行政院國家科學委員會專題研究計畫成果報告

原發性與轉移性胃癌同源關係和基因變化的特徵分析(1/2)

計畫類別：■個別型計畫　□整合型計畫

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個別型計畫：計畫主持人：吳明賢醫師

整合型計畫：總計畫主持人：

子計畫主持人：

註：整合型計畫總報告與子計畫成果報告請分開編印各成一冊，彙整一起繳送國科會

處理方式：□可立即對外提供參考

■一年後可對外提供參考

□兩年後可對外提供參考

(必要時，本會得展延發表時限)

執行單位：台大醫學院一般醫學科

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中 文 摘 要

胃癌的發生為多步驟伴隨很多重要的調控生長基因的改變，雖然有關於胃癌進展的基因慢慢開始有一些眉目，但是到底有多少基因參與胃癌轉移？其發生頻率多少？其臨床及生物學上意義為何？等重要問題仍不清楚，而且目前也沒有直接比較原發性和轉移性胃癌間基因變化的報告。導致此領域資料不完整或付之闕如的主因包括效率不佳的顯微分離和非全基因體探討等技術問題，最近發展出來的比較基因雜交法(CGH)和雷射捕捉顯微切割系統(LCM)可以用來克服上述困難，進行對特殊細胞群體的基因變化分析，而且可與微衛星體及異質性喪失等資料互補。

為了克服研究胃癌轉移的困難，吾人在第一年的計畫裏首先建立 CGH 及 LCM 的技術。利用 CGH，成功地分析 53 例胃癌的基因變化(圖 1)。此初步的結果顯示原發性胃癌的一些基本染色體變化，可做為將來進一步比較的參考，而且吾人也發現不同組織型態和共同別胃癌有不同的染色體改變。另外，利用 LCM，則可清楚地將腫瘤細胞與非腫瘤細胞從組織切片分離(圖 2)。由於這些經 LCM 取得細胞的 DNA 量不足以直接進行 CGH，吾人採用 degenerate oligonucleotide primed-polymerase chain reaction (DOP-PCR)將原發腫瘤部(T)、轉移腫瘤部(M)和非腫瘤部(N)的 DNA 成功地放大(圖 3)，這些放大的 DNA 足以進行 CGH，而且不影響 CGH profiles。

利用第一年所成功發展出的 LCM/DOP-PCR 配合 CGH 成為一種有效分析原發性胃癌和轉移性胃癌基因變化的有效方法，吾人在第二年的計畫將進一步以上述方法探討第一年所收集不同轉移部位(淋巴結、肝臟等)胃癌和原發性胃癌基因變化的差異，以了解其同源關係並嘗試尋找有用的癌細胞轉移標記。

關鍵語：胃癌，比較基雜交法，雷射捕捉顯微切割系統，職合酵素反應

Abstract

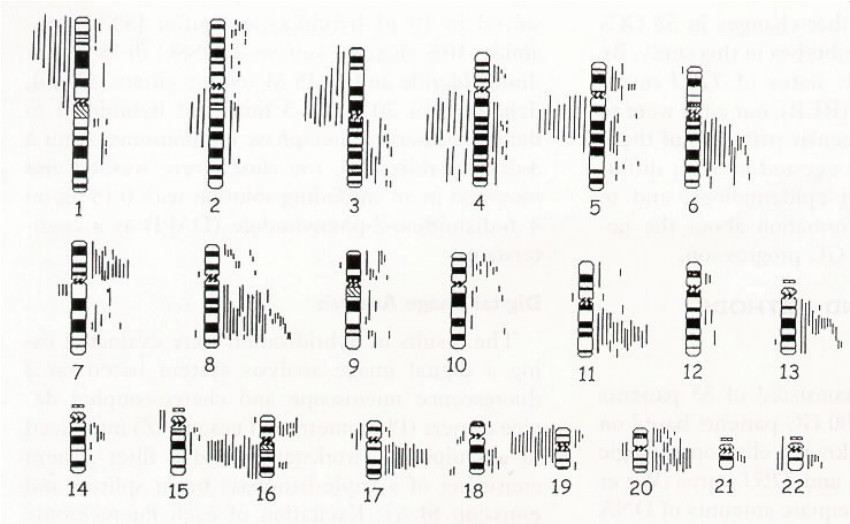
Gastrocarcinogenesis is consistent with the theory of invasive cancer arising from a series of histological abnormalities in the multistep process with accumulation of multiple alterations of critical growth-regulatory genes. Although genetic changes associated with progression that may be used for tumor characterization are beginning to be defined for GC, the range of genetic abnormalities that are involved in metastases, their frequency of occurrence, and their clinical and biological significance remain poorly understcod. Furthermore, there have been no published data directly comparing systemic genetic alterations in primary GC and their metastases. The reasons why previous studies in this field were incomplete and lacking are due to technical problems including inefficient microdissection and incomprehensive investigations of whole genome. To overcome these limitations, a comprehensive molecular cytogenetic technique called comparative genomic hybridization (CGH) and a powerful microdissection technique named laser capture microdissection (LCM) have recently been developed. In the first year grant period, we have successfully established the technique of CGH and utilized CGH to analyzed genetic alterations in 53 primary GC (figure 1). Our preliminary results have demonstrated genomewide chromosomal alterations in primary GC and such genetic profiles could provide a basis for further comparison with metastatic GC. These findings also revealed histology and stage-related chromosomal alterations. In addition, the application of LCM could precisely dissect tumorous and nontumorous components from histologic sections (figure 2). Since genomic DNA from LCM cells were not sufficient to perform CGH, we used degenerate oligonucleotide primed-polymerase chain reaction (DOP-PCR) to amplify DNA (figure 3). The amplified DNA then could be subjected to CGH and the analyses were accurate to reflect the genetic profiles of original cells.

