長期酒精曝露模式探討肝癌細胞之生長週期與化療藥物之抗藥性研究

Use of long term ethanol treatment to study the ethanol effect on cell cycle and chemosensitivity in hepatocellular carcinoma cell.

中文摘要

Hepatitis B Virus (HBV)是亞洲常見的病毒性肝炎。長期酒精處理會導致肝細胞不正常增生，因此我們利用基因已被嵌入 HBV genome，且持續表現 HBsAg 的肝癌細胞株 Hep3B，在長期的酒精處理下，推測長期飲酒對肝癌病患及同時感染 B 肝病毒對細胞週期與抗藥性的影響。利用 MTT 技術與計數細胞的結果皆指出長期酒精培養會加速細胞生長。因此酒精可能改變及影響細胞的細胞週期與週期相關蛋白。結果顯示 cyclin D1、cyclin D3 是增加的。這與上述酒精增加細胞生長，有可能因爲 cyclin D1、D3 蛋白增加，促進細胞往 S 期進行所產生的結果。進一步利用流式細胞儀分析酒精對細胞週期的影響，結果顯示酒精的作用明顯促進細胞週期的進行，且與 G0/G1 期的 cyclin D1 與 cyclin D3 蛋白變化有一致性。並進一步探討酒精是否增加或減少細胞對化療藥的敏感性。幾種臨床上常見的肝癌藥包含 Cisplatin、Doxorubicin、Paclitaxel、Mitomycin C、5-Fluorouracil，觀察細胞在酒精處理下對各種化療藥的反應。結果顯示在這些化療藥中，酒精在 48 小時、2 μg/ml 的 Cisplatin 或 10 μM 的 Mitomycin C 的處理下明顯增加細胞的耐受性。相反地，酒精增加細胞對 Paclitaxel、Doxorubicin 的敏感性。在合併 Cisplatin 與 Mitomycin C，更增加酒精組細胞耐受性，但對細胞凋亡蛋白並未有顯著差異。此研究提供醫師在酗酒會影響治療肝癌同時感染 B 型肝炎病毒病人在化療治療效果的一些資訊。

英文摘要

Hepatitis B virus infection is common in south Asia and causes the hepatitis in human. Alcoholism causes the abnormal proliferation of hepatocytes, especially in chronic hepatitis patient or carrier. In order to investigate the effect of ethanol on the cell cycle and chemosensitivity of HBV-infected cells under the condition of the long-term ethanol treatment, the Hep3B cells were chosen as a study model, which are integrated with full length HBV genome and constitutively express HBsAg. By using the MTT technique and direct counting cell number, results appeared that long-term ethanol culture enhanced the cell growth. Therefore, ethanol might alter the cell cycle and cycle-related proteins. Under the treatment of ethanol, cyclin D1 and cyclin D3 were increased resulting in cells toward S phase. By using the flow cytometric technique, ethanol facilitated the progression of cell cycle, which is correlated to the

alteration of cyclin D1 and cyclin D3. Because most chemodrugs were effective on the cell division, the sensitivity of chemodrugs of ethanol-treated Hep3B cells was also investigated. Clinically, cisplatin, doxorubicin, mitomycin C, paclitaxel and 5-fluorouracil were used for the treatment of liver cancer. Data appeared that ethanol increased the cells tolerance to cisplatin , mitomycin C ,or combination when cells were treated with 2 μg/ml cisplatin or 10μM Mitomycin C for 48 hours. But they had no differents on cell apoptosis protein. In constrast with paclitaxel or doxorubicin, ethanol could increase Hep3B cells the sensitivity to paclitaxel or doxorubicin. In conclusion, this study provided physicians the information that HBV-infected liver cancer patients could be influenced by alcoholism in chemotherapy.