國科會專題研究計畫成果報告

急性白血病人CD44變異型的表現及其臨床應用

CD44 variant isoforms in acute leukemia and its clinical implications

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主持人 ： 田蕙芬

執行機構： 台大醫學院內科

一、計畫中文摘要：

關鍵詞：CD44, 急性骨髓性白血病，急性淋巴芽細胞白血病

(CD44s)與淋巴細胞的歸鄉(homing)及正常造血(hemopoiesis)的調控有關。CD44s還有許　　CD44是一種黏著分子，在很多細胞包括造血細胞的表面都可以出現。典型的CD44多分子量較大的變異型(CD44v)，是由至少10個exons (v1到v10)選擇性裁剪(splicing)所造成。CD44v的表現被發現與細胞的惡性轉化及腫瘤的轉移有關。在非何金氏淋巴瘤中，CD44-v6主要表現於具侵略性的淋巴瘤，且與預後差有關。在多發性骨髓瘤中，CD44-v9的表現常出現在臨床表現及預後較差的一群病人。至於急性白血病CD44v的表現，除了最近的一篇摘要外，至今尚未有正式的論文發表。在這個計畫中，我們將對急性骨髓性白血病及急性淋巴芽細胞白血病，以免疫細胞化學染色的方法偵測白血病細胞上CD44s及各種CD44v (v3 to v10)的表現。這些結果將與病人的臨床表現、血液學變化、染色體異常及預後做一對照，找出在臨床上應用的可能性。

二、計畫英文摘要：

keywords: CD44, acute myeloid leukemia, acute lymphoblastic leukemia

CD44 is an adhesion molecule that is expressed on the surface of many cells including hemopoietic cells. The standard form of CD44 (CD44s) has been implicated in lymphocyte homing and in the regulation of normal hemopoiesis. There are many higher-molecular-weight variant isoforms (CD44v) generated by alternative splicing of 10 exons (v1 through v10) within the CD44 gene. Expression of particular isoforms of CD44 has been associated with malignant transformation and acquisition of metastatic potential. In non-Hodgkin's lymphomas, the expression of variant isoforms containing CD44-v6 was observed predominantly in aggressive lymphoma and was associated with poor prognosis. Overexpression of CD44-v9 was related to

unfavorable prognosis in multiple myeloma. No data concerning CD 44v expression in acute leukemia, except one abstract presented recently, have been published in literature. In this project, we would like to study the expression of various CD44v (v2 to v10), by immunocytochemical staining, in acute myeloid leukemia and acute lymphoblastic leukemia. Patients will be serially followed-up and reanalysed when relapse occurs. The results will be correlated with the clinical and hematological features, cytogenetic findings and prognosis of the patients. The clinical implications of CD44v expression will be determined.

三、研究計畫之背景、目的：

CD44 is an adhesion molecule which is widely expressed on the surface of many cells, including leukocytes and fibroblasts (1,2). It is implicated in lymphocyte homing and in the regulation of normal hemopoiesis (2-6). In long-term bone marrow cultures, formation of myeloid or lymphoid cells was completely suppressed by CD44 monoclonal antibodies at an early stage (5). Hyaluronan is the most widely recognized ligand of CD44 but other ligands such as fibronectin, collagen, and serglycin has also been reported (2,7,8).

CD44 is encoded by 20 exons: 10 of these code for standard form, CD44s. and the other exons contribute to multiple variant isoforms (CD44v) which are generated by alternative splicing of the RNA and insertion of the encoded variant regions in the extracellular domain of CD44(9,10). Expression of particular isoforms of CD44 has been associated with malignant transformation and acquisition of metastatic potential (11-16). Overexpressionof CD44-v6 in a non-metastasizing cell line of rat pancreatic carcinoma sufficed to establish full metastatic behavior (11). Upregulation of CD44v has also been demonstrated during tumor progression and disseminationin in humans, including gastric ca, colon ca and lymphoma (12-16).

In non-Hodgkin's lymphomas, the expression of variant isoforms (CD44v) containing CD44-v6 were observed predominantly in aggressive lymphoma and

were associated with a shorter overall survival of patients (17). In multiple myeloma, overexpression of v9-containing isoforms of CD44 was related to unfavorable clinical presentation and prognosis (18).

No data concerning the expression of variant isoforms of CD44 in acute leukemia have been published in literature, except one abstract presented in Annual Meeting of American Society of Hematology one month ago (19). A higher expression of CD44-v5, -v6, and v9 in early preB-acute lymphoblastic leukemia (ALL) and thymic T-ALL was found in that report. We plan to study various CD44v (v2 to v10) expression in AML and ALL patients and correlate the results with clinical manifestations and prognosis of patients.

四、研究結果：

（一）急性骨髓性白血病 (acute myeloid leukemia, 簡稱 AML):

我們對 73 例 AML 做了免疫細胞化學染色檢查，結果如下：

I. CD44 – V6 的表現

1. 73 例 AML 中，6 例病人有 CD44 – v6 的表現佔 8%，這 6 例病人中，除 1 例外，其他 5 例病人 CD44s 仍然有表現，且均有 90%以上的細胞為陽性，其他 CD44 – v6 陰性的病人，CD44s 均為陽性。
2. 分析 CD44 –v6 表現與國際 FAB 分類各亞型的關係（如下表），可見 M0 亞型所佔比例特別高：4 例中有 2 例為陽性。

FAB	病人數	CD44 – v6 陽性數(%)
M0	4	2(50%)
M2	26	1
M3	7	1
M4	9	2
Others (M1,M5, M6,M7)	29	0
Total	73	6 (8%)

3. 成人及孩童 AML 間的差別：
46 例成人 AML 中，4 例（9%）有 CD44 –v6 的表現，27 例孩童 AML 中，2 例(7%）有此變化

II. CD44 -v3, v4 -5,v9 在所檢驗的病人中，均無陽性者

(二）急性淋巴芽細胞白血病（ acute lymphoblastic leukemia,簡稱 ALL ）：
對 69 例 ALL 檢查的結果如下：

1. 69 例 ALL 中，2 例病人有 CD44 –v6 的表現，這 2 例病人 CD44s 陽性的細胞均小於 10%。另外有 12 例病人，雖然無 CD44 –v6 的表現，但 CD44s 陽性細胞比率減低，小於 50%，與其他病人大多數細胞為陽性不同，其中 7 例病人，CD44s 陽性細胞更小於 10%。

2. 分析 CD44 –v6 與 CD44s 表現與 ALL 免疫表型的關係，可見 B-lineage ALL，GrVI，也就是 B-ALL，CD44 –v6 陽性比率 最高(2/5 或 40%)，如下表：

Subtype	病人數	CD44 –v6 陽性者	CD44s 陽性細胞<50%
B – lineage			
Gr Ⅰ			
Ⅱ	12		
Ⅲ	22		6
Ⅳ	15		3
Ⅴ	6		1
Ⅵ	5	2	3*
T-lineage	7		
Unknow	2		1
Total	69	2	14*,**

*包括 2 例 CD44 –v6 陽性者
**其中 7 例，CD44s 陽性細胞 <10%

3. 成人及孩童 ALL 間的差別：
10 例成人中，1 例有 CD44 –v6 的表現這 1 例 CD44s 陽性細胞亦小於 50%；59 例孩童中，1 例有 CD44 –v6 的表現，13 例 CD44s 陽性細胞小於 50%（包括 CD44 –v6 陽性一例）。

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