and showed normal blood flow in portal and suprahepatic veins. Percutaneous liver biopsy was performed and revealed intracellular cholestasis, preservation of hepatic architecture, microbiologic cultures were negative. Renal function was secondarily impaired with oliguria followed by hypotension that required management with vasoactive drugs. Dexamethasone treatment was begun and, because a bone marrow aspirate confirmed complete remission, ATRA was discontinued. After three days, the clinical symptoms resolved, and there was a reduction in the hepatomegaly; normalization of hepatic and renal function was achieved in the following two weeks. At present, the patient remains in complete morphologic, cytogenetic and molecular remission.

Although ATRA is generally well tolerated, serious adverse effects have been reported, the ATRA syndrome being the most important of them. This syndrome occurs with an incidence ranging from 15 to 27%,1-3 ATRA liver toxicity, consisting in mild, transiently increased aminotransferases or bilirubin, is a well-described adverse effect.1,4,5 In our case, the patient developed life-threatening hepatic toxicity consisting of acute hepatomegaly with severe elevation of cholestatic enzymes and secondary renal failure, but without other signs of the ATRA syndrome. We ruled out other causes of acute hepatomegaly and because of the suspicion of hepatic injury by ATRA, this drug was discontinued. The clinical condition of the patient improved dramatically in the following days with normalization of parameters of liver function. The biopsy findings observed were consistent with our hypothesis of ATRA-induced hepatic toxicity.

To our knowledge, only two other cases of severe acute toxicity due to this drug have been reported.3,6 Mechanisms of liver damage caused by ATRA are unknown, but impaired glucuronidation of its secretion resulting in cholestatic jaundice have been proposed.7 Addition of ATRA to other chemotherapeutic drugs, including idarubicin, does not necessarily result in an increase of hepatic toxicity, although other adverse effects could be increased.6,10 Because ATRA therapy can, in some cases, induce a life-threatening hepatic complication, as in the case reported here, liver enzymes should be monitored carefully. If hepatic toxicity becomes apparent, the drug should be withdrawn.

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Key words
Acute promyelocytic leukemia, all-trans-retinoic acid, hepatotoxicity, hepatomegaly, adverse effects.

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References

Long-term disease-free acute myeloblastic leukemia with inv(16) is associated with PCR undetectable CBFβ/MYH11 transcript

This study investigates the benefits of minimal residual disease in a subset of AML patients with inv(16) by means of non-quantitative detection of CBFβ/MYH11 fusion transcript. We found that CBFβ/MYH11 RT-PCR negativity is associated with cure of disease.

Sir,
The expression of CBFβ/MYH11 fusion mRNA provides a potential molecular marker that can be detected in leukemic cells taken from a subset of patients with acute myelocytic leukemia (AML).1-4 (Gene Bank
accession numbers AF249898, AF251768, AF249897). Some authors\(^5\) have reported favorable clinical outcomes despite persistence of CBF\(^{b}\)/MYH11 transcript detected by reverse transcription-polymerase chain reaction (RT-PCR), so the significance of minimal residual disease (MRD) is presently not defined. We have applied RT-PCR assay for CBF\(^{b}\)/MYH11 analysis\(^4\) on bone marrow samples from 9 AML patients with long-lasting complete remission (CR) (defined as >36 months disease free), after induction chemotherapy and consolidation (overall survival (OS) median 49 months; range 40-102 months; disease free survival (DFS) 45 months, range 63-17 months from CR) in order to verify their MRD status. Patients

