

CORRESPONDENCE

bcr BREAKPOINT AND PROGNOSIS OF CHRONIC PHASE CHRONIC MYELOID LEUKEMIA

To the Editor:

In a recent article, Morris et al¹ concluded that the location of breakpoints within the breakpoint cluster region (BCR) of chromosome 22 does not influence the duration of chronic phase or survival in chronic myeloid leukemia (CML). This conclusion conflicted with two previous independent studies^{2,3} from our laboratories that associated a 3' breakpoint with shortened chronic phase duration, but was consistent with other early reports^{4,5} that failed to note any such association.

Morris et al¹ base their conclusion on the analysis of 80 patients selected from a larger population (118 patients) who underwent leukapheresis between 1977 and 1982. They acknowledge that they may have missed a subset of patients with early conversion to blast crisis because patients with aggressive disease were excluded from the autotransfusion program. Clinical information was collected after the completion of molecular studies and excluded patients with inadequate clinical or molecular information. Therefore, it is quite likely that additional patients with early disease progression would be excluded because these patients would be the ones for which follow-up information would be hardest to obtain.

The possibility that ascertainment bias strongly influenced this study is suggested by the unusually long median survival data. For the 80 patients included in the study, median survival appeared to be approximately 53 months while median survival in CML is generally considered to be 36 to 48 months in most studies.⁶ A further indication that the population studied was atypical is indicated by the median age of patients, which was approximately 37 years at

diagnosis. The median age of CML at diagnosis is generally considered to be 50 years and, moreover, there is evidence for longer survival in younger patients.⁷

The initial studies that associated 3' breakpoints with shorter disease duration have been expanded^{8,9} and our overall conclusions remain the same. We suggest that the study by Morris et al¹ does not, in fact, "closely approximate a prospective study" and it should not be concluded on the basis of their paper that chronic phase duration and survival are uninfluenced by *bcr* breakpoint location. A true prospective study that includes a population of all newly diagnosed cases of CML is probably required to resolve this controversy.

We have previously pointed out¹⁰ that the consequences of a particular breakpoint may be complex in terms of the amount and types of transcripts produced. Investigation of the nature of different *bcr-abl* fusion products arising from the various possible breakpoints and/or alternative splicings of the *bcr-abl* gene transcripts may ultimately be of considerable importance in understanding clinical variation in CML.

PETER BENN

University of Connecticut Health Center
Farmington, CT

GEORGE D. BIRNIE

ELAINE MACKENZIE

The Beatson Institute for Cancer Research

KEN I. MILLS

Leukemia Research Fund Laboratories
Glasgow, Scotland

REFERENCES

- Morris SW, Daniel L, Ahmed CMI, Elias A, Lebowitz P: Relationship of *bcr* breakpoint to chronic phase duration, survival and blast crisis lineage in chronic myelogenous leukemia patients presenting in early chronic phase. *Blood* 75:2035, 1990
- Eisenberg A, Silver R, Soper L, Arlin Z, Coleman M, Bernhardt B, Benn P: The location of breakpoints within the breakpoint cluster region (*bcr*) of chromosome 22 in chronic myeloid leukemia. *Leukemia* 2:642, 1988
- Mills KI, MacKenzie ED, Birnie GD: The site of the breakpoint within the *bcr* is a prognostic factor in Philadelphia-positive CML patients. *Blood* 72:1237, 1988
- Dreazen O, Berman M, Gale RP: Molecular abnormalities of *bcr* and *c-abl* in chronic myelogenous leukemia associated with a long chronic phase. *Blood* 71:797, 1988
- Shtalrid M, Talpaz M, Kurzrock R, Kantarjian H, Trujillo J, Gutterman J, Yoffe G, Blick M: Analysis of breakpoints within the *bcr* gene and their correlation with the clinical course of Philadelphia-positive chronic myelogenous leukemia. *Blood* 72:485, 1988
- Champlin RE, Golde DW: Chronic myelogenous leukemia: Recent advances. *Blood* 65:1039, 1985
- Silver RT: Chronic myeloid leukemia: A perspective of the clinical and biologic issues of the chronic phase. *Hematol Oncol Clin North Am* 4:319, 1990
- Mills KI, Hynds SA, Burnett AK, MacKenzie ED, Birnie GD: Further evidence that the site of the breakpoint in the major breakpoint cluster region (M-*bcr*) may be a prognostic indicator. *Leukemia* 3:837, 1989
- Grossman A, Silver RT, Arlin Z, Coleman M, Camposano E, Gascon P, Benn PA: Fine mapping of chromosome 22 breakpoints within the breakpoint cluster region (*bcr*) implies a role for *bcr* exon 3 in determining disease duration in chronic myeloid leukemia. *Am J Hum Genet* 45:729, 1989
- Birnie GD, Mills KI, Benn P: Does the site of the breakpoint on chromosome 22 influence the duration of the chronic phase in chronic myeloid leukemia? *Leukemia* 3:545, 1989

