of p210 bcr/abl fusion protein in LG7 cells, detection
and quantification of p210 bcr/abl transcript in these
cells was done using a quantitative, competitive
PCR. The presence of bcr-abl transcripts in the LG7
cell line was further demonstrated by western blot
analysis (data not shown). Our data suggest that
ATRA can potentiate the inhibitory effects of IFN-α
both on Ph-negative and Ph+ leukemic cells. The
mechanism(s) of synergism is unknown. It does not
seem to be related to p210 expression, but appears
to be influenced by preincubation of target cells with
ATRA. These findings suggest that pretreatment with
ATRA could induce activation of IFN-α-induced genes
which in turn could favor the clinical response to
IFN-α.

Domenico Russo, Gianluca Tell, Lucia Marin,
Mario Tribelli, Maria Alessandra Santucci, Carlo Pucillo
Chair and Division of Hematology, Department of Medical and
 Morphologic Research; *Section of Immunology, Department of
Biomedical Science and Technology, Udine University, Udine,
* Institute of Hematology and Medical Oncology
“L. e A. Seragnoli”, Bologna University, Bologna, Italy

Key words
ATRA, IFN-α, Ph+ CM L, p210 bcr/abl, cell lines

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Correspondence
Dr. Domenico Russo, M.D., Clinica Ematologica, Policlinico
Universitario, piazza S. Maria della Misericordia, Udine, Italy.
Phone international +39-0432-559662-64 – Fax international
+39-0432-559661- E-mail: domenico.russo@drmm.uniud.it

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Retinoic acid activates interferon regulatory factor-1 gene

Possible evolution of human parvovirus B19 infection into erythroleukemia

Sir,

Since its discovery in 1975, HPV B19 has been identified with an acute form of bone marrow failure in susceptible hosts. It is now well established that HPV B19 is cytotoxic for human erythroid precursors and causes a lytic process in infected cells.

We report the case of a patient with HPV B19 bone marrow infection who developed erythroleukemia (EL). A 69-year-old man was admitted to a local hospital following several days of high fever and pancytopenia. The patient was given antibiotics and several units of red blood cells and then transferred to our institution for further evaluation.

Cytologic and histologic examination of bone marrow cellularity showed markedly reduced and dysplastic erythropoiesis. Blast cells such as myeloblasts and monocyctic blasts accounted for 20% of bone marrow cells. We were impressed by the high number of giant, frequently binucleated, erythroblasts, some of which were morphologically normal while others showed atypical irregularly-shaped nuclei, prominent nucleoli and vacuoles (Figure 1a). Cell pictures suggesting a fusion phenomenon were occasionally observed (Figure 1b). A provisional diagnosis of myelodysplastic syndrome was formulated.

One week after admission a rapid increase in WBC, platelet and reticulocyte count was observed. Bone marrow examination was repeated ten days after admission; the marrow showed marked erythrophagocytic hyperplasia, a few giant proerythroblasts were still present and there were numerous megakaryocytes while the number of blast cells was reduced. A clinical diagnosis of transient aplastic crisis due to HPV B19 was considered. Serologic examination revealed the presence of elevated anti-HPV B19 IgM antibodies and a low level of IgG, by ELISA immunoassay. Virus DNA, tested by PCR, was negative. In the following days a dramatic reticulocytosis followed, the blood count returned to normal values, the patient’s clinical condition improved and he was discharged. He remained in good health for the next six months. He then started complaining of
fatigue and exertional dyspnea for which he was admitted to our hospital. His Hb level was 8.9 g/dL, WBC count was 2.1×10⁹/L. A peripheral blood examination showed the presence of many blast cells (57%). Cytologic and histologic examination showed a hypercellular marrow with 90% agranular blasts. Residual cells comprised erythroblasts with a variety of abnormal cytologic features, from slight megaloblastic characteristics to vacuolization and the presence of bi- and multinucleated giant cells (Figure 1c). Again pictures suggestive of fusion phenomena could be observed, with morphologic details reminiscent of the characteristic features of cell fusion as described in HIV-induced cell-cell fusion: cells made contact by using microspikes to touch and adhere to adjoining cells (Figure 1d). A preliminary diagnosis of acute leukemia was made and later confirmed by established cytochemical studies. Reactivity with anti-glycophorin A was strongly positive in 60% of blasts (APAAP stain with hematoxylin counterstain). A diagnosis of EL was made.

