Clinical Significance of the BCR-ABL Fusion Gene in Adult Acute Lymphoblastic Leukemia: A Cancer and Leukemia Group B Study (8762)


The Philadelphia (Ph1) chromosome, or its molecular counterpart, the BCR-ABL fusion gene, is a rare but important prognostic indicator in childhood acute lymphoblastic leukemia (ALL), but its impact on adult ALL has not been well ascertained. A prospective study of the BCR-ABL fusion gene was begun on patients entered on clinical trials conducted by the Cancer and Leukemia Group B (CALGB). All patients received intensive, multiagent chemotherapy that included daunorubicin. Over 2 years, 56 patients were studied for molecular evidence of a BCR-ABL gene using Southern blot and pulsed-field gel hybridization analysis. Results were compared with cytogenetic detection of a Ph1 chromosome, and clinical features were compared for the BCR-ABL–positive and –negative groups. Molecular methods detected the BCR-ABL gene in 30% of cases compared with cytogenetic detection of the Ph1 chromosome in only 23%. The majority of cases (76%) showed the p190 gene subtype similar to pediatric ALL: the BCR-ABL–positive cases displayed a more homogeneous immunophenotype than the BCR-ABL–negative cases and were predominantly CALLA positive (86%) and B-cell surface antigen positive (82%). The rate of achieving complete remission was similar in the BCR-ABL–positive and –negative groups (71% and 77%, respectively, P = .72). There were more early relapses in the BCR-ABL–positive group, resulting in a shorter remission duration that was especially marked in the CALLA-positive and B-cell antigen–positive populations. These preliminary data suggest that the impact of the BCR-ABL gene on clinical outcome in ALL may be on maintenance of complete remission (CR) rather than achievement of CR when aggressive, multiagent chemotherapy is used. This study identifies the BCR-ABL gene as an important factor in adult ALL and demonstrates the utility of molecular methods for its accurate diagnosis.

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THE BCR-ABL FUSION GENE, usually the result of the Philadelphia (Ph1) chromosome translocation1 is universally present in cases of chronic myelogenous leukemia (CML).2 Advances in molecular biology, especially in vivo studies of BCR-ABL gene activity,3,4,5 have highlighted the contribution of this gene to the development of acute leukemia as well. Acute lymphoblastic leukemia (ALL) has been of particular interest in this regard because the Ph1 chromosome is seen frequently in this disorder, which bears a close similarity to the lymphoid blast phase of CML. Acute lymphoblastic leukemia (ALL) has been the major diagnostic abnormality in adult acute lymphoblastic leukemia as well. Acute lymphoblastic leukemia (ALL) has been the major diagnostic abnormality in adult acute lymphoblastic leukemia.

In pediatric ALL, cytogenetic studies have demonstrated that the Ph1 chromosome is an important contributor to mortality though present in only 2% to 3% of cases.9,11 Recent studies have underscored this contribution, showing that the Ph1 chromosome continues to carry a poor prognosis12,13 even within the context of highly intensive, multiagent therapy for childhood ALL that has improved survival in other cytogenetic subgroups.14 In adult ALL, the clinical impact of the Ph1 chromosome has not been as well delineated as it has in childhood ALL, although it is estimated to have a much higher incidence in this age group.15 Earlier studies suggested a poor outcome,16 but there have been no prospective studies that accurately reflect the incidence or prognosis of this abnormality within the context of contemporary therapies. Furthermore, it has been suggested that the BCR-ABL fusion gene of childhood and adult ALL have a different molecular basis, with the BCR-ABL fusion gene in adult ALL of the “p210” subtype resembling that found in CML, whereas the childhood subtype is “p190.”17 The two subtypes differ in the amount of the BCR gene included in the fusion and may have different biologic activity. Thus, it is difficult to extrapolate from the pediatric experience to adult ALL.

We therefore initiated a prospective study to determine the incidence, molecular subtypes, and clinical outcome of the BCR-ABL fusion gene in newly diagnosed patients with adult ALL enrolled in CALGB clinical trials. We used methods of Southern hybridization and pulsed-field gel electrophoresis well-suited to small sample volumes that could reliably detect the BCR-ABL fusion gene, or confirm its absence, in greater than 90% of cases. We compared our results with cytogenetic diagnosis and analyzed associations with clinical and pathologic parameters.