RESPONSE

The letter of Benn et al raises several issues that they contend may have biased our analysis of the relationship of *bcr* breakpoint location to chronic phase duration and survival in patients with chronic myelogenous leukemia (CML). To address these points, we

systematically reviewed our data to determine if the manner in which our patient population was obtained or the study performed may have led to spurious results.

As mentioned in our paper,¹ 37 of the 118 patients studied for *bcr*

breakpoints were excluded from our final analysis, leaving 81 patients in whom complete molecular and clinical information was available. Sixteen of these 37 patients were excluded because they were either *bcr*-negative (6 patients) or because the natural history of their disease was interrupted by allogeneic bone marrow transplantation in early chronic phase (7 patients) or by early death from causes unrelated to CML (3 patients). Obviously, this group of patients would have been excluded from final analysis in any type of study attempting to relate the course of *bcr*-positive disease to the *bcr* breakpoint location. The remaining 21 patients were excluded from consideration because we were unable to obtain complete clinical data on them after determining their breakpoint location. Of these cases only one was truly lost to physician follow-up. Clinical information on the remaining 20 was existent, but unobtainable for the following reasons: (1) loss or misplacement of leukapheresis records, or lack of information in these records on referring hospitals or physicians or patient addresses; (2) misplacement of hospital records or hospital policy prohibiting our review of these records; and (3) unavailability of outpatient records because of retirement or relocation of private physicians. Thus, no suppositions as to the course of disease in these patients can be made simply on the basis of lack of complete clinical data on them. Moreover, even if one makes the assumption that these 21 cases all had aggressive disease, the fact that the distribution of breakpoints among them does not differ significantly from that of the 81 patients analyzed ($P = .94$, χ^2 test) suggests that it is highly unlikely that their inclusion would have altered our published results.

The low median age of the patients analyzed in our study reflects the preferential referral of younger patients for eventual autologous transfusion after aggressive therapy to reverse blast crisis. As mentioned in our paper, we were especially interested in studying young patients in early chronic phase who might be candidates for curative allogeneic transplantation because knowledge of disease prognosis could be useful in the management of such cases,² especially in suboptimal situations in which HLA-identical sibling donors are not available.

Although patients were not segregated by age in our paper, we took age into account in reexamining our data to better address the relationship of age, *bcr* breakpoint location, and CML course. Of 79 patients in whom the impact of breakpoint upon disease was reexamined (two patients were excluded because they fell in the rare subgroup with breakpoints in *bcr* subregion 4), 58 were less than 45 years of age. Median chronic phase duration and survival in this group (46 and 53.5 months, respectively) were somewhat longer than expected for CML patients as a whole, but are in line with figures reported for younger patients.³ Thus, we feel this group is a

representative sampling of CML patients under 45 years of age. Analysis of this group of patients failed to show any correlation between median duration of chronic phase or survival and *bcr* breakpoint location ($P = .96$ and $.95$ for median chronic phase duration and survival, respectively; log rank test).

