Is the Aberrant Expression of p53 by Immunocytochemistry a Surrogate Marker of TP53 Mutation and/or Deletion in Chronic Lymphocytic Leukemia?

Correspondence

To the Editor

We read with interest the article by Chang et al in which the aberrant expression of nuclear p53 detected by immunohistochemical analysis in 10% or more of bone marrow biopsy cells of 14 of 110 patients affected by chronic lymphocytic leukemia (CLL) identified all but 1 case with del(17p) in 10% or more of cells by fluorescent in situ hybridization (FISH).

Deletion of chromosome 17p13 occurs in 5% to 7% of untreated CLLs and in 20% to 30% of relapsed or resistant cases and is strongly associated with TP53 mutations of the remaining allele; however, in 3% to 4.5% of cases, the TP53 mutation is not associated with the deletion, but it still maintains its adverse prognostic value.3,4 TP53 mutations lead to an accumulation of an abnormal prolonged half-life p53 protein, which can be detected by immunohistochemical or immunocytochemical analysis.1,5,6

In CLL, the detection of TP53 alterations is strongly associated with an aggressive disease resistant to therapy and overall poor survival.7-9 It is so far the only biologic parameter that can direct the choice of treatment and the indication for allogeneic stem cell transplantation.10

Owing to the importance of a rapid and reliable screening of TP53 status in patients with CLL, we evaluated a cohort of 440 CLL cases (36% at diagnosis, 53% at first progression, 11% relapsed/resistant) for TP53 dysfunction combining 2 different methods: direct sequencing of TP53 exons 5 to 8 by polymerase chain reaction and p53 detection by immunocytochemical analysis on peripheral blood cells, as previously described.1,5,11 The anti-p53 monoclonal antibody (DO-7, DAKO, Glostrup, Denmark) was the same used by Chang et al.1 In a subgroup of 300 patients, del(17p) by FISH was also assessed, as previously described.12

Of the 440 cases analyzed, 19 (4.3%) were found to have p53 protein expression in 10% or more of circulating cells and 35 (8.0%) harbored a TP53 mutation; of the 300 cases evaluated by FISH, 27 (9.0%) had del(17p) in 10% or more of the cells.

Cases immunocytochemically positive for p53 carried a mutated TP53 in 10 cases and were wild-type in 9; immunocytochemically negative cases carried a mutated TP53 in 25 cases and were wild-type in 396. Thus, the sensitivity and specificity of p53 protein assessment by immunocytochemical analysis to detect a TP53 mutation were 29% and 98%, respectively; the positive predictive value (PPV) and negative predictive value (NPV) were 53% and 94%, respectively.

Cases that were immunocytochemically positive for p53 carried a del(17p) in 7 cases and were nondeleted in 6; negative cases carried a del(17p) in 20 cases and were nondeleted in 267. Thus, the sensitivity and specificity of p53 protein assessment by immunocytochemical analysis to detect a del(17p) in 10% or more were 26% and 98%, respectively; the PPV and NPV were 54% and 93%, respectively.

Overall, based on the immunocytochemical results, 25 (71%) of 35 mutated TP53 cases and 20 (74%) of 27 deleted cases were not identified. Focusing on the 20 patients with an amount of CLL cells carrying del(17p) of 20% or more, p53 protein expression was still not detectable in 65% of cases.

To our knowledge, our series is one of the largest assessing the value of p53 protein expression as a screening tool to identify patients with CLL with TP53 aberrations due to gene mutations and/or deletions. The clinical impact of false-negative cases is the most relevant. Some of them can be justified by the type of TP53 mutation. In fact, 9 (36%) of 25 immunocytochemically false-negative cases with mutated TP53 showed microdeletions or nonsense mutations that do not cause protein accumulation.5,13 Moreover, 8 (40%) of 20 false-negative immunocytochemical cases with del(17p) of 10% or more showed the same type of mutations or a wild-type configuration on the TP53 remaining allele.

False-positive cases were also detected, but they can be justified by the fact that protein accumulation may also occur in the absence of TP53 mutations due to other mechanisms of protein stabilization (ie, mutations and deletions of the ataxia telangiectasia-mutated gene or MDM2 alterations).14

Two studies assessed the value of p53 immunocytochemical detection in CLL. Our group previously published on the predictive value of p53 immunocytochemical expression in identifying gene mutations in 88% of cases.6 However, no data on false-p53-negative cases with TP53 abnormalities were provided. Lepelley et al15 found concordant results of p53 immunocytochemical analysis and TP53 mutations in 123 (96%) of 128 patients with various hematologic malignancies, including CLL. False-negative immunocytochemical cases showed nonsense TP53 mutations, which presumably led to reduced levels of truncated p53.

In our experience in a large series of CLL cases, the sensitivity of p53 assessment by immunocytochemical
analysis is such that we can not recommend it as a valid tool for identifying all patients at high risk. Although the difference between the study by Chang et al and ours could be due to technical differences between immunohistochemical and immunocytochemical procedures, even adopting the same anti-p53 antibody, unfortunately, it cannot be ignored that in more than one fourth (9/35) of TP53-mutated cases, the type of mutation leads to the deletion or truncation of the protein that, as a consequence, does not accumulate and cannot be detected by immunocytochemical analysis.

References