Deletions of the long arm of chromosome 6 have been described in acute and chronic lymphocytic leukemia (ALL and CLL) and prolymphocytic leukemia (PLL), and have been associated with t(14;18)(q21;q21) in non-Hodgkin’s lymphomas (NHL). Of 55 cases of small lymphocytic (sm) NHL, deletions of 6q(21q23) were the most common recurring cytogenetic abnormality. Among 14 sm lym NHL with del(6q)q21q23), this abnormality occurred as a solitary change in 3 cases. Each of these 3 cases, and 5 additional cases with del(6q) and other abnormalities, showed atypical larger forms with the morphologic appearance of prolymphocytes or paraimmunoblasts in the peripheral blood. In comparison, of the 11 cases without del(6q) and circulating abnormal cells, prolymphocytoid forms were observed in 4 cases (P < .001). Of the 31 sm lym without del(6q), trisomies of chromosomes 3, 12, or 18, or t(11;14)(q13;32) occurred in greater than 10% of cases. Proliferation centers or infiltration by larger forms were observed in similar proportions of tissue sections derived from sm lym NHL with or without del(6q). The presence of the larger forms in the peripheral blood did not have an adverse prognostic impact on the survival of the del(6q) cohort, who experienced a median survival in excess of 6 years. All 14 cases of del(6q) sm lym NHL were characterized by a mature B-cell phenotype. Expression of CD11c, a feature of a CLL/PLL variant previously described, was not detected in 9 cases analyzed. In 5 cases of del(6q) sm lym NHL, no circulating abnormal lymphocytes were noted. Twelve cases presented with, or developed, clinical splenomegaly. These results suggest that deletion of a gene or genes at 6q21-23 is associated with the pathogenesis of a subset of B-cell sm lym NHL that may display larger prolymphocytoid cells in the peripheral blood, but that follows a clinical course typical of other well-differentiated lymphocytic neoplasms.

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Submitted July 23, 1993; accepted December 22, 1993.

Supported by Grants No. CA-34775 and CA-08748 from the National Institutes of Health and the Lymphoma Foundation, K.O. is supported by a Clinical Oncology Career Development Award from the American Cancer Society.

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0006-4971/94/8309-0010$3.00/0

Comparison of cases with differing cytogenetic abnormalities were made using the method of inferences from proportions based on $\chi^2$ analysis.12

RESULTS

Cytogenetic and morphologic features. Of 55 sm lym NHL, the most frequent recurring cytogenetic abnormality was a del(6q) or add(6q), which was observed in 14 (26%) cases. Other abnormalities previously described in low-grade NHL2 that were observed in greater than 10% of cases included trisomy 3, trisomy 12, trisomy 18, and t(11;14)(q13;q32). Of these, only del(6q) and t(11;14) were observed as sole karyotypic abnormalities in individual cases. The t(11;18)(q21;q21) translocation, previously described in sm lym NHL,14 was not observed in this series.

The full karyotypes of the 14 cases with del(6q) or add(6q) are shown in Table 1. All cases shared a common region of deletion encompassing 6q21-23. In 3 cases (nos. 753, 1000, and 1046), the del(6q) was the sole karyotypic abnormality. Recurring additional nonrandom cytogenetic aberrations comprised trisomy 18 (cases no. 279, 625, and 829) and trisomy 12 (cases no. 304 and 1145). Case no. 910 had a t(11;14)(q13;q32).

Whereas all cases were classified as sm lym NHL on tissue section, biopsies of 8 of the 14 cases, including the cases with del(6q) as the solitary abnormality, demonstrated a mixed population of both small and large neoplastic cells in proliferation centers (pseudofollicles) or interspersed with the smaller well-differentiated cells. A similar proportion of the 41 cases of sm lym NHL with aberrations other than del(6q) showed pseudofollicles or populations of larger cells ($P > .2$). Of 8 cases of del(6q) NHL with circulating cells, all showed atypical larger forms, compared with 4 of 11 cases of sm lym NHL with circulating cells and abnormalities other than del(6q) ($P < .001$). All 3 cases with del(6q) as a solitary abnormality demonstrated larger atypical forms as 10% to 50% of the lymphoid population in the peripheral blood.

