抽菸,喝酒,砷暴露,第 I、II 相代謝酵素的基因多形性與泌尿道上皮癌之相關性研究

Cigarette smoking, alcohol consumption, arsenic exposure, genetic polymorphisms of phase I、II metabolized-enzymes and urothelial carcinoma

中文摘要

研究背景：抽菸、無機砷及危險職業暴露是泌尿道上皮癌已知的危險因子。近來有研究指出喝酒可能與膀胱癌有關，但可能的機制尚未被詳細探討。人體接觸到環境致癌物質之後，通常先由第 I 相代謝酵素[例如細胞色素 P450 酵素, cytochrome P-450 (CYPs)]將有毒化學物質經代謝活化後形成親電子性物質，再由第 II 相代謝酵素[例如麩胺基硫轉移酵素, Glutathione S-transferases (GSTs)或 sulfotransferase (SULT)]將活性中間產物代謝成毒性較低的親水性物質排出體外。若體內的第 I、II 相代謝酵素的功能失去平衡，長期累積之後會增加罹患癌症的風險。另外，與酒精代謝相關的酵素包括 CYP2E1、乙醇去氫酵素(alcohol dehydrohenase, ADH)及乙醛去氫酵素(aldehyde dehydrogenase, ALDH)，假若這些酒精代謝相關的酵素產生變異，可能會增加罹患癌症的風險。因此，本論文將抽菸,喝酒,砷暴露及第 I、II 相代謝酵素基因多形性與泌尿道上皮癌進行合併分析，藉以探討基因與基因及基因與環境危險因子間的交互作用對於罹患泌尿道上皮癌的影響。

材料與方法：本論文選取 540 位經病理診斷確定的泌尿道上皮癌(包括腎盂、輸尿管及膀胱部位的癌症)患者作爲病例組個案，另外選取 540 位與病例組個案的年齡頻率匹配且未曾罹患任何癌症的患者作爲對照組個案。基因多形性主要是以聚合酵素連鎖反應-限制片段長度多形性方法來判定。第 I 相代謝酵素包括 CYP1A1、CYP2A6 及 CYP2E1，第 II 相代謝酵素包括 GSTO1、GSTO2 及 SULT1A1，與酒精代謝相關的酵素包括 ADH2、ADH3 及 ALDH2。利用 goodness-of-fit 卡方檢定進行哈溫平衡(Hardy-Weinberg Equilibrium)檢定。以非條件式多變項邏輯斯迴歸估計危險對比值(odds ratios, ORs)及 95%信賴區間(confidence intervals, CIs)。而基因的單套型(haplotype)是採用 Haploview 3.2 分析軟體(http://sourceforge.net/projects/ haploview)來進行分析。

研究結果：本研究發現抽菸、無機砷及職業暴露與泌尿上皮癌有顯著相關。同時具有抽菸及喝酒習慣者有較高的危險性(OR=3.0)及顯著的協同作用，而經常使用消炎止痛藥者有 2 倍危險性。在各別的基因多形性分析中，帶有 CYP1A1 A4889G 的 G/G 基因型有顯著較高的危險性(OR=1.9)。CYP1A1 危險雙套型有 1.8 倍顯著的危險性。帶有 3 個或以上第 I 相代謝酵素的危險基因型/雙套型者有 2.5 倍的顯

著危險性。同時帶有抽菸/喝酒及職業暴露(≥1)、無機砷暴露(high)及危險基因型(≥3)者有 7.7 倍顯著的危險性。另外，帶有 GSTO2 A424G-G/G、GSTO2 A-183G-G/G 及 SULT1A1 G638A-G/G 基因型有顯著較高的危險性。GSTO1/2 的危險雙套型也有 1.8 倍顯著的危險性。帶 1 個或以上第 II 相代謝酵素的危險基因型/雙套型者有 2.3 倍的顯著危險性。同時帶有抽菸/喝酒及職業暴露(≥1)、無機砷暴露(high)及危險基因型(≥1)者有 6.8 倍顯著的危險性。將抽菸/喝酒、無機砷暴露及職業暴露合併分析後，結果發現帶有其中 1 個及帶有其中 2 個或以上環境暴露者分別有 1.5 及 2.6 倍顯著的危險性。最後，在帶有其中 2 個或以上環境危險因子暴露之下，同時帶有 3 個或以上第 I 相代謝酵素的危險基因型/雙套型及 1 個或以上第 II 相代謝酵素的危險基因型/雙套型者具有最顯著的危險性 (OR=19.4, 95% CI =4.5-83.1)。