MATERIALS AND METHODS

Fifty-six untreated patients with ALL who were 15 years of age or older were referred for molecular diagnostic studies as part of this prospective study: Cancer and Leukemia Group B (CALGB) 8762—Molecular Genetic Abnormalities in Adult Acute Lymphoblastic Leukemia. Forty-nine of these patients were treated on CALGB treatment protocols; 8 on CALGB 851316 and 41 on CALGB 8811.17 The protocols included intensive, five-drug induc

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tion chemotherapy that included daunorubicin. These 49 patients represent 41% of cases entered on these protocols from October 1, 1987 until February 1, 1990. The other seven patients were included in the molecular, cytogenetic, and immunophenotypic studies but were not treated on CALGB protocols because none were active at the time; all received standard treatment at their home institutions.

The diagnosis of ALL was confirmed by central review of cytologic and cytochemical features according to the French-American-British (FAB) criteria. Cytologic characteristics were examined on Wright-Giemsa-stained smears. Bone marrow or blood specimens were evaluated for myeloperoxidase, using diaminobenzidine substrate at pH 7.6, Sudan Black B, α-naphthyl acetate esterase, and with periodic acid–Schiff stain in standard cytochemical assays. All patients had fewer than 3% myeloperoxidase-positive blasts. Cases that could be subclassified into the L1 or L2 categories are included; those with L3 morphology, or Burkitt’s type leukemia/lymphoma, were excluded.

**Molecular detection of the BCR-ABL fusion gene.** Heparinized bone marrow aspirates or blood specimens were obtained before treatment and sent by overnight mail to a central reference laboratory at the University of Chicago Medical Center. Although bone marrow was requested for study on all cases, blood was accepted if marrow could not be provided either because the marrow was inaspirable or the sample was too scanty. A buffy coat was prepared, red blood cells were lysed by hypotonic ammonium chloride, and white blood cells were prepared directly into agarose plugs, each containing 1.6 million white blood cells (approximately 10 μg of DNA). The plugs were used for PFGE or for standard gel electrophoresis using previously described methods. Molecular diagnosis of the BCR-ABL fusion gene was performed by Southern blot and PFGE analysis as described previously. For every case, Southern blot analysis was performed by hybridization of probes bcr exon 2, bcr exon 4, and pAEB2.3 to BamHI, HindIII, and Bgl II digests; PFGE analysis was performed by hybridization of probes bcr exon 2 and pAEB2.3 to Not I digests. Translocation breakpoints are manifest as nongermline restriction bands within the BCR gene that are unique to individual cases. The presence of a new Not I BCR band is considered positive for the BCR-ABL fusion gene of any subtype. The molecular subtype is then specified by Southern blot analysis. Rearrangements detected with bcr exon 2 or bcr exon 4 probes specify the p210 subtype; rearrangement detected with probe pAEB2.3 or the absence of Southern blot rearrangement (in the presence of a positive PFGE finding) indicates the p190 subtype. PFGE and Southern hybridization to probes from the ABL gene were confirmatory, but isolated rearrangement was not considered positive in the absence of BCR findings.

**Cytogenetic analysis.** Cytogenetic analysis was performed as part of CALGB 8461, a prospective study of chromosomes in acute leukemia. Analysis of banded chromosomes from bone marrow or, if inaspirable, peripheral blood taken at diagnosis were performed in CALGB institutional cytogenetic laboratories. Specimens were generally processed using short-term unstimulated cultures (24 to 72 hours). G-banding was usually used. A minimum of 20 cells were analyzed in each case. Two karyotypes demonstrating each abnormal clone, or normal cells if the case lacked a clonal abnormality, were centrally reviewed. Karyotypes were designated according to the ISCN nomenclature.

**Immunophenotype determination.** Heparinized bone marrow aspirates and blood specimens were used to determine the surface antigen phenotype by means of monoclonal antibodies. The samples were sent by overnight courier to a reference laboratory at the University of California, San Diego Cancer Center. Cytofluorometric analysis, as previously described, and a panel of monoclonal antibodies was used for indirect immunofluorescence. The criteria for surface marker positivity was expression by at least 20% of the leukemic blast population. B-antigen expression was defined as B4 (CD19) or B1 (CD20) positivity; T-antigen expression as T101 (CD5) or T11 (CD2) reactivity; and myeloid antigen expression as MCS-2 (CD13), My7 (CD13), or MY9 (CD33) positivity. We defined B-lineage as expressing B antigens only (without T or myeloid); T-lineage as expressing T antigens only; myeloid lineage as expressing only myeloid antigens; mixed as being positive for myeloid antigens plus either B, T, or both markers; and miscellaneous as any other combination (B+T or no positive markers). CALLA antigen was assessed by J5 (CD10) expression.

