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Detect the level of expression of Cluster Differentiation (CD) marker (CD13 and CD33) in acute myeloid leukamia Iraqi patients

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Abstract: The present study shedding light on the expression of Cluster Differentiation (CD) marker (CD13 and CD33) among AML subtypes.The expression of (CD13 and CD33) was investi-gated in (33) biopsy of Iraqi patients with Acute Myeloid Leukemia (AML) (18 males and 15 females) from the Hematology Clinic department in the teaching hospitals of Baghdad during the period from May 2011 to May 2012 .The tissue sections were analyzed immunohistochemically (IHC) to detection the Expression of CD13 and CD33, Our study showed a high positive immunohistochemical expression of bone marrow tissues for CD13and CD33. The investigation on the percentage of this immunological parameters for patients with AML showed a significant rise (P<0.01) to this parameter levels between groups of patients.

Introduction

AML is the most common subtype of leukemia in adults and accounts for 15-20 % of childhood leukemia [1]. AML is character-ized by continued proliferation and suppressed differentiation of haemopoietic progenitors in the bone marrow with disease cells characterized by enhanced survival and self-renewal. Thus, accumulating numbers of immature haemopoietic progenitors replace the normal red blood cells, white blood cells and platelets [2].

The diagnosis of acute myeloblastic leukemia(AML)is based on cell morphology, cytogenetic and molecular changes, cell markers and clinical data [3]. Immunopheno-typing marker studies have demonstrated that AMLcells are antigenically heterogeneous [4].

Leukemic myeloblasts express a variety of CDs, which reflect commitment to the myeloid lineage as well as a level of maturation [5]. Our study indicates that immunohistochemistry is an alternative to FC for the classification of AML when only a bone marrow biopsy specimen is available and bone marrow aspirate morphologic features and flow cytometry (FC) are unavail-able for analysis [6]. Immunohistochemistry now represents a universally accessible immunophenotyping technique that can be rapidly and accurately applied to diagnosis [7], and classify acute leukemia [5].

A variety of clinical and biologic parameters, including immunophenotype, have been examined for potential value in predicting treatment response and survival. Some reports have suggested a relationship between some antigens(e.g.CD7,CD9, CD11b , CD13, CD14, CD15, CD33, and CD34) and AML prognosis [8]. In the attempt to define the optimal number of markers to determine the immunophenotype in acute leukaemia with a sensitivity of 95%, some recommended markers for AML are: myeloperoxidase (MPO), CD33, CD13, CD14,CD15, CD117, CD34 [9].The association between some immunogenetic markers (CD 13 and CD 33) and the acute myeloid leukemia subtype classification in order to obtain a better understanding of susceptibility to develop the disease, pathogenesis and diagnostic the disease we suggest this study.

Material and method

The (33) patients sample were collected from some Hospitals in Baghdad . The sections were put on positive charged slides and stained immunohistochemically for CD13 and CD33. Immunohistochemical staining was carried out dilution 1:50 (Dako, Denemark). After washing, the samples were stained with diluted liquid DAB, and then counter stained with hematoxylin. Slides washed, dehydrated then mounting, and examining under light microscope at 10X, 20X, 40X magnification using Envision technique using non-commercial material the slides were deparaffinized, rehydrated then blocked. All of the slides were treated with Mouse antihuman CD13 and CD33 primary antibody,

Result and Discussion:

The results concerning the prognostic value of surface antigens expression in AML. The investigation tissue expression of CD13 and CD33 markers, on the percentage of this immunological parameters for patients with AML showed a significant rise (P<0.01) to this parameter levels between groups of

patients.CD13 distinguishes antigens on the surfaces of the granulocyte and monocytic mature and immature, CD33 Is a trans-membrane receptor expressed on cells of myeloid lineage[10]. It is usually considered myeloid-specific, but it can also be found on some lymphoid cells [11]. Our study explains CD13& CD33 were positive in the majority of cases studied. CD13 or CD33, or both may be seen in all subtypes of AML and this agreed with other investigator, Al-Moussawi said that the percentage of the CD13 and CD33 marker in lymphoid cells isolated from AML patients showed the significant increased in this marker levels when compared with healthy people [12]. Moreover, compared between AML and ALL the CD33, CD13 and CD117 had much higher specificity for AML since it was rarely observed in ALL [13], the same writer referred to myeloid markers: CD13 and CD33 expressed in the vast majority of AML samples at the rate of 86.7% , 96.1% [13], and so refer Chianese and his group to the relationship between the antigens (CD13and CD33) and AML prognosis[8].