In cases with circulating larger forms, the malignant cells showed moderate cytoplasm, condensed chromatin, and one or more nucleoli suggestive of lymphocytes or paraimmunoblasts (Figs 1 and 2). In other cases, there were variable numbers of prolymphocytoid forms intermixed with cells showing nuclear indentations (Fig 3A) or cytoplasmic projections (Fig 3B), or plasmacytoid forms of varying size and morphology (Fig 4). In some cases (eg, no. 304), the larger
Fig 1. (A) Morphology of neoplastic lymphocytes in a case of sm lym with del(6q) as the sole cytogenetic abnormality (case no. 1046). A population of larger cells shows moderate cytoplasm and condensed nuclear chromatin with nucleoli. (B) Histologic sections from the same case. Sections show a dimorphic population of malignant lymphocytes consisting of small round lymphocytes, prolymphocytes, and scattered large paraimmunoblasts. Immunophenotypes of lymphoma cells of del(6q) sm lym NHL. Immunohistochemical and flow cytometric analysis of the tissue and bone marrow specimens of the del(6q) sm lym NHL was consistent with a B-cell phenotype in all cases (Table 1). In 9 cases tested, these findings were confirmed by demonstration of clonal rearrangement of the IGH gene. A circulating paraprotein was shown in 5 cases. Nine cases expressed CD5 on neoplastic cells that coexpressed B-cell markers in the absence of other T-cell antigens (Table 1). Two of these cases (nos. 910 and 1000) showed less than 10% mouse red blood cell rosetting (mrbcR). The other cases with less than 10% mrbcR (nos. 279 and 941) demonstrated plasmacytoid features. CD10 and CD11c reactivity was absent in tissue sections of all 9 cases assessed by immunohistochemical analysis, including 2 cases in which the diagnosis of hairy cell leukemia (HCL) was entertained (nos. 941 and 1068), the larger cells comprised greater than 80% of the lymphocytes, whereas in other cases (eg, no. 1068), the larger cells comprised only 10% to 20% of the lymphoid population.

In 6 cases of del(6q) NHL (nos. 753, 910, 912, 1000, 1023, and 1145), biopsies were performed at the time of relapse of disease (Table 2). Examination of histologic sections or samples of bone marrow from the initial biopsies of 5 of these cases in which original material was available for review showed no significant change in morphology or percentage of larger forms compared with the relapse biopsies. Of the 8 del(6q) sm lym NHL showing larger forms in the peripheral blood, 3 were studied at the time of initial diagnosis, whereas all 4 cases with prolymphocytic cells in the peripheral blood but lacking del(6q) were studied at relapse. One of these latter cases was a prolymphocytic transformation of a long-standing sm lym lymphoma/leukemia.
Both cases were negative for tartrate-resistant acid phosphatase (TRAP).

Clinical features. The mean age at diagnosis of the 14 patients with del (6q) sm lym NHL was 61 years (median, 58.5), compared with 58 years for cases with other abnormalities (P > .4). The median leukocyte count at diagnosis was $9.4 \times 10^9$/L. Four patients presented with stage I-III disease. As shown in Table 2, treatments were heterogeneous. Survival from the time of initial diagnosis was 84 months for the 14 cases of sm lym with del(6q), compared with 108 months for the 41 cases with karyotypic abnormalities other than del(6q) (P = .06). There was no difference in leukocyte count, proportion with early stage disease, or proportion with extranodal disease between the groups.

In 6 of the 14 cases of del (6q) sm lym NHL, the peripheral leukocyte count was normal at the time of diagnosis, with no evidence of circulating neoplastic lymphocytes. In 6 cases, there were circulating malignant lymphocytes and bulky (>10 cm) lymphadenopathy. One of these (case no. 304) had a peripheral leukocyte count greater than $200 \times 10^9$/L. This elderly patient had massive adenopathy, including a subcutaneous neck mass, and expired 6 months after chemotherapy was initiated. The remaining 2 cases (nos. 941 and 1000) were characterized by primary leukemic presentations with splenomegaly in the absence of adenopathy. These patients demonstrated anemia, thrombocytopenia, and (>70%) lymphocytosis in peripheral blood, with total leukocyte counts of 8.5 and $63.4 \times 10^9$/L, respectively. Both patients were effectively palliated by splenectomy, with normalization of hemoglobin and platelet counts. Of the 14 cases of del(6q) sm lym NHL, 12 showed clinical splenomegaly during the course of their disease.
There was no difference in survival from diagnosis between the 8 cases of del(6q) sm lym NHL with larger forms in tissue sections compared with the 6 cases with monomorphic infiltrates \((P > .1)\), or between the 8 cases with leukemic populations including larger forms and those without circulating cells \((P > .2)\).