The appearance of EL in a patient who suffered an infectious disease of the erythron a few months before prompted us to investigate whether the leukemic cells where harboring HPV B19. B19 DNA in marrow cells was repeatedly absent when searched for by an in situ hybridization assay. Despite courses of cytotoxic therapy and intensive supportive measures, the patient succumbed to an overwhelming infection.

Our findings suggest the evolution of an acute, primarily self-limiting HPV B19 transient aplastic crisis into EL. The characteristic giant proerythroblasts in the context of acute bone marrow failure first suggested the diagnosis of HPV B19 infection; additionally we observed multiple forms of bi-multinucleated giant cells and occasional morphologic evidence of cell fusion of erythroid precursors. The presence of...
Hybrid (myeloid-erythroid) blasts has been proposed to be a characteristic bone marrow feature in EL, but has not as yet been emphasized as a morphologic feature of the disease. Erythroblasts with peripheral chromatin condensation are sufficiently characteristic for a provisional diagnosis of B19 infection to be made on the histology and rare erythroblasts with this morphologic feature could be observed (Figure 1e). While we can firmly correlate the infectious phase of the disease to HPV B19, the association with the neoplastic phase is only hypothetical because of the absence of the virus in the marrow cells when searched for by in situ hybridization.

An alternative interpretation is that the patient had already subclinical EL which was unmasked by HPV B19 infection. If the virus hit compromised marrow, in which a shrunken compartment of normal hematopoiesis coexists with a still subclinical competing leukemia clone, pancytopenia may follow. Resolution of the virus infection might have given the normal marrow a chance to re-establish normal blood counts transiently, but EL finally expanded and took over. Since this is a case report it would be inappropriate to draw general conclusions. Nevertheless interesting speculation arise from this report: is there an association between HPV B19 and EL and can cell fusion play a role in the cytogenesis of giant multinucleated erythroblasts?

Key words
Parvovirus B19 infection, erythroleukemia, cell fusion.

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Correspondence
Gianmaria Sitar, M.D., Istituto di Medicina Interna e Oncologia M edica, IRCCS Policlinico S. Matteo, 27100 Pavia, Italy. Phone: international +39-0382-502567 - Fax international +39-0382-526223.

Pulmonary thromboembolism in thalassemia intermedia patients

Sir,
We and others have reported thromboembolic phenomena in patients with β-thalassemia major (TM). Thalassemia intermedia (TI) patients have a phenotypically milder disease, not requiring regular blood transfusions. However, TI also predisposes to thrombotic events, similar to the life-long hypercoagulable state that exists in TM patients. It is unknown whether TI patients who suffer thrombotic events have other inherited or acquired predisposing risk factors. We report the cases of two TI patients with no other risk factors for thrombosis who suffered from potentially life-threatening pulmonary thromboembolism. Both patients presented with increasing dyspnea of recent onset. Investigations revealed bilateral pulmonary thromboembolism. Pertinent clinical details are summarized in Table 1. The results of a screen for hypercoagulable states are shown in Table 2.

These two patients with TI suffered from bilateral pulmonary thromboembolic disease with unusual features. No risk factors (other than thalassemia) were present. Furthermore, no source of emboli could be

Table 1. Clinical details of the patients.

<table>
<thead>
<tr>
<th>Patient #1</th>
<th>Patient #2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>Sex</td>
</tr>
<tr>
<td>51/Female</td>
<td>46/Female</td>
</tr>
<tr>
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<td>IVS1nt6/?</td>
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<tr>
<td>Splenectomy</td>
<td>yes</td>
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<tr>
<td>Cholecistocstomy</td>
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<tr>
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<tr>
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<td>Hb (g/dL)</td>
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<td>PO2 (mm Hg)</td>
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</tr>
<tr>
<td>PCO2 (mm Hg)</td>
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</tr>
</tbody>
</table>
| Ventilation/Perfusion scan | normal/
| Abdominal ultrasound | normal |
| Other imaging studies | doppler of legs normal |

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