Expression of Cell Differentiation Antigens as a Prognostic Factor in Acute Leukemia

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A. Introduction

Much attention has been devoted to the study of prognostic factors in acute leukemia. Most of the published studies have been concerned with easily measured clinical and hematological parameters [6, 8] while studies requiring specialized techniques such as chromosome or molecular analysis [9] and immunophenotyping of leukemia [1–3, 7] have generally been rare.

In this report, based on the analysis of a large group of leukemia patients, we document that some cell differentiation antigens can be regarded as potential markers, predicting the ability to achieve complete remission and the duration of survival.

B. Materials and Methods

A total of 1054 untreated patients with acute leukemia were subclassified on the basis of the morphocytochemical FAB criteria and typed by means of a panel of 6–21 monoclonal antibodies (MoAb) of the VI series, donated by W. Knapp [5]. The mononuclear cells were isolated from the peripheral blood and/or bone marrow. Only samples with more than 70% blasts in the differential count were tested in indirect immunofluorescence [4, 5] using FITC-labeled goat Fab_2 antibodies against mouse IgG + IgM (Grubb) as the secondary reagent. The criterion for positivity was expression of the antigen by at least 15% of the blast cell population.

In this report special attention will be paid to the subgroup of 437 adult patients with acute nonlymphocytic leukemia (ANLL) – 236 men and 201 women, mean age 53 years (range 16–89 years) – and to the expression of antigens typical for the myelomonocytic line, such as VIM* 2, CD 15 (detected by VIM D5), CD 11_b (VIM 12), and CD 14 (VIM 13).

Patients were treated using one of the following remission-induction protocols: daunorubicin + Ara-C: 3 + 7 days or TAD, and those who reached complete remission (CR) received cyclic mainte-

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* CDw 65

nance chemotherapy administered for periods of 12–36 months. The computer analysis was carried out using the BMDP1L and the Leukos 3 programs. The study was designed to correlate expression of differentiation antigens with CR rate and survival. Survival was measured from the date of entry into the study and included all cases with sufficient data regardless of the remission-induction result. Comparisons involving survival were based on the following tests; generalized Wilcoxon (Breslow), generalized savage (Mantel-Cox), and/or log-rank test.

C. Results

Using the FAB criteria and a panel of reagents detecting the determinants VIM 2, CD 15, 11, 14, glycophorin A, PL* 1–3, CD 24/19, CD 10, E-receptor, CD 7, CD 3, 4, 8, peroxidase (POX), unspecific esterase (ANAE) and PAS, 97.5% of the cases studied were classified as ALL or ANLL, while 2.5% remained unclassifiable.

At the time of this analysis data on the response to therapy of 437 adult patients with ANLL were available and form the basis for evaluation. The overall response rate in this group of patients (age 16–89 years) was 51% (42% CR, 9% PR). The correlation of CD 15 expression with the response to therapy was tested in all these cases. The remaining analyses were carried out in fewer cases, dependent upon the current availability of clinical data.

The relationship of antigen expression to CR rate is presented in Table 1. Significant differences were found only between the subgroups with different CD-15 expression.

The CR-rate was significantly higher in the CD 15-positive group than in the CD 15-negative one ($P<0.05$). A significant difference in the CR rate was also found between the subgroups with the proportions of CD 15-positive blasts above 50% and those with lower rates of expression ($P<0.02$). Expression of the other five antigens studied and of POX and ANAE did not correlate with the response to induction therapy.

An attempt was made to exclude the influence of other risk factors on the response. We found no statistically significant differences in age, WBC count, percentages of blasts and granulocytes in bone marrow, blast counts in peripheral blood, proportions of M_2-FAB subtype, platelet counts, and Hb level between the subgroups compared.