Table (1) Comparative between subtype of AML in CD13 and CD33 expression (%).

Subtype	CD13	CD33
M0	36.25	37.50
M1	46.00	51.00
M2	43.33	46.67
M3	26.25	38.75
M4	26.25	56.25
M5	20.00	45.00
M6	20.00	45.00
Chi-quare	10.025 **	6.331 **
** (P<0.01).		

A B

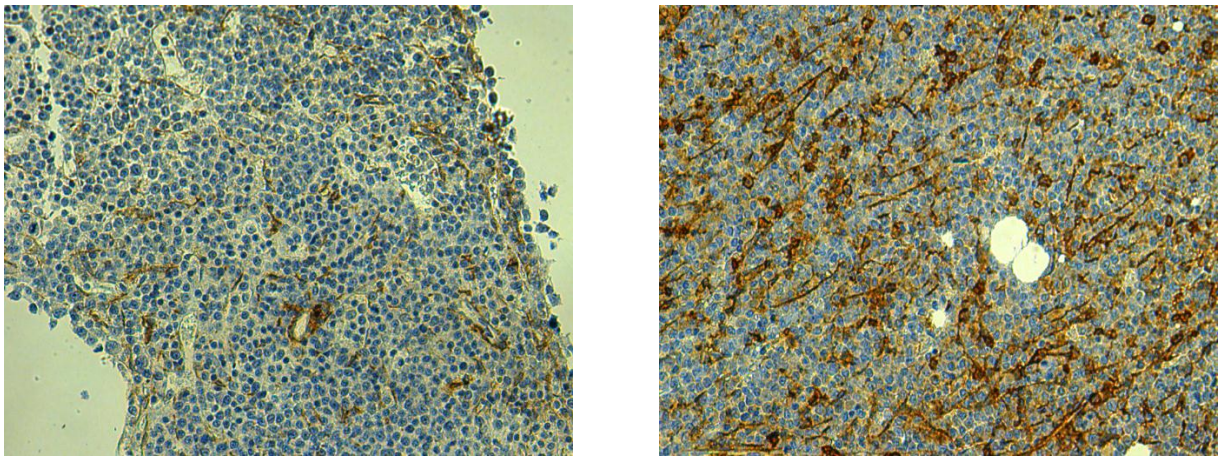


Figure (1)Immunohistochemical tissue expression of CD13 on AML patients (BMB) [A:negative ; B:positive for CD13] X20

