To the editor:

GATA3 risk alleles are associated with ancestral components in Hispanic children with ALL

In this issue of *Blood*, Migliorini et al identify rs3824662 in *GATA3* as a novel susceptibility locus for childhood acute lymphoblastic leukemia (ALL) via genome-wide association analyses of European children.[1] Their results suggest that rs3824662 is strongly associated with nonhyperdiploid B-cell ALL (B-ALL), especially those cases lacking the TEL-AML1 fusion and having later age at diagnosis. Importantly, they and others have also linked the *GATA3* region to poorer ALL outcomes.[1,2] Hispanic children have a greater incidence of ALL,[3] a later average age at diagnosis,[4] and increased relapse compared with white children.[5] We therefore investigated whether single nucleotide polymorphisms (SNPs) in *GATA3* increase risk of ALL in Hispanics and whether they are associated with genetic

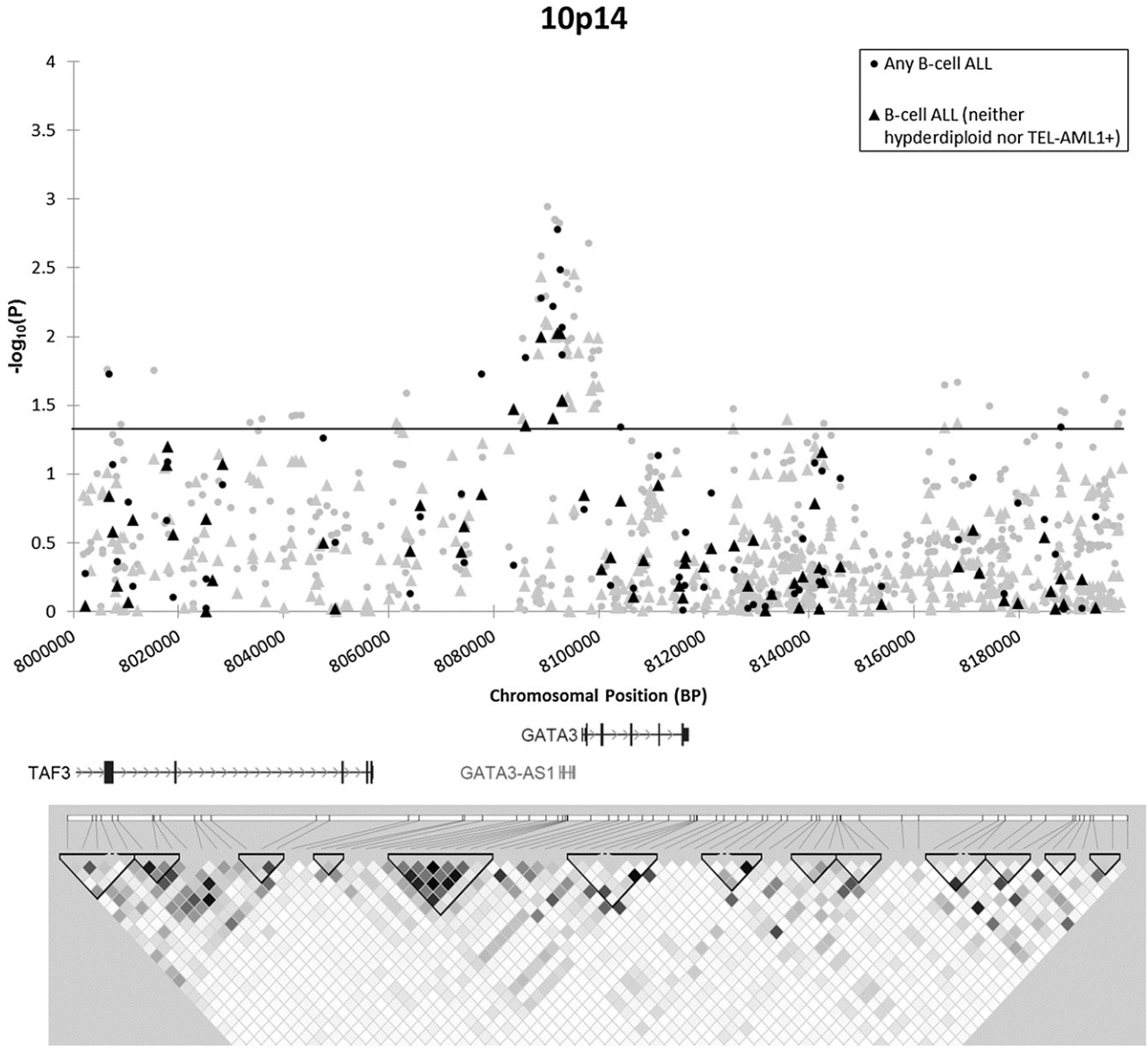

Figure 1. Association of SNPs in the *GATA3* locus with B-ALL risk among Hispanic children, by subtype. Association of 68 directly genotyped SNPs (black) and 477 imputed SNPs (gray) with B-ALL risk, adjusted for the first 5 principal components. Circles denote associations for B-ALL cases compared with controls. Triangles denote associations for nonhyperdiploid B-ALL cases without TEL-AML1 fusion compared with controls. Haplotype structure in the Hispanic sample appears below gene names, with darker shading indicating higher R^2 values and greater correlation between SNP genotypes. The strongest association in the Hispanic samples is at rs1271899 located in haplotype block 5. The strongest association from Migliorini et al,[1] rs3824662, is located between blocks 5 and 6 and is weakly but significantly linked to rs1271899 (R^2, 0.10; $P < .0001$).

ancestry by using genome-wide SNP data from 297 Hispanic children with B-ALL (97 hyperdiploid, 39 TEL-AML1 fusion–positive, 118 nonhyperdiploid/non-TEL-AML1 fusion–positive, 43 unknown molecular subtype) and 454 Hispanic controls from the California Childhood Leukemia Study.[6]

