Ketoconazole potentiates the antitumor effects of nocodazole: in vivo therapy of human tumor xenografts in nude mice

林建煌

Wang YJ;Jeng JH;Chen RJ;Chen LC;Liang YC;Lin CH;Chen CH;Chu JS;Ho WL and Ho YS

Abstract

Our previous studies demonstrated that the oral antifungal agent ketoconazole (KT) induces apoptosis and G0/G1 phase cell cycle arrest in human cancer cell lines. In this study, we first demonstrated that KT (1 mM) potentiated the apoptotic effects of nocodazole (ND, 1 nM) in COLO 205 cancer cells. We further demonstrated the therapeutic efficacy of a combined treatment of KT (50 mg/kg/three times per week) and ND (5 mg/kg/three times per week) in vivo by treating athymic mice bearing COLO 205 tumor xenografts. The antitumor effects of ND were significantly potentiated by KT in mice after 6 wk of treatment. No gross signs of toxicity were observed in mice receiving these treatment regimens. The apoptotic cells were detected in a microscopic view of the terminal deoxynucleotidyl transferase-mediated dUTP nick-end labeling staining and by observation of DNA fragmentation in KTþND-treated tumor tissues. The levels of cell cycle regulatory proteins were determined by Western blot analysis. Treatment with KT inhibits tumor growth through elevation of p53, p21/CIP1, and p27/KIP1 as well as inhibition of cyclin D3 and cyclin-dependent kinase 4 protein expression. Immunohistochemical staining analysis showed that p53, p21/CIP1, and p27/KIP1 immunoreactivity were induced in the tumor tissues. To clarify the roles of the p21/CIP1 and p27/KIP1 protein expression involved in G0/G1 arrest and/or apoptosis induced by a combined treatment with KT and ND, antisense oligodeoxynucleotides (ODNs) specific to p21/CIP1 and p27/KIP1 were used. Our results demonstrated that apoptotic phenomena, including BAX induction and cytochrome C released from mitochondria induced by KTþND, were significantly attenuated by pretreatment the cells with the p27/KIP1-specific antisense ODNs. These results indicate that p27/KIP1 protein does indeed play a critical role in the KTþND-induced apoptosis. Our study revealed the molecular mechanism of KTþND in regression of the tumor growth. The apoptotic effects of KT in a great variety of cancer cells make it a very attractive agent for cancer chemotherapy.