**Statistical methods.** The study was designed to analyze the association between the BCR-ABL gene and morphotype, immunophenotype, karyotype, presenting clinical features, response to therapy, survival, and remission duration and to compare the concordance of molecular and cytogenetic methods.

Survival was defined as the time from study entry until the time of death or last follow-up and was measured for all patients. Remission duration was defined only for those achieving complete response (CR) as time from CR until relapse (either bone marrow or non–bone marrow), death, or last follow-up. Patients were censored for analysis of survival if they were alive at the date of last follow-up, and they were censored for analysis of remission duration if relapse or death had not occurred by the time of last follow-up. Because of possible treatment effects on survival and remission duration, differences in these endpoints between BCR-ABL groups were tested, controlling for treatment study, by using the stratified logrank test. Survival and remission duration probabilities were estimated by the Kaplan-Meier method. Comparison of the BCR-ABL groups with respect to surface markers, lineage, sex, organomegaly, lymphadenopathy, mediastinal mass, and response was based on Fisher’s exact test, whereas differences in age, leukocyte count (WBC), and hemoglobin were tested with the Wilcoxon rank sum statistic. A comparison of the proportion of BCR-ABL–positive cases found by molecular and cytogenetic detection methods was performed with McNemar’s test. All reported P values are based on two-sided tests of significance.

### RESULTS

**Detection of the BCR-ABL fusion gene.** The molecular methods used permitted diagnosis of the BCR-ABL fusion and its molecular subtype in 93% of patients (Table 1). The technique was well suited to the analysis of specimens that contained low numbers of cells (as few as 5 million cells) or cells of poor viability resulting from shipping delays, conditions that are unavoidable in multi-institutional studies. The BCR-ABL fusion gene was detected as a rearrangement within the BCR gene by PFGE, and the molecular

<table>
<thead>
<tr>
<th>Molecular No. (%)</th>
<th>Cytogenetic No. (%)</th>
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<tbody>
<tr>
<td>Total cases studied</td>
<td>56</td>
</tr>
<tr>
<td>BCR-ABL positive</td>
<td>17 (30)</td>
</tr>
<tr>
<td>bcr positive (p210)</td>
<td>4 (24)</td>
</tr>
<tr>
<td>bcr negative (p190)</td>
<td>13 (76)</td>
</tr>
<tr>
<td>5 PFGE only</td>
<td>—</td>
</tr>
<tr>
<td>7 PFGE and Southern blot</td>
<td>—</td>
</tr>
<tr>
<td>1 Southern blot only</td>
<td>—</td>
</tr>
<tr>
<td>BCR-ABL negative</td>
<td>35 (63)</td>
</tr>
<tr>
<td>Not evaluable</td>
<td>4 (7)</td>
</tr>
<tr>
<td>BCR-ABL positive/evaluable</td>
<td>0.33</td>
</tr>
</tbody>
</table>
subtypes was specified by Southern analysis. In Ph1-positive
ALL, chromosome 22 translocation breakpoints occur in
one of two regions of the BCR gene: the 5.8-kb break-
point cluster region, which is detected by Southern blot
using bcr exon 2 and 4 probes, or the 70-kb first intron of the
BCR gene. Translocations within the breakpoint cluster
region usually produce the p210 protein, similar to that
seen in CML, whereas the latter produces the p190 protein.

The results of molecular studies are summarized in Table
1. Of 56 cases enrolled on CALGB study 8762, 52 had all
molecular tests successfully completed. This included 36
bone marrow and 16 peripheral blood specimens. Seventen
cases (30% of the total cases, or 33% of evaluable
cases) were BCR-ABL positive. Of these 17, only one-
fourth showed findings consistent with the p210 subtype;
the remainder were of the p190 subtype.

Of the molecular methods used, PFG was the single most
successful, detecting 15 of the 17 cases (88%), whereas
Southern blot analysis detected 12 of 17 cases (71%),
including 2 that were missed by PFG (one p210 and one
p190). Of interest, Southern hybridization with pAEB2.3
detected 8 of the 13 p190 cases, further confirming that
these breakpoints tend to cluster within this region. An
ABL rearrangement was detected in 10 of the BCR-ABL–
positive cases (59%), as well as in one BCR-ABL–negative
case that had a cytogenetic Ph1 chromosome (see below);
no other ABL rearrangements were detected, although
extensive ABL analysis was not performed.