**DISCUSSION**

The earliest descriptions of CLL and sm lym NHL identified a subset of cases with varying degrees of larger immature forms with features of prolymphocytes or paraimmunoblasts clustered in pseudofollicles (or "proliferation centers") or dispersed throughout the tumor. In a study of 300 cases of CLL and PLL, a subset of 84 cases was found to demonstrate features of both entities, with 10% to 50% circulating prolymphocytes in the setting of both splenomegaly and lymphadenopathy. Pugh et al. described 16 cases of a rapidly progressive "paraimmunoblastic variant" of sm lym NHL characterized by larger forms, rarely including "blast" like cells in peripheral blood. Recently, 29 variant cases with morphologic and immunologic features of CLL, PLL, or HCL have been shown to express the CD11c antigen. In all prior series, karyotypic analysis was either not performed or was limited to small numbers of patients.

The data presented here comprise the largest series of sm lym NHL, serially analyzed for karyotypic abnormalities; the frequency of del(6)(q21q23) surpassed that of all other cytogenetic aberrations. The del(6q) sm lym NHL appear to share most of the clinical features described for the CLL/PLL.
variant, including the same age at diagnosis and a clinical presentation with both lymphadenopathy and splenomegaly. Although not every case of del(6q) sm lymph NHL demonstrated all features of CLL/PLL or the paraimmunoblastic variant of sm lymph NHL previously described, there was a statistically significant \( P < .001 \) association between the recurring del(6q)(q21q23) and the leukemic manifestation of larger forms in the peripheral blood. The characteristics of the del(6q) cohort were in contrast to PLL, which typically presents in the eighth decade with massive splenomegaly, absence of adenopathy, high levels of peripheral leukocytes (mean, \( 355 \times 10^9/L \)), and a brief survival.\(^{22,23} \) Although case no. 305 demonstrated a leukocyte count typical of PLL, this patient presented with massive adenopathy more characteristic of NHL. Only 2 cases of del(6q) in this series had a leukemic presentation in the absence of significant adenopathy. In 1 of these cases, the total leukocyte count was \( 8.5 \times 10^9/L \), and both cases followed indolent courses after splenectomy.