A B

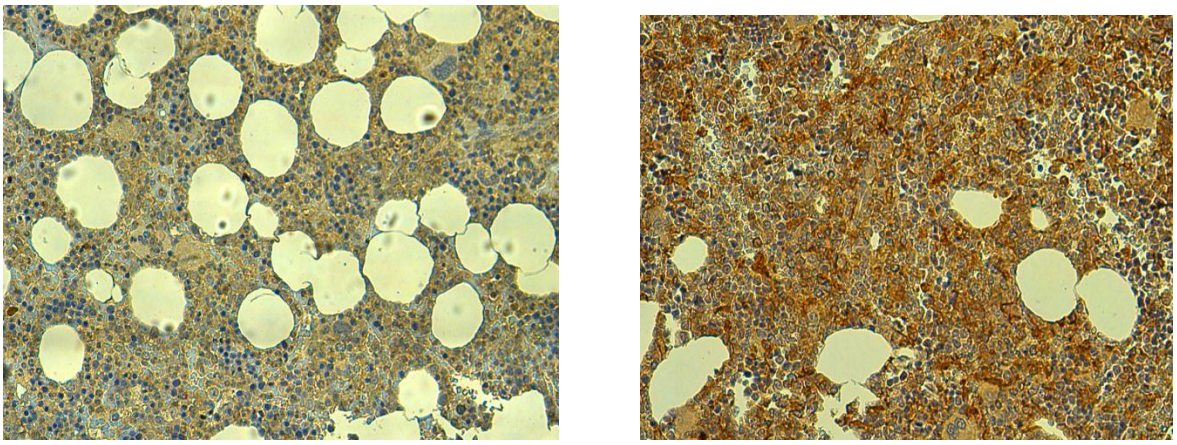
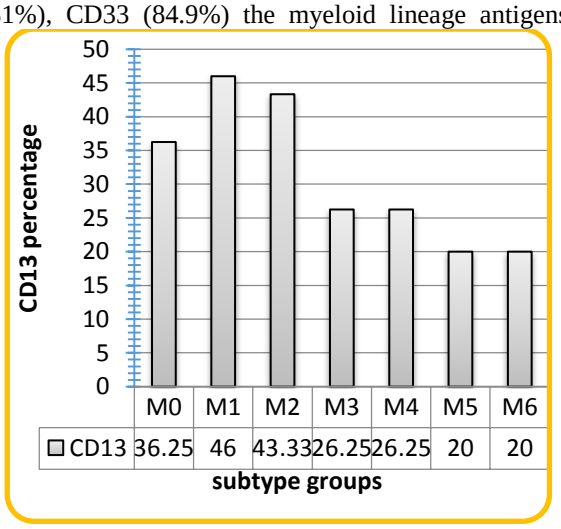
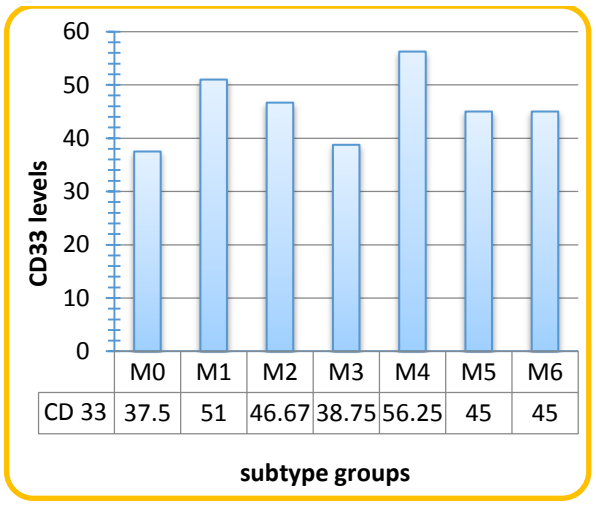


Figure (2) Immunohistochemical tissue expression of CD33 on AML patients (BMB) [A:negative ; B:positive] X20

So Sanaat and her group found among different markers, the most positive markers the following CD13 (81%), CD33 (84.9%) the myeloid lineage antigens, and the hematopoiesis progenitor cell markers HLA-DR (46.1%) [14].



Percentage of CD13+ immunohistochemical tissue expression in AML bone marrow biopsy



Percentage of CD33+ immunohistochemical tissue expression in AML bone marrow biopsy

In the diagnostic By FC the CD13 was positive in the majority of cases studied [6], whereas the M3 variable (heterogeneous) CD13 and bright CD33 [15]. CD13, CD33, or both may be seen in all subtypes of AML.

In some of the prior investigations, presence of CD33 was considered a favorable prognostic factor [16]. By expression in segmented neutrophils was very weak to absent. Strong expression of CD33 was maintained in mature monocytes.CD33 stain-ing in eosinophils appeared similar to that in neutrophils and mature eosinophils showed (weak–absent) staining; however, there was some CD33 expression seen in more immature eosinophilic precursors [17], for that we prefer the dependent on this CDs marker as a biomarker for diagnostic. Where CD13 and CD33 are

FC the CD33 was more intensely positive than CD13 and was negative in 1 case each of M0 and M2 and 2 cases of M1. All cases were positive for CD13 or CD33 [6]. There was a progressive decrease in CD33 staining intensity with granulocytic maturation;

important markers for distinguish between AML subtypes and ALL, and specifically between M0 and ALL.

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