血小板凝集反應以及 ATP 釋放反應。Resveratrol (0.15 與 0.25 M) 亦可抑制 collagen (1 g/ml) 所引起的細胞內鈣離子移動、 phosphoinositide 減少以及 thromboxane A2 (TxA2) 的合成。此外，resveratrol (0.15 與 0.25 M) 可以增加人類血小板內 nitrite 與 cyclic GMP 的含量，但對於 cyclic AMP 的含量並沒有顯著增加；另一方面 resveratrol (0.15 與 0.25 M) 可以有效清除 collagen (1 g/ml) 刺激產生的 hydroxyl radicals。此天然成分也抑制由 collagen (10 g/ml) 所刺激的 p38 MAPK 磷酸化，但是對於 extracellular signal regulated kinases (ERKs) 和 c-Jun-NH2-terminal kinases (JNKs) 以及 Akt 的磷酸化不具影響。Resveratrol (0.15 與 0.25 M) 可有效抑制 PDBu (150 nM) 和 collagen (1 g/ml) 所刺激 47 kDa protein 磷酸化。

由結果證實，resveratrol 抑制血小板活性的作用可能牽涉下列路徑：(一) Resveratrol 會刺激 NO 的生成，接著增加血小板細胞內 cyclic GMP 的含量，並且誘發 VASP 磷酸化、抑制 phospholipase C 的活性。(二) Resveratrol 利用其清除 hydroxyl radicals 的作用以及抑制 p38 MAPK 磷酸化，影響 phospholipase A2-cyclooxygenase 路徑的反應，進一步抑制 TxA2 的生合成。綜合以上結果，導致 resveratrol抑制血小板細胞內鈣離子的移動以及濃度的增加，最後因而抑制血小板的凝集反應。此項作用意味著 resveratrol 可有效地應用在治療與血小板過度活化相關之疾病。

英文摘要

Resveratrol is an edible nonflavonoid polyphenolic stilbene, that is produced by several plants, berries and fruits, especially grapes. This polyphenolic stilbene is reported to have anti-cancer, anti-inflammatory, free radical scavenging, antioxidant, and prevent cardiac vascular disease properties. However, the pharmacological functions of resveratrol on platelets were not yet understood, we are interested in investigating the inhibitory effect of resveratrol of cellular signal transduction in platelet activation. In this study, resveratrol concentration dependently (0.02-45 M) inhibited collagen (1 g/ml)-, U46619 (1 M)-, and arachidonic acid (AA) (60 M)- induced human platelets aggregation and ATP-release reaction. In addition, resveratrol (0.15 and 0.25 M) markedly inhibited intracellular Ca2+ mobilization in Fura-2 AM-loaded platelets, phosphoinositide breakdown and thromboxane A2 formation stimulated by collagen (1 g/ml). Furthermore, resveratrol (0.15 and 0.25 M) significantly increased the formations of nitrate and cyclic GMP but not cyclic AMP in human platelets. Moreover, resveratrol (0.15 and 0.25 M) obviously scavenged collagen (1 g/ml)-induced hydroxyl radicals and significantly inhibited p38 MAPK phosphorylation in human platelets, but not significantly inhibited ERK、JNK and Akt phosphorylation in human platelet. Rapid phosphorylation of a protein of Mr. 47,000 (P47), a marker of protein kinase C activation, was triggered by PDBu (150 nM) or collagen (1 g/ml). This phosphorylation was inhibited by resveratrol (0.15 and 0.25 M) in human platelets. In conclusion, our study suggested that the possible pathways of anti-platelet aggregation of resveratrol may involve in the following pathway: (1) Resveratrol stimulated nitrite formation, followed by increasing amount of cyclic GMP formation and inhibited phospholipase C activity. (2) Resveratrol significantly inhibited thromboxane A2 formation through inhibition of phospholipase A2-cyclooxygenase pathway and following resulted in inhibition of intracellular Ca2+ mobilization、p38 MAPK phosphorylation and scavenging the hydroxyl radicals. Taken together, resveratrol may be used as an effective tool in treating pathological disorder associated with platelet hyperaggregability.