The documentation of circulating prolymphocytoid forms

### Table 2. Clinical Features of 14 Cases of Small Lymphocytic Lymphoma With del(6q)

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Sex/Age</th>
<th>Treatment Status</th>
<th>Stage</th>
<th>BM</th>
<th>Adenopathy</th>
<th>Circulating Cells</th>
<th>Extramedullary Disease</th>
<th>WBC at DX*</th>
<th>Paraprotein</th>
<th>Rx</th>
<th>Survival (mos)</th>
</tr>
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<tbody>
<tr>
<td>279</td>
<td>F/75</td>
<td>Pre IV</td>
<td>NI, P</td>
<td>+</td>
<td></td>
<td>D</td>
<td>6.9</td>
<td>IgMκ</td>
<td>M2</td>
<td></td>
<td>84.0+</td>
</tr>
<tr>
<td>304</td>
<td>F/84</td>
<td>Pre IV</td>
<td>I</td>
<td>+</td>
<td>+ LF</td>
<td>A</td>
<td>274</td>
<td>IgMκ</td>
<td>CMOPP</td>
<td></td>
<td>6.0e</td>
</tr>
<tr>
<td>625</td>
<td>M/75</td>
<td>Pre FE</td>
<td>IE</td>
<td>-</td>
<td>- B</td>
<td>6.1</td>
<td>-</td>
<td>Surgery</td>
<td>10.0+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>753</td>
<td>M/88</td>
<td>Post IV</td>
<td>I</td>
<td>+</td>
<td>+ LF</td>
<td>-</td>
<td>12.0</td>
<td>-</td>
<td>C, P, FAMP</td>
<td></td>
<td>69e</td>
</tr>
<tr>
<td>759</td>
<td>M/65</td>
<td>Post III</td>
<td>NI</td>
<td>+</td>
<td>- B</td>
<td>7.1</td>
<td>-</td>
<td>CHOP</td>
<td>16.0e</td>
<td></td>
<td></td>
</tr>
<tr>
<td>829</td>
<td>F/34</td>
<td>Pre III</td>
<td>IE</td>
<td>+</td>
<td>- C</td>
<td>6.0</td>
<td>-</td>
<td>Surgery, M2</td>
<td>27.0+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>910</td>
<td>M/52</td>
<td>Post IIIE</td>
<td>NI, P</td>
<td>+</td>
<td></td>
<td>D</td>
<td>8.5</td>
<td>MBACOD, M2</td>
<td>56.0+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>912</td>
<td>F/55</td>
<td>Post IV</td>
<td>I</td>
<td>+</td>
<td>+ LF</td>
<td>-</td>
<td>10.2</td>
<td>IgMκ</td>
<td>C, P, FAMP, VAD</td>
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<td></td>
</tr>
<tr>
<td>941</td>
<td>M/55</td>
<td>Pre IV</td>
<td>I</td>
<td>-</td>
<td>+ LF, a</td>
<td>E</td>
<td>8.5</td>
<td>IgMκ</td>
<td>Surgery</td>
<td>41.0+</td>
<td></td>
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<tr>
<td>1020</td>
<td>F/87</td>
<td>Post IV</td>
<td>NI, P</td>
<td>-</td>
<td>+ LF, a</td>
<td>E</td>
<td>63.4</td>
<td>-</td>
<td>Surgery</td>
<td>36.0+</td>
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<td>1023</td>
<td>M/45</td>
<td>Post IV</td>
<td>I</td>
<td>+</td>
<td>+ LF, a</td>
<td>D</td>
<td>22.1</td>
<td>-</td>
<td>C, M2, FAMP</td>
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<td>M/55</td>
<td>Pre IV</td>
<td>I</td>
<td>+</td>
<td>+ LF</td>
<td>F</td>
<td>21.0</td>
<td>PrMOPP, FAMP</td>
<td>21.0+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1068</td>
<td>M/40</td>
<td>Pre IV</td>
<td>NI</td>
<td>+</td>
<td>- G</td>
<td>8.5</td>
<td>IgMκ</td>
<td>M2, CHOP, FAMP</td>
<td>86.0e</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1145</td>
<td>M/62</td>
<td>Pre IV</td>
<td>I</td>
<td>+</td>
<td>+ LF</td>
<td>-</td>
<td>11.0</td>
<td>C, P, FAMP</td>
<td>85.0e</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Abbreviations: Pre, cytogenetic analysis before cytotoxic therapy; Post, cytogenetic analysis posttreatment; BM, bone marrow; NI, not involved; NI, P, not involved at diagnosis, involved during progression; I, involved at diagnosis; LF, larger forms with prolymphocytoid features present as well as small lymphocytes; LF, a, larger forms included prolymphocytoid forms as well as atypical cells with indented nuclei, some with cytoplasmic projections; A, neck mass; B, lung lesions; C, breast mass; D, bowel/mesenteric mass; E, splenomegaly as dominant feature; F, skin involvement; G, chest wall mass; M2, cyclophosphamide, vincristine, CCNU, melphalan, prednisone; CMOPP, cyclophosphamide, vincristine, procarbazine, prednisone; CHOP, cyclophosphamide, daunorubicin, vincristine, prednisone; M2, cyclophosphamide, vincristine, procarbazine, prednisone; PrMOPP, same drugs as CHOP plus etoposide, nitrogen mustard, vincristine, prednisone, procarbazine; C, chlorambucil; P, prednisone; FAMP, fludarabine; MBACOD, CHOP + methotrexate, alemtuzumab, dexamethasone. Survival in months; e = expired; "+" = alive at last follow-up.

Leukocyte count (10^9/L) at diagnosis.
at the time of diagnosis in 3 cases of del(6q) sm lym NHL studied at initial diagnosis suggests that this morphology may present de novo in a subset of cases. The presence of prolymphocytic forms in the peripheral blood of 4 cases without del(6q) studied at the time of relapse suggests that subsets of “prolymphocytic transformation” of sm lym NHL may be associated with other cytogenetic abnormalities.