On the other hand, the 21 patients 45 years of age and older had an unusually long median chronic phase duration and survival (79 months for both) and therefore appear to represent a biased patient sampling. Closer examination of this group showed seven patients with lags of 2 or more years between diagnosis and leukapheresis and prolonged median chronic phase and survival durations (90 and 93 months, respectively) as compared with the remaining 14 patients in this age group. These 14 patients had median chronic phase and survival durations of 42 and 48.5 months, respectively, and appear to represent more typical CML cases. Analysis of the patients 45 years of age and older either as a complete group or without the seven "lag patients" also failed to show any correlation between *bcr* breakpoint location and clinical course ($P = .89$ and $.61$ for median chronic phase duration and $P = .85$ and $.39$ for survival; log rank test).

Based upon our initial report and this reexamination of our data, we continue to believe that *bcr* breakpoint analysis as currently performed by Southern blotting cannot be used reliably as a prognostic indicator in CML patients presenting in early chronic phase. Moreover, five recently published studies⁴⁻⁸ in addition to the two older studies noted in our original report support our conclusion. We believe that the natural history of CML is more likely determined by unrecognized forms of alternative splicing of *bcr-c-abl* messenger RNA (mRNA) resulting in subtle differences in protein structure, other abnormalities in *bcr-c-abl*,^{9,10} or abnormalities accumulated in other genes as disease progresses.¹¹ Nevertheless, we wholeheartedly agree with Benn et al that a large, prospective study of newly diagnosed CML patients is required to resolve the issue of CML course and prognosis in relation to *bcr-c-abl* structure and expression, preferably one with mRNA analysis to address the role of alternative splicing.

STEPHAN W. MORRIS

MICHAEL L. HANCOCK

Departments of Hematology-Oncology and Biostatistics
St Jude Children's Research Hospital
Memphis, TN

PAUL LEBOWITZ

Department of Internal Medicine
Yale Medical School
New Haven, CT

REFERENCES

- Morris SW, Daniel L, Ahmed CMI, Elias A, Lebowitz P: Relationship of *bcr* breakpoint to chronic phase duration, survival and blast crisis lineage in chronic myelogenous patients presenting in early chronic phase. *Blood* 75:2035, 1990
- Thomas E, Clift, RA: Indications for marrow transplantation in chronic myelogenous leukemia. *Blood* 73:861, 1989
- Sokal JE, Baccarani M, Tura S, Fiacchini M, Cervantes F, Rozman C, Gomez GA, Galton DAG, Cannellos GP, Braun TJ, Clarkson BD, Carbonell F, Heimpel H, Extra JM, Fiere D, Nissen NI, Robertson JE, Cox EB: Prognostic discrimination among younger patients with chronic myelogenous leukemia: Relevance to bone marrow transplantation. *Blood* 66:1352, 1985
- Przepiorka D: Breakpoint zone of *bcr* in chronic myelogenous leukemia does not correlate with disease phase or prognosis. *Cancer Genet Cytogenet* 36:117, 1988
- Dyck JA, Bosco JJ: Clinical stage of chronic granulocytic leukemia and BCR breakpoint location in South-East Asian patients. *Br J Haematol* 72:64, 1989
- Ogawa H, Sugiyama H, Soma T, Masoaka T, Kishimoto S: No correlation between locations of *bcr* breakpoints and clinical states in Ph⁺-positive CML patients. *Leukemia* 3:492, 1989
- Jaubert J, Martiat P, Dowding C, Ifrah N, Goldman JM: The position of the M-BCR breakpoint does not predict the duration of chronic phase or survival in chronic myeloid leukemia. *Br J Haematol* 74:30, 1990
- Tien HF, Wang CH, Chen YC, Shen MC, Wu HS, Lee FY, Chuang SM, Liu CH: Chromosome and *bcr* rearrangement in chronic myelogenous leukemia and their correlation with clinical states and prognosis of the disease. *Br J Haematol* 75:469, 1990

9. Romero P, Berai M, Shtalrid M, Anderson B, Talpaz M, Blick M: Alternative 5' end of the *bcr-abl* transcript in chronic myelogenous leukemia. *Oncogene* 4:93, 1989
10. Shore SK, Bogart SL, Reddy EP: Activation of murine *c-abl*

- proto-oncogene: Effect of a point mutation on oncogene activation. *Proc Nat Acad Sci USA* 87:6502, 1990
11. Bernstein R: Cytogenetics of chronic myelogenous leukemia. *Semin Hematol* 25:20, 1988
-