結論：從本論文的研究結果綜合來看，環境危險因子的暴露及第 I、II 相代謝酵素的危險基因型/雙套型分別對於罹患泌尿道上皮癌有顯著的影響之外，尤其是當環境與基因之間具有協同作用時，影響的程度會更加明顯。因此，未來需要更大的樣本及納入其他具功能性的基因多形性一併分析，以期對泌尿道上皮癌的致病機轉有更完整的瞭解。

英文摘要

Background：Cigarette smoking and exposures of arsenic and risk-occupations are known risk factors for urothelial carcinoma (UC). Recent studies indicated that alcohol consumption is likely to be associated with bladder cancer, but the possible mechanism has not been well investigated. After human exposed to environmental carcinogens, most of these substances are firstly converted into reactive carcinogenic metabolites by phase I enzymes including Cytochrome P-450 (CYPs) and these reactive components were removed by phase II enzymes including Glutathione S-transferases (GSTs) or Sulfotransferase (SULT). The lone-term imbalance of enzyme activity between phase I and phase II enzymes will lead to the development of cancer. Besides, CYP2E1、alcohol dehydrohenase(ADH) and aldehyde dehydrogenase (ALDH) are known alcohol-related enzymes and higher frequencies of variations of these enzymes are also likely to lead to the development of cancer. Therefore, to investigate the joint effects of gene-gene and gene-environment interactions on risk of UC, this study included environmental exposures of cigarette smoking, alcohol consumption, arsenic and risk-occupations and genetic polymorphisms of phase I 、II enzymes in combined analyses.

Materials and Methods：A total of 540 pathologically-confirmed UC cases including cancers of renal pelvis, ureter and bladder were selected as UC cases group. A total of 540 cancer-free controls, frequency-matched on age, were recruited from individuals who admitted to the same hospitals with UC cases for a health examination. Genetic polymorphisms of were determined by polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP). Selected enzymes including phase I enzymes-CYP1A1、CYP2A6 and CYP2E1, phase II enzymes-GSTO1、GSTO2 and SULT1A1, and alcohol-metabolized enzymes-ADH2 、 ADH3 and ALDH2. A goodness-of-fit X2 test was performed to examine Hardy-Weinberg Equilibrium (HWE). We used a unconditional multivariate logistic regression to estimate odds ratios (ORs) and 95% confidence intervals (CIs). Haplotype analysis was calculated by Haploview 3.2 (http://sourceforge.net/projects/ haploview).

Results：Cigarette smoking, arsenic exposure and risk occupational exposure are significantly associated with UC risk. Study subjects with both of cigarette smoking and alcohol consumption have a significantly higher UC risk of 3.0. Individuals with frequent NSAID usage have a significantly higher UC risk of 2.0. In genotyping analysis, subjects with CYP1A1 A4889G G/G genotype have a significantly higher UC risk (OR=1.9). Those carrying risk diplotypes of CYP1A1 have a significantly higher UC risk of 1.8. Subjects who carried three or more risk genotypes/diplotypes of phase I enzymes have a significantly increased UC risk of 2.5. Individuals with cigarette smoking/alcohol consumption and occupational exposures (≥1), arsenic exposure (high), and risk genotypes/diplotypes of phase I enzymes (≥3) have a significantly increased UC risk (OR=7.7). Significantly increased UC risks of 1.6, 2.5 and 1.8 were found for subjects carrying GSTO2 A424G-G/G, GSTO2 A-183G -G/G and SULT1A1 G638A-G/G genotypes, respectively. Individuals with risk diplotypes of GSTO1/2 also have a significantly higher UC risk of 1.8. Those carrying one or more risk genotypes/diplotypes of phase II enzymes have a significantly increased UC risk (OR=2.3). Individuals who carried cigarette smoking/alcohol consumption and occupational exposures (≥1), arsenic exposure (high), and risk genotypes/diplotypes of phase II enzymes (≥1) have a significantly increased UC risk (OR=6.8). In a combination analysis of cigarette smoking/alcohol consumption, arsenic exposure and occupational exposure, significantly higher UC risks of 1.5 and 2.6 were found for subjects with one risk factor and those with two or more risk factors, respectively. Finally, subjects who exposed to two or more of environmental risk factors carrying three or more risk genotypes/diplotypes of phase I enzymes and one or more risk genotypes/diplotypes of phase II enzymes have the significantly highest risk of UC (OR=19.4, 95% CI =4.5-83.1).

Conclusion：In addition to significant effects from exposures of environmental risk factors and risk genotypes/diplotypes of phase I、II enzymes on UC risk, the effects on the development of UC will be more predominant especially under the existence of gene-environment interaction. Therefore, a larger sample size and other functional polymorphisms of candidate genes should be took into consideration to provide a

more comprehensive understanding of UC.