* CD 41–42

Table 1. Relationships of cell-differentiation antigen expression to CR rate in adults with ANLL

Antigen	Expression (% of blasts)	No.	CR (%)	Significance (chi^2)
CD15	<15%	170	34.7	4.4, $P<0.05$
	⩾15%	267	45	
	<50%	323	38	5.4, $P<0.02$
	⩾50%	114	51	
VIM2	<15%	64	42	n.s.
	⩾15%	302	39.7	
CD14	<15%	126	44.7	n.s.
	⩾15%	44	50	
CD11	<15%	131	48	n.s.
	⩾15%	122	37.4	
Glycophorin A	<15%	221	43.3	n.s.
	⩾15%	8	25	

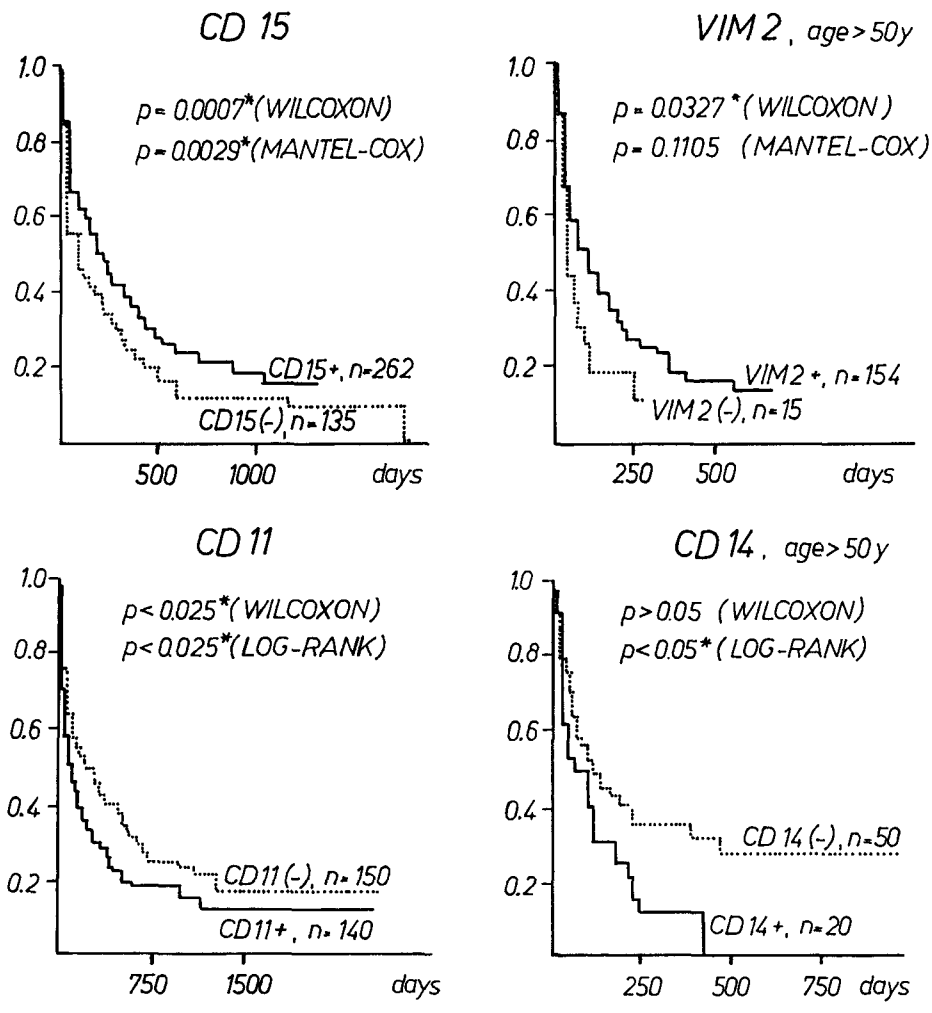

Fig. 1. Kaplan-Meier survival curves in patient groups defined by expression of selected antigens (the analyses of VIM 2 and CD 14 expression deal with subgroups of patients older than 50 years). *Asterisk*, Significant difference

Analysis of Kaplan-Meier survival curves (Fig. 1) shows that the CD 15-positive group had a significantly better outcome (mean 648 ± 353 days, median 218 days, 1-year survival = 41%) compared with the CD 15-negative one (mean 423 ± 279 days, median 76 days, 1-year survival = 29%), $P = 0.0007$ Wilcoxon, $P = 0.0029$ Mantel-Cox. Analogous analysis demonstrated that VIM 2 expression does not correlate significantly with survival, in spite of a higher median survival of the VIM 2-positive group. Significant differences were found when the effect of VIM 2 expression was studied in patients stratified into subgroups by age.