With the aforementioned CGH, LCM, and DOP-PCR, we have established an effective method to analyze genetic aberrations in primary and metastatic GC. In the second year grant period, we will adopt these methods to analyze genomewide alterations of primary and metastatic GC

from different locations (e.g. lymph nodes, liver etc.)collected in the first year. Such efforts might elucidate the clonal relationship between primary and metastatic GC and disover useful predictive markers for GC.

Key Words: Gastric cancer, comparative genomic hybridization, laser capture microdissection, degenerative oligonucleotide primed-polymerase chain reaction

圖一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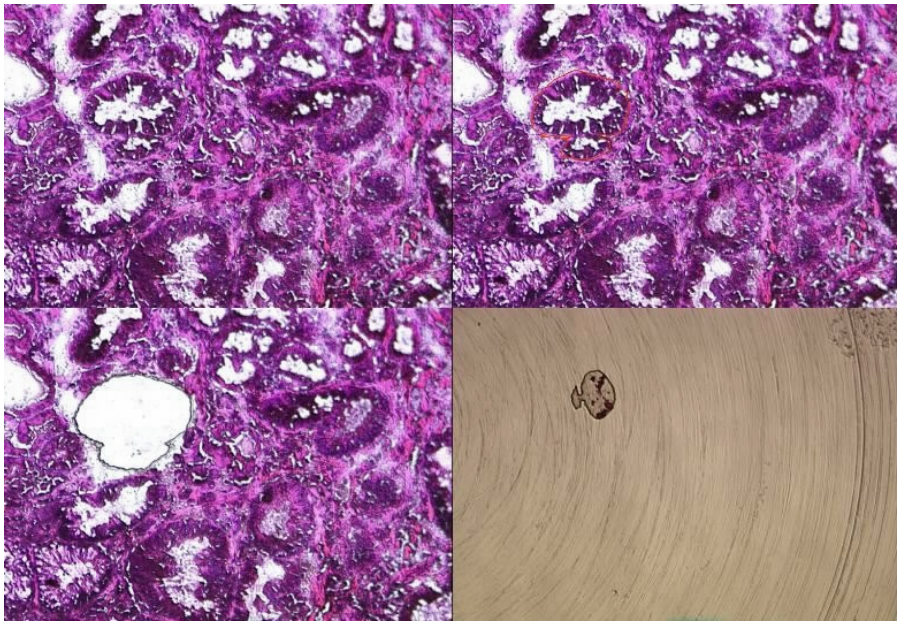
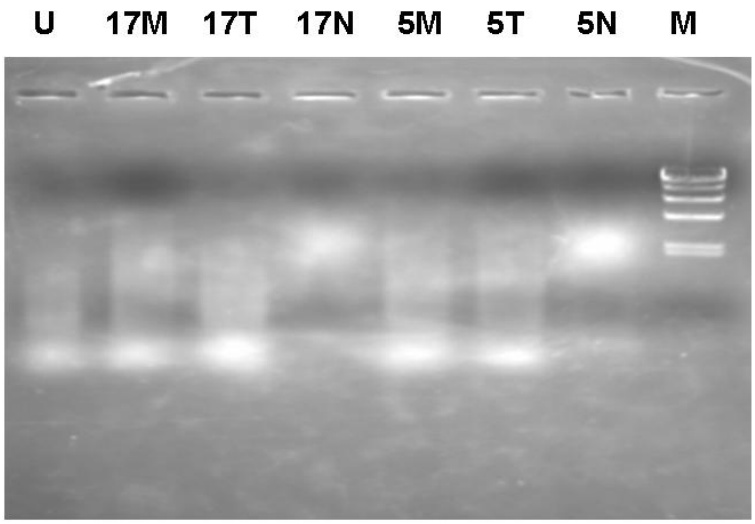
圖二



圖三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