Cytogenetic detection of Ph1. Of 53 cases sent for cytoge-
enetic analysis, 44 were specimens of bone marrow and 9
were peripheral blood. Complete karyotypes with adequate
banding could be obtained on 40 cases (71%). A higher
cytogenetic success rate was obtained when the analysis was
limited only to the presence or absence of a Ph1 chromo-
some. When reviewed in this manner, 45 of 53 cases (85%)
produced evaluable metaphases, and the results are shown in
Table 1. Of the 53 cases, 12 (23%) were Ph1 positive. If
only the evaluable cases are considered, the incidence of
Ph1 chromosome is 12 of 45 (27%). Cytogenetic analysis
was more likely to fail in peripheral blood (4 failures of 9
specimens) than in marrow (4 failures of 44 specimens).

We directly compared molecular and cytogenetic meth-
ods by analyzing the 53 cases that had samples sent for both
studies. Molecular methods detected 16 BCR-ABL–
positive cases, about one-third more than did cytogenetics
(12 cases). When both methods were completely evaluable
(43 cases), agreement was good (P = .18), but there were
five discrepancies. Four of these discrepancies were the
result of a BCR-ABL fusion that was missed by cytogenet-
ics, and one was a Ph1-positive case that was missed by
molecular analysis.

These five discrepant cases were analyzed in detail. Of
the four cases with negative cytogenetic studies, there were
two in which no cytogenetic abnormality was reported. One
case (41081) had a marrow sample analyzed. Although
banding was poor and the karyotype was incomplete, a Ph1
chromosome was not present. The other case (43419) had
a normal karyotype, but only peripheral blood was available
for cytogenetics. Because the total white blood count was
low (only 3.3 × 109/L) it is possible that the only met-
aphases obtained actually originated in residual normal
cells. On molecular evaluation, both cases showed a Southern
blot rearrangement with pAEB2.3 (Fig 1A and B) as
well as a positive PFG result (not shown).

Two other cases demonstrated the BCR-ABL fusion, but
did not show the Ph1 chromosome, although cytogenetic
studies were adequate. Case 44503 had the karyotype
46,XY,t(9;13) (q34;q12), suggesting a breakpoint on chromo-
some 9 at the ABL locus, but because both chromo-
somes 22 were normal, it could not be classified as Ph1
positive. Molecular results demonstrate a bcr rearrange-
ment with the ABL probe (Fig 2E) but no BCR
rearrangement with either Southern or PFG analysis, and
thus did not meet the criteria for BCR-ABL fusion. By our
choice of probes and enzymes, an ABL rearrangement is
not consistently demonstrated when a BCR-ABL fusion is
present, so we have not included it in our criteria. On the
other hand, we have never observed an ABL rearrange-
ment in normal cells or in leukemia cells that are Ph1 or
BCR-ABL negative by other criteria. This case remains
unconfirmed for a BCR-ABL gene but, if present, it would
represent a false negative of molecular analysis and would
support an expansion of our diagnostic criteria.

Immunophenotype and FAB class. Of 52 submitted cases,
46 were evaluable for immunophenotyping, and the results
are shown in Table 2. Twenty-nine of 46 (63%) expressed B
antigens, including 14 of 15 BCR-ABL–positive cases.
Expression of T antigens was less common in the BCR-ABL–
positive than in the −negative cases and never occurred in
the absence of B antigens. Ten of the 14 B-antigen–
positive cases with the BCR-ABL gene expressed only B antigens,
resulting in a B-lineage phenotype; 4 showed a mixed
lineage phenotype that included B antigens (2 My+ B+ and
2 My− B+ T+), and 1 had no surface markers positive.
There were no pure myeloid-lineage or T-lineage cases of
BCR-ABL–positive ALL. Viewed from another perspec-
tive, the BCR-ABL gene is present in half of the cases of
adult ALL with B-cell surface markers, but it is found in

