探討大豆異黃酮素對於老鼠造骨細胞增生與骨蛋白表現的影響

The Effects of Soy Isoflavones (Genistein and Daidzein) on MC3T3-E1 Cells Proliferation and Bone Protein Expression

中文摘要

本研究針對大豆異黃酮素(Genistein、Daidzein)對於老鼠造骨細胞株(MC3T3-E1 cells)增生之影響機制進行探討。由細胞增生實驗結果(MTS assay)發現投與 10-9M~10-5M 濃度的異黃酮素，培養 24 小時，與控制組比較，可顯著刺激 MC3T3-E1 細胞的增生($p<0.05$)；培養 24~96 小時，計算細胞增生數目，發現 10-9M~10-6M 均可刺激細胞增生。故由上述結果，選擇投予 10-9M~10-5M 之大豆異黃酮素，培養 48 小時，作爲後續實驗的投予濃度及培養時間點。投與 10-9M~10-5M 之大豆異黃酮素，抽取細胞內的 DNA 進行測試，發現可顯著增加 MC3T3-E1 細胞內 DNA 的量，以 10-8M 與 10-7M 兩組所增加細胞內 DNA 的效果最佳，此符合 MTS assay 所得到的結果。當異黃酮素之添加濃度爲 10-9M~10-6M，可顯著刺激 MC3T3-E1 細胞內鹼性磷酸酶(ALP)的酵素活性，尤以 10-7M 組刺激 MC3T3-E1 細胞內 ALP 的活性最佳。投與大豆異黃酮素，抽取細胞內的蛋白質，以西方墨漬法進行骨蛋白(Osteopontin, OPN、第一型膠原蛋白)表現量的測定，發現投予 10-9M~10-7M 三組，OPN、第一型膠原蛋白的表現量較高。投予造骨細胞株異黃酮素，抽取細胞內的 RNA，給予 OPN primers，進行 RT-PCR 的測試，發現 OPN mRNA 在各組的表現量並無顯著的差異。故本研究之結論爲，大豆異黃酮素可刺激老鼠造骨細胞株之細胞存活率與增生情形，提高 DNA 的含量與 ALP 的酵素活性，且可增加 MC3T3-E1 細胞內骨蛋白(Osteopontin、Type I collagen)的表

英文摘要

The proliferation effect of isoflavones(genistein and daidzein) were investigated using an MTS assay and DNA content on MC3T3-E1 cells. The samples were tested at levels of 10-9 to 10-5M of isoflavones for 24, 48, 72 and 96 hr incubation. The results obtained from the two assys were consisted and also exhibited dose-dependent effects except 10-5M. At the level of 10-8 and 10-7M, isoflavones had the best result for cell proliferation. Genistein and daidzein stimulated an increase in alkaline phosphatase(ALP) activity on MC3T3-E1 cells in a dose-dependent manner(10-9 ~10-7M). Isoflavones treatment(10-9 ~10-7M) significantly increase the production of osteopontin(OPN) and Type I collagen protein, but not on OPN mRNA expression. In summary: isoflavones at dietary achievable level(10-9 ~10-7M) stimulated the proliferation and DNA content of MC3T3-E1 cells. Isoflavones also increased bone marker synthesis(ALP, OPN and Type I collagen) and could be beneficial for bone

growth.