α-Naphthoflavone 抑制血小板凝集作用之機轉探討

Mechanisms involved in the antiplatelet activity of α-naphthoflavone

中文摘要

從植物 Acroptilon repens (Russian knapweed) 根部的分泌物發現具有毒害植物的作用，而這個分泌物被認定是 7,8-benzoflavone (α-naphthoflavone)。Alpha-naphthoflavone (α-NF) 已經被發現經由誘導細胞外的鈣離子流入和內皮細胞的 NO 生成，而影響血管舒張。然而，正確的機轉仍然不清楚，需要更進一步的確定它的特性。血小板在心血管系統中扮演重要的角色，因此，本實驗想進一步探討 α-naphthoflavone 是否能夠有效地抑制血小板凝集。在這個研究中，實驗發現在經由 collagen (1 μg/ml)、ADP (20 μM) 或 AA (60 μM) 刺激血小板之後，再加入 α-naphthoflavone，其濃度爲 5-20 μM，以 collagen 作用下較顯著的抑制血小板的凝集。α-naphthoflavone 抑制 collagen 誘發血小板凝集的 IC50 濃度約 5 μM。除此之外，α-naphthoflavone 可以顯著地抑制 collagen 刺激之血小板內的鈣離子移動、phosphoinositide 減少和 thromboxane A2 生成。更進一步，α-naphthoflavone 會增加人類血小板中 cyclic GMP 的生成，但並不會影響 cyclic AMP 的生成。總而言之，依實驗結果推測α-naphthoflavone 的機轉可能和以下有關：(1) 它會抑制 phospholipase C (PLC) 的活性，透過影響膜的流動性，接著會抑制 47 kDa 蛋白質的磷酸化和細胞內鈣離子的移動。(2) α-naphthoflavone 會影響 phospholipase A2 (PLA2) 的活性，導致 thromboxane A2 生成減少。(3) α-naphthoflavone 可能會經由活化血小板內的 guanylate cyclase，使 cGMP 的含量增加。

英文摘要

Root exudates from Acroptilon repens (Russian knapweed) were found to be phytotoxic and the phytotoxin in the exudate was identified as 7,8-benzoflavone (α-naphthoflavone). Alpha-naphthoflavone (α-NF) has been found to induce vasorelaxation through induction of extracellular calcium influx and NO formation in endothelium. However, the exact mechanism is still unclear and requires further characterization. Platelet plays an important role in the cardiovascular system, therefore, we want to further know whether α-NF can effectively inhibit platelet aggregation. In this study, we found that α-NF (5-20 μM) significantly inhibited platelet aggregation stimulated by collagen (1 μg/ml), ADP (20 μM) and AA (60 μM) in washed human platelets. The IC50 of α-NF at inhibition of platelet aggregation were about 5 μM stimulated by agonists. In addition, α-NF significantly inhibited intracellular Ca2+ mobilization, phosphoinositide breakdown and thromboxane A2 formation. Moreover, α-NF increased the cGMP level, but not increased cyclic AMP formation in washed human platelets. In conclusion, we suggest that the mechanisms of a-NF may be involved : (1) it may inhibit phospholipase C (PLC) activity via influence of membrane fluidity, followed by the inhibition of 47 kDa protein phosphorylation and intracellular calcium mobilization. (2) α-naphthoflavone affected phospholipase A2 (PLA2) activity leading to reduce thromboxane A2 formation. (3) a-naphthoflavone may activate guanylate cyclase, and increase cGMP levels.