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Fig 1. Molecular findings of the discrepant cases. (A) Southern blot, Bgl II restriction digest hybridized to probe a (pAEB2.3). Lane 1, case 41081, Ph' negative by cytogenetics (discrepant case); lane 2, case 41075, unevaluable cytogenetics. Lane 2 shows the normal restriction pattern with a band of size 10 kb, whereas lane 1 shows a rearranged band (arrow) of size 14 kb, constituting a positive BCR-ABL gene of p190 subtype. (B) Southern blot, BamHI restriction digest, hybridized to probe a (pAEB2.3). Lane 1, case 43419, Ph'-negative cytogenetics (discrepant case); lane 2, case 43368, Ph'-positive by cytogenetics (concordant case, with molecular tests positive for p210, not shown here). Lane 1 demonstrates the common polymorphism seen with this probe, of sizes 5.5 kb and 4.4 kb (shown by asterisks), and a rearranged band of size 10 kb indicated by the arrow. In this case, the smaller (4.4 kb) allele is rearranged, constituting a positive BCR-ABL gene of p190 subtype. In lane 2, a single band of size 5.5 kb is shown, and the case is thus not polymorphic. (C) Southern blot, Bgl II restriction digest, hybridized to probe b (bcr exon 2). Lane 1, case 44518, Ph'-positive by cytogenetics (concordant case, with molecular tests positive for p210, not shown here); lane 2, case 44503, Ph'-negative by cytogenetics (discrepant case). Lane 1 shows the normal restriction pattern with a band of 5.5 kb, whereas Lane 2 shows a rearranged band of size 4.8 kb (arrow), constituting a positive BCR-ABL gene of subtype p210. (D) PFG blot, Not I restriction digest hybridized to probe c (bcr exon 4). Lane 1, case 42265, Ph' negative by cytogenetics (discrepant case); lane 2, SUP-B13 cell line used as a positive control (BCR-ABL of p190 subtype). Both lanes show the two polymorphic BCR Not I bands of size 1.800 and 450, as well as a positive band signal from undigested DNA in the "compression zone," at which all fragments larger than 1,000 kb migrate. Rearrangements are seen in lanes 1 and 2 (arrows), constituting a positive BCR-ABL gene; this test does not specify the subtype, which in both cases is p190 (data not shown). (E) Pulsed field gel blot, Not I restriction digest, hybridized to probe pHabl5'. Lane 1, case 43217, Ph' positive by cytogenetics (discrepant case); lane 2, SUPB-13 cell line here used as a negative control (SUP-B13 is BCR-ABL positive, but rearrangements are only seen with the BCR probe, Fig 2D, lane 2). The SUPB-13 cell line shows only a germline band with this probe, but lane 1 shows a new band of approximately 400 kb.

less than 6% of cases with T-cell surface markers and in none of the cases that classify as pure T-lineage or pure myeloid lineage. The preponderance of BCR-ABL-positive cases among those with B-cell surface markers is statistically significant compared with BCR-ABL-negative cases (P = .01).

Although one might have predicted that myeloid antigen expression would be more common in the p210 cases because of their molecular similarity to CML, this was not the case. Of the four p210 cases that were immunophenotyped, three were pure B lineage and one was negative for surface markers, but none of them expressed myeloid antigens. The four BCR-ABL cases with a mixed-lineage phenotype were all of the p190 subtype.

CALLA was another marker that was frequently present in the BCR-ABL group. Thirty-four of 45 evaluable cases were CALLA positive, including 12 of 14 BCR-ABL positives; the BCR-ABL cases thus comprised 35% of the total CALLA-positive population. There were no statistical differences in numbers of CALLA positive and negative cases between the two BCR-ABL groups (P = .28). Likewise, there were no apparent differences between the FAB subtypes of the BCR-ABL-positive and -negative groups. Of the 52 cases, 5 (29%) of the BCR-ABL positive and 14 (40%) of the BCR-ABL negatives were L1, 9 (53%) of the positives and 18 (51%) of the negatives were each L2, and the remainder were unclassifiable. The differences in numbers of L1 and L2 cases in the BCR-ABL groups are not statistically significant (P = .75).

Clinical features and treatment outcome. Presenting clinical features for the 52 patients for whom this information is available are shown in Table 3. There were no significant
Clinical follow-up is available for the 45 patients fully evaluable by molecular methods who received treatment on CALGB protocols (8 on CALGB 8513 and 37 on CALGB 8811). When stratified by treatment protocol, the overall CR rate of 76% was nearly identical for both the BCR-ABL-positive and -negative patients (71% and 77%, respectively; Table 4). There were more early failures in the BCR-ABL-positive cases (Fig 2); there was a trend toward a longer remission duration in the BCR-ABL-negative cases, but it did not reach statistical significance ($P = .06$). Median follow-up for patients still at risk of relapse is 15.5 months. The survival (Fig 3) is also not significantly different between the BCR-ABL-positive and -negative groups (median survival, 11.1 and 21.9 months, respectively; overall survival, $P = .26$). The median follow-up for those patients still alive is 23 months.