### Table 1. Clinical and therapeutic characteristics of the AML patients with inv(16) or involvement of 16q22 band

<table>
<thead>
<tr>
<th>No.</th>
<th>Name</th>
<th>Sex/Age</th>
<th>FAB cytotype</th>
<th>Therapy 1st/2nd/3rd line</th>
<th>Type of transcript</th>
<th>OS/DFS mos.</th>
<th>Clinical outcome after CT</th>
<th>Karyotypic analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>B.A.</td>
<td>F/44</td>
<td>M1</td>
<td>ICE/NOVIA/FLAG/ABMT</td>
<td>A</td>
<td>67/63</td>
<td>CR/AW</td>
<td>46,XX(3)/46,XXinv(16) (p13q22)/23(17)</td>
</tr>
<tr>
<td>2</td>
<td>M.D.</td>
<td>M/27</td>
<td>M4E</td>
<td>ICE/NOVIA/ABMT</td>
<td>A</td>
<td>62/61</td>
<td>CR/AW</td>
<td>46,XY(1)/46,X;del(7)(q22),inv(16)(p13q22)/13(13)</td>
</tr>
<tr>
<td>3</td>
<td>V.M.</td>
<td>M/32</td>
<td>M4E</td>
<td>ICE/NOVIA/BMT</td>
<td>A</td>
<td>61/60</td>
<td>CR/AW</td>
<td>46,XY(2)/46,X;inv(16) (p13q22)/13(13)</td>
</tr>
<tr>
<td>4</td>
<td>R.I.</td>
<td>F/49</td>
<td>M4E/breast c.</td>
<td>ICE/NOVIA/FLAG/FLAG/BMT</td>
<td>A</td>
<td>47/44</td>
<td>CR/AW breast c.</td>
<td>46,XX(1)/46,XXinv(16) (p13q22)/23(1)</td>
</tr>
<tr>
<td>5</td>
<td>D.P.</td>
<td>F/53</td>
<td>M4E</td>
<td>ICE/NOVIA/ABMT/PBSC/FLAG/FLANG</td>
<td>A</td>
<td>49/17</td>
<td>2nd CR/AW</td>
<td>46,XY(2)/46,XXinv(16) (p13q22)/13(28)</td>
</tr>
<tr>
<td>6</td>
<td>C.R.</td>
<td>M/58</td>
<td>M4E</td>
<td>ICE/NOVIA/FLANG/ABMT/PBSC</td>
<td>A</td>
<td>47/45</td>
<td>CR/AW</td>
<td>46,XY(2)/46,XXinv(16) (p13q22)/13(19)</td>
</tr>
<tr>
<td>7</td>
<td>B.C.</td>
<td>M/32</td>
<td>M4E (GS)</td>
<td>ICE/NOVIA/BMT</td>
<td>A</td>
<td>46/45</td>
<td>CR/AW</td>
<td>46,XY(3)/46,XXinv(16) (p13q22)/13(16)</td>
</tr>
<tr>
<td>8</td>
<td>V.P.</td>
<td>M/53</td>
<td>M4E</td>
<td>ICE/NOVIA/ABMT</td>
<td>A</td>
<td>40/38</td>
<td>CR/AW</td>
<td>46,XY(1)/46,XXinv(16) (p13q22)/13(11),+22(12)/47,XY(t(9;19)(q22;q13),inv(16)(p13q22)+22(8)</td>
</tr>
<tr>
<td>9</td>
<td>B.A.</td>
<td>M/34</td>
<td>M4E</td>
<td>MECS/MEC4/ABMT/FLAG/FLANG</td>
<td>A</td>
<td>102/39</td>
<td>2nd CR/AW</td>
<td>46,XY(1)/46,XXinv(16) (p13q22)/14(14)</td>
</tr>
</tbody>
</table>

OS = overall survival; DFS = disease-free survival; CT = chemotherapy; AW = alive and well.

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**Figure 1. Display of RT-PCR results of all tested patients.** The figure shows the data of our series of patients after the achievement of CR. The time of follow-up is denoted in months which represent the patients’ follow-up. Open triangle (▲) and full triangle (▼) represent samples that were negative and positive, respectively, for cytogenetic analysis. Each dot represents an RT-PCR assay performed at the indicated time. Open dots (○) represent samples that were negative for the presence of a CBF\(^{b}\)/MYH11 transcript by RT-PCR analysis. Full dots (●) represent positive samples. Only samples with adequate RNA quality for amplification of control RNA are included.
received different protocols of induction chemotherapy including an anthracycline (daunorubicin or idarubicin) alone or in combination with cytosine arabinoside (Some biological and clinical data are given in Table 1). The 9 patients (#1-9) achieved CR after different schedules of ablative induction and consolidation chemotherapy protocols (Table 1). All 9 patients who achieved clinical remission are currently alive in first (7 cases) (77%) or second (2 cases) (23%) CR (Figure 1), confirming that AML with inv(16) is curable by ablative therapy in a high percentage of cases. Two patients experienced relapse (#5 and #9 in Table 1) but they achieved a second CR, one of these lasting more than 36 months. On these nine patients, remission bone marrows aspirates were obtained after achievement of CR and used for molecular analysis.5 Cytogenetic studies and RT-PCR analysis were performed as reported.4 Experiments using inv(16) RNA in serial dilution of total RNA from normal individual were also conducted: the level of sensitivity of type A fusion sequence amplification after nested PCR was of 1 tumor cell in 106-107 non-neoplastic cells.4 At diagnosis, chimeric cDNAs were detected after amplification in all 9 patients (Table 1). Only one type out of nine reported chimeric transcripts was found, representing 1921 position fusion point within MYH11 spliced to position 495 of CBFβ (type A). This finding is in line with the concept that AML with inv(16) is strictly associated with the type A transcript.4, 5 The results of RT-PCR analysis in remission samples are schematically represented in Figure 1. In no cases, were CBFβ/MYH11 transcripts visible on the ethidium bromide gels. In the 2 cases who experienced relapse, (patients #9 and #13), no prediction of re-emerging residual cells expressing the CBFβ/MYH11 transcript was found, representing 1921 position fusion point within MYH11, indicating that: 1) PCR negativity should be considered the therapeutic goal in these patients; 2) the cure of AML with inv(16) by chemotherapy is accompanied by elimination of at least below our RT-PCR sensitivity levels, of residual cells expressing the CBFβ/MYH11 transcript. Quantitative PCR analysis could be useful for improving the significance of MRD.8 This means that the RT-PCR assay is a useful prognostic tool not only in the induction and consolidation treatment phases, but also during long-lasting follow-up.10

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Myelofibrosis in myeloid malignancies with 3q26 cytogenetic abnormalities

3q abnormalities define a subtype of myeloid malignancies characterized by similar clinical, morphologic and cytogenetic features, poor response to therapy and short survival. Trilineage myelodysplasia, affecting particularly the megakaryocytic line, is