LETTER TO THE EDITOR

External validation of the accuracy of 'CLLflow score'

B-cell chronic lymphoproliferative disorders (B-CLDs) are a group of heterogeneous diseases in both morphological and immuphenotypic features, as well as in clinical behavior.[1] Flow cytometry has a relevant importance for the accurate diagnostic definition of the various type of B-CLDs. Unfortunately there is not a single marker that can univocally define a specific disease entity. For this reason, in practical laboratory routine, a combination (the so-called 'panel') of monoclonal antibodies is currently used.

Aiming at better classifying B-CLDs, >20 years ago, the Royal Marsden British group in London proposed a scoring system based on the surface expression of five markers (CD5, CD23, FMC7, CD22 and surface immunoglobulins (SmIg)).[2] The same group later showed that the accuracy of this score could be improved by the use of CD79b instead of CD22 ('revised Matutes score').[3] Despite the progresses of the recent years, especially in the field of genetics and molecular biology, the Matutes score is still widely used. However, the diagnosis of some B-CLDS remains inconclusive.

Recently, the transmembrane type Ia glycoprotein belonging to the immunoglobulin superfamily identified by CD200 has been shown to have differential expression in B-CLDs.[4] In particular CD200 was found to be useful in distinguishing chronic lymphocytic leukemia (CLL), the most common form of B-CLDs, from mantle cell lymphoma (MCL).[5] In light of this, Kohnke *et al* very recently reported on the diagnostic CD23+/CD5+ B cells and then subtracting the percentages of CD79b+ and FMC7 +B cells.[6] They used CD200 instead of SmIg and the percentage expression of each monoclonal antibody on CD19-positive cells. Overall, a CLLflow score >0 is suggestive of CLL, while a score ≤0 is consistent with a diagnosis of non-CLL disorder.

Very recently, our group also evaluated the accuracy of a simplified score for the diagnosis of CLL in which only four markers are used (CD5, CD23, CD200 and SmIg).[7] In our hands, the proposed score showed a higher sensitivity and specificity with respect to the modified Matutes score.

Here we report the results of a retrospective multicenter study on 216 patients evaluated for B-CLDs at our Institutions between September 2009 and April 2018, aiming at comparing the accuracy of 'CLLflow score' and our score. Median age at diagnosis was 70 years (range 38–97 years), and 131 (61 per cent) were male. The diagnosis, according to investigator assessment, was CLL in 161 patients (75 per cent). The non-CLL patients (n=55, 25 per cent) were 32 marginal zone lymphoma (MZL) (15 per cent), 10 MCL (5 per cent), 8 follicular lymphoma (FL) (4 per cent), 4 lymphoplasmacytic lymphoma (1 per cent) and 1 hairy cell leukemia (<1 per cent). Sensitivity and specificity for the diagnosis of CLL (vs non-CLL) were calculated for 'modified Matutes score', 'CLLflow score' and our score. McNemar's test was used to compare the accuracy between different scores. Statistical analyses were performed using IBM SPSS Statistics for Windows, V.22.0.

Sensitivity and specificity for the diagnosis of CLL (vs non-CLL) for 'CLLflow score', 'modified Matutes score' and our score are depicted in table 1. In this patient cohort, the accuracy of 'CLLflow score' and our score was not significantly different. In fact, only 5 out 216 patients (2.3 per cent) had a discordant diagnosis when the two scores were applied: two patients with CLL ('CLLflow score −5 and 98, respectively; our score 3 and 2, respectively), two patients with MZL ('CLLflow score' 22,9 and 99,4, respectively; our score 1 and 2, respectively), and 1 FL patient ('CLLflow score' −34,1; our score 3). On the contrary, the diagnostic accuracy of both 'CLLflow score' and our score was significantly different as compared with 'modified Matutes score' (p<0001 and 0.002, respectively), with a higher number of discordant cases (13 cases for 'CLLflow score' vs 'modified Matutes score', and 14 cases for our score vs modified 'Matutes score').

Taken together, the results of this analysis confirm the usefulness of both 'CLLflow score' and our score for the diagnosis of CLL. In our opinion, the latter score has the advantage of requiring few monoclonal antibodies (four instead of five). Obviously, these findings need further confirmation by means of external validation studies, before being proposed as standardized diagnostic procedures. The usefulness of CLLflow score has been already confirmed in a study focusing only on B-CLDs with inconclusive immunophenotype.[8] Overall, data supporting the relevance of CD200 expression evaluation for the diagnosis of CLL are undoubtedly strong, thus suggesting the mandatory inclusion of this marker in the diagnostic panel for B-CLDs.

Table 1 Sensitivity and specificity of different diagnostic scores for the diagnosis of chronic lymphocytic leukemia (CLL) in the present cohort

Score	Sensitivity for CLL diagnosis (%)	Specificity for CLL diagnosis (%)
Matutes modified≥3	91	95
Matutes modified≥4	67	96
CLLflow score>0	98	91
Our score≥3	97	93
Our score=4	48	98

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