Because the CALLA-positive and the B-cell surface antigen-positive subgroups contained a high proportion of BCR-ABL cases, they were examined separately for differences in clinical outcome. Within each group, the BCR-ABL cases showed a much shorter CR duration (Table 4B.

Table 3. Presenting Clinical and Laboratory Characteristics of BCR-ABL-Positive and BCR-ABL-Negative Cases

<table>
<thead>
<tr>
<th>BCR-ABL+ (n = 17)</th>
<th>BCR-ABL- (n = 35)</th>
<th>$P$ Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median age (range)</td>
<td>39 (17-80)</td>
<td>37 (16-71)</td>
</tr>
<tr>
<td>% Male</td>
<td>65</td>
<td>51</td>
</tr>
<tr>
<td>Lymphadenopathy</td>
<td>18%</td>
<td>43%</td>
</tr>
<tr>
<td>Mediastinal mass</td>
<td>0%</td>
<td>20%</td>
</tr>
<tr>
<td>Splenomegaly</td>
<td>29%</td>
<td>37%</td>
</tr>
<tr>
<td>Hepatomegaly</td>
<td>24%</td>
<td>20%</td>
</tr>
<tr>
<td>Median hemoglobin (g/100 mL)</td>
<td>10.5</td>
<td>9.8</td>
</tr>
<tr>
<td>Median WBC ($\times10^9$/L)</td>
<td>26.3</td>
<td>17.5</td>
</tr>
</tbody>
</table>

$P = .03$ by Fisher’s exact test for a 5 $\times$ 2 contingency table.

$P = .28$ by Fisher’s exact test.
Table 4. Clinical Outcome for Patients Treated on CALGB Protocols 8513 and 8811

<table>
<thead>
<tr>
<th></th>
<th>BCR-ABL+ (n = 14)</th>
<th>BCR-ABL- (n = 31)</th>
<th>P Value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>A. All Cases</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Complete remission</td>
<td>10 (71%)</td>
<td>24 (77%)</td>
<td>.72</td>
</tr>
<tr>
<td>Median remission duration (mo)</td>
<td>10.0</td>
<td>18.4</td>
<td>.06</td>
</tr>
<tr>
<td>Median survival (mo)</td>
<td>11.2</td>
<td>21.8</td>
<td>.26</td>
</tr>
<tr>
<td>B. CALLA-Positive Cases</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Complete remission</td>
<td>9 (75%)</td>
<td>18 (61%)</td>
<td></td>
</tr>
<tr>
<td>Median remission duration (mo)</td>
<td>9.5</td>
<td>NA†</td>
<td>.003</td>
</tr>
<tr>
<td>Median survival (mo)</td>
<td>11.1</td>
<td>NA†</td>
<td>.15</td>
</tr>
<tr>
<td>C. B Antigen-Positive Cases</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Complete remission</td>
<td>9 (65%)</td>
<td>10 (67%)</td>
<td></td>
</tr>
<tr>
<td>Median remission duration (mo)</td>
<td>9.1</td>
<td>NA†</td>
<td>.03</td>
</tr>
<tr>
<td>Median survival (mo)</td>
<td>11.4</td>
<td>12.5</td>
<td>.64</td>
</tr>
</tbody>
</table>

*Logrank, stratified by treatment.
†Median not yet achieved.

and C). The CALLA-positive subgroup, which comprised 82% of the entire study population, showed especially striking differences. Remission duration was considerably longer in the BCR-ABL-negative cases (P = .003); median CR duration in BCR-ABL positives was 9.5 months, whereas 60% of the negative cases are still in CR at 2 years (Fig 3). It is clear from Figs 2 and 3 that the observed differences are primarily the result of better outcome in the comparison group. These results suggest that the CALLA-positive, BCR-ABL-negative subgroup may be a subgroup of adult ALL that does considerably better than others; this observation warrants confirmation in a larger study.

DISCUSSION

The present study is an accurate portrayal of the incidence of BCR-ABL-positive cases in an unselected population of adult ALL. Molecular analysis allowed the accurate specification of BCR-ABL-negative as well as BCR-ABL-positive groups, which facilitated clinical comparisons. Although the population is small and follow-up is short, there are intriguing differences in clinical outcome that warrant further investigation.