An increased proportion of prolymphocytes or other larger forms was suggested to denote a prognostic significance in some series of CLL/sm lym NHL, but not in others. No such prognostic significance of the presence of larger forms in tissue sections or peripheral blood was noted in the current series. Although there was a trend for a shorter median survival of the del(6q) cohort compared with cases with other abnormalities, the observation of continuous relapse in all cases and the heterogeneity of treatments precluded the assessment of the prognostic significance of del(6q). The survival of the del(6q) cohort (85 months) contrasts with the median survivals of 96 months for patients with CLL end of 4 to 36 months for PLL. Half of the del(6q) sm lym NHL patients succumbed despite intensive chemotherapy therapy regimen, highlighting the progressive nature of the disease. The recent introduction of fludarabine and related compounds has resulted in improved clinical responses in both CLL and PLL as well as HCL, this drug was of limited palliative benefit in 5 of 6 cases in this series.

The immunophenotype of many of the del(6q) sm lym NHL was intermediate between the more mature phenotype of IgG-expressing HCL/intermediate-grade NHL and the uniform mrbcR expression of CLL. The intensity of surface membrane Ig, a useful marker of the subset of CLL/PLL, could not reliably be assessed from the immunohistochemical preparations used in this study. The absence of expression of CD11c in these cases distinguished them from the previously described subset of CD5-expressing CLL/HCL, whereas the absence of TRAP positivity in those cases that showed morphologic features of HCL contrasted with recently described cases of CLL/PLL. The splenic presentation, involvement of both red and white pulp, and immunologic features of 1 of these cases (no. 941) was consistent with the description of splenic lymphoma with villous lymphocytes, although the prolymphocytic forms observed in the peripheral blood are not a typical feature of this entity.

The t(11;14)(q13;q32) translocation has been noted in 2 of 13 cases of PLL, in 2 cases of the “paraimmunoblastic variant” of sm lym NHL, as well as in diffuse intermediate differentiation lymphoma (DIL). Although noted in greater than 10% of diffuse low-grade NHL in this series, t(11;14) was observed in a single case with del(6q). This tumor demonstrated features of intermediate differentiation NHL and was the only t(11;14) case that presented in leukemic phase, suggesting a “secondary” role for the del(6q) in this instance. In both the sm lym NHL with and without del(6q), the additional cytogenetic aberrations del(13)(q14q31), +3, +18, +12, and +21 were observed. Each of these aberrations has been associated with the progression of NHL. Although tumors derived from mucosa-associated lymphoid tissues (MALT) were identified in the series, the recently described t(11;18)(q21;q21) associated with MALT NHL was not observed.

In addition to the 3 cases in the current study, prior large series have identified del(6q) as the solitary cytogenetic abnormality in 2 cases of sm lym NHL as well as in 5 cases of CLL. Although observed in 4% of CLL and 10% to 20% of PLL, del(6q) is a common feature of t(14;18) follicular NHL, in which it is thought to play a role in tumor progression. In a prior analysis, we attempted to compare the regions of 6q deletion in low-grade NHL with and without t(14;18). A larger region of deletion encompassing 6q23-27 was noted in the t(14;18) tumors, with a commonly deleted segment at 6q23 in low-grade NHL without t(14;18). Detailed loss of heterozygosity analysis in the 6q21-23 region will be necessary to determine more precisely the incidence, the association with other abnormalities, and the prognostic significance of del(6q) in diffuse low-grade NHL treated uniformly. Molecular characterization of del(6q)(q21q23) in the low-grade NHL described here will also facilitate identification of specific genetic alterations at this site. This report suggests an important pathogenetic role of del(6q) in sm lym NHL based on its high frequency in this histologic subset, and by the observation of del(6q) as the solitary cytogenetic abnormality in individual cases of diffuse low-grade lymphoid malignancy.

ACKNOWLEDGMENT

We are grateful to Kin Kong, Stephanie Alton, and Amelia Panico for their expert photographic assistance; to Judith Brydson and Marilyn Evans for secretarial assistance; to Drs Carol Portlock, David Strauss, Mark Weiss, and James P. O’Brien for providing clinical information regarding the cases; and to Susan McKenzie and Dr Gianluca Guidano for their comments on the peripheral blood morphologies.

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Clinical and morphologic features of B-cell small lymphocytic lymphoma with del(6)(q21q23)

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