In the subgroup of those over 50 years of age, the VIM 2-positive patients had a better survival (median 101 days) compared with the VIM 2-negative one (median 26 days); $P = 0.03$ by Wilcoxon test.

In contrast to those "positive" interrelationships, the CD 11-positive group of ANLL patients had a shorter survival (median: 95 days, 1-year survival: 28%) than the CD 11-negative one (median: 219 days) 1-year survival: 43%); $P < 0.025$ by Wilcoxon test.

Similarly, a higher CD 14 expression appears to correlate with a shorter survival, but significant differences were found only in subgroups of patients older than 50 years (1-year survival equaled 10% and 35% respectively; $P < 0.05$ by log-rank test).

No association could be found between the survival and the expression of the following determinants; Ia (VID1), p45 (VIP 2b), transferrin (VIP 1), and glycophorin A (VIE G4).

D. Discussion

Today, when a number of significant therapeutic options are available, an attempt should be made to individualize

therapy and to use new, more aggressive therapies mainly in prognostically poor groups of patients who are identified with the use of prognostic indices. The potential utility of cell differentiation antigens as prognostic factors has been investigated to a limited extent mainly in ALL [2, 7], but similar studies in ANLL are rare [1, 3, 4].

In our previous studies we showed that the expression of CD 15 provides prognostic information on the probability of achieving CR [4]. The present study confirms this observation in a larger group of 437 ANLL patients (Table 1) and shows that the predictive significance of CD 15 expression is not associated with other risk factors. As expected from our previous observations, the higher CD 15 expression was found to correlate with a better survival (Fig. 1). Thus, the expression of higher levels of CD 15 antigen is predictive of both a higher CR rate and a longer survival.

Of seven other antigens tested, the expression of CD 11_b, VIM 2, and CD 14 was found to correlate to some degree with survival. However, the significance of these interrelationships was lower. Moreover, VIM 2 and CD 14 antigen expression was predictive only in patients over the age of 50 years. It must be stressed that a high level of CD 15 and VIM 2 expression identified a group of patients with better outcome, whereas CD 11_b and CD 14 positivity were predictive of a shorter survival.

There are few publications referring to the prognostic relevance of immunophenotyping in ANLL. Griffin et al. [3] noted a correlation of My7 (CD 13) and My4 (CD 14) expression with a poor CR rate but found no significant relation to survival. In the same study no correlation could be shown between MCS1 (CDw15) expression and the response to therapy.

It is rather difficult to compare these results with our findings because of different reactivities of the monoclonal antibodies used in both studies. The frequencies of VIM D5 positivity in particular FAB subtypes in our study were as follows: M_1 – 60%, M_2 – 70%, M_3 – 65%, M_4 – 74%, M_5 – 71%. Corresponding frequencies obtained with MCS1 were lower, particularly in FAB subtypes M_1, M_2, and M_3 [3]. Thus, the reactivity of MCS1 is rather of the myelomonocytic type, whereas VIM D5 appears to be more specific for the granulocytic line [5].

Explanations for the higher CR rate and a better survival of the CD 15-positive group of ANLL patients are purely speculative. The CD 15 antigen can be found on normal granulocytic cells, from blasts to granulocytes [5]. It appears to be more specific for the granulocytic line than the VIM 2 antigen, which, in our experience, is expressed more strongly on both granulocytic and monocytic cells and may serve as a key marker of ANLL, being present in over 90% of cases. Assuming that patterns of antigen expression by normal hematopoietic cells are conserved by their malignant counterparts, it appears that CD 15 positivity reflects to some degree the maturity of leukemic cells. From this point of view, our findings are in agreement with the observations indicating a better prognosis in more mature subtypes of ANLL [6, 8].

Finally, the results presented here suggest that examination of the expression of CD 15, CD 11, VIM 2, and CD 14, and probably other myeloid antigens could be a useful addition to the existing systems of risk assignment in ANLL and could contribute to the improvement of therapy.

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