The BCR-ABL fusion gene appears to be a common finding in adult ALL, being present in one-third of all cases and fully one-half of cases with B-cell surface markers. Previous studies suggested a 20% incidence, but these were based on cytogenetics, which, as we have shown, tends to underestimate the prevalence of this abnormality. Not surprisingly, these BCR-ABL cases present a more homogeneous phenotype than the comparison BCR-ABL-negative groups. The majority of BCR-ABL-positive cases demonstrated B-cell markers and were CALLA positive, with a notable lack of T-lineage cases and myeloid cases. This is similar to findings in pediatric ALL, in which T-cell cases made up only 9% of Ph1-positive cases. There were no other important differences in presentation except for a complete absence of mediastinal mass in the BCR-ABL-positive group, also noted in pediatric ALL, again reflecting the lack of T-cell phenotype.

An unexpected result is that the majority of cases show a molecular rearrangement consistent with the p190 subtype. Because of the small number of p210 cases, it was not possible to determine if there are clinical differences between these two subgroups; this question will require a larger study to answer it. It had previously been assumed that the p210 is more common in adult ALL, and the p190 more common in children, but these rates have never been examined prospectively. The distribution of 23% p210 and 77% p190 in adults reported here is similar to 15% and 85%, respectively, reported in pediatric ALL by Suryanarayan et al using PCR. These molecular and clinical findings underscore the similarities of adult and pediatric ALL.
Phl-positive ALL, which share more features with each other than with other types of ALL in the same age groups, supporting a view that BCR-ABL-positive ALL is a single clinical entity with a broad age range.

Previous studies in adult\textsuperscript{30,31} and pediatric\textsuperscript{11,12,13} ALL show that the presence of a Ph\textsuperscript{1} chromosome is associated with low remission rates and shortened survival; these findings have been especially marked in pediatric ALL, in which long-term survival is frequent among the Ph\textsuperscript{1}-negative cases. The results of the present study differ somewhat, as the aggressive, multiagent chemotherapy that was used produced a high overall remission rate of greater than 70\% in both the BCR-ABL-positive and BCR-ABL-negative groups. Despite this, the BCR-ABL cases showed a trend toward shorter remission duration that was especially marked in certain subgroups; in the CALLA-positive cases; in particular, the BCR-ABL marker identified cases with a short remission duration among those that otherwise would appear to do extremely well. This observation warrants further study, as the CALLA-positive cases make up a sizable majority of adult ALL cases. The shortened remission duration has not yet translated into statistically significant survival differences, but follow-up is still early.

The present study underscores the limitations of cytogenetics for the diagnosis of the BCR-ABL gene in ALL; adequately banded metaphases are difficult to obtain and, even when adequate, may fail to detect a Ph\textsuperscript{1} chromosome. The finding of a positive molecular test for the BCR-ABL fusion in the face of Ph\textsuperscript{1}-negative cytogenetics has been well documented in CML,\textsuperscript{7} and this emphasizes the importance of confirmatory molecular analysis. Several molecular methods have been used to detect the BCR-ABL fusion in ALL,\textsuperscript{8} including PFG,\textsuperscript{25} polymerase chain reaction (PCR),\textsuperscript{29,32-33} and fluorescence in situ hybridization.\textsuperscript{34-36} Southern blot analysis as used in CML is inadequate because breakpoints occur outside of the 5.8-kb breakpoint cluster region, in an area much larger than can be surveyed by standard methods. Results obtained with any acceptable molecular method should be similar and allow comparisons with other clinical trials. Although PCR is now the preferred method for speed and cost considerations, the PFG approach used here has been shown to be comparable.\textsuperscript{33}

In summary, this study demonstrates that the BCR-ABL fusion gene is a common finding in adult ALL, and defines a group of pre-B-cell ALL with similar features in all age groups. The effect on clinical outcome depends somewhat on the comparison group, but these preliminary results suggest that the BCR-ABL cases are easily induced into remission with aggressive chemotherapy and fail because of early relapse. If so, this group might benefit by early intensified treatment, such as allogeneic bone marrow transplantation. It will be necessary to test this hypothesis with a larger population, ideally combined with close monitoring by molecular methods to elucidate the effects of treatment and the extent of residual disease, to confirm remission, and to predict relapse.

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