We found the most strongly associated variant from Migliorini et al (rs3824662; $P = 8.62 \times 10^{-12}$) has a significant effect in the same direction in our data ($P = .046$; odds ratio [OR], 1.23). The magnitude of this effect was not stronger in our nonhyperdiploid, non-TEL-AML1 fusion–positive cases (OR, 1.25). In total, 11 of 68 genotyped SNPs across the *GATA3* region were associated with B-ALL in our sample ($P < .05$) (Figure 1), with the strongest association at rs1271899 (OR, 0.62; 95% confidence interval [CI], 0.46 to 0.84; $P = 1.7 \times 10^{-3}$). rs1271899 was also the most strongly associated variant when analysis was limited to nonhyperdiploid, non-TEL-AML1 fusion–positive cases, in whom it showed a slightly stronger effect than among all B-ALL (OR, 0.59; 95% CI, 0.40 to 0.88; $P = 9.5 \times 10^{-3}$).

Imputation to 1000 Genomes revealed 45 additional SNPs near *GATA3* that were associated with B-ALL in our data ($P < .05$), although these variants had effect sizes similar to our top genotyped SNP, rs1271899. We found a modest association between the rs3824662 risk allele and later age at diagnosis ($P = .085$). This attenuated effect may be due to our smaller sample size or to a dilution of any age effect by the later average age at diagnosis experienced by Hispanic children with B-ALL.[4]

We have previously shown that B-ALL risk alleles in *ARID5B*, *CDKN2A*, *CEBPE*, and *PIP4K2A* are associated with Amerindian ancestry in Hispanics.[7] Here, we observed that the *GATA3* rs3824662 risk allele was also significantly associated with increased genome-wide Amerindian ancestry, in both Hispanic cases (r, 0.29; $P < 1.0 \times 10^{-4}$) and Hispanic controls (r, 0.23; $P < 1.0 \times 10^{-4}$). By using previously described methods,[7] we quantified the contribution of this SNP to the increased ALL incidence observed in Hispanics relative to populations of exclusively European ancestry. Rs3824662 accounted for a 1.11-fold increased rate of B-ALL in Hispanics versus Europeans (95% CI, 1.02 to 1.21), equivalent to the ethnic incidence rate ratio previously observed for rs7089424 in *ARID5B*.[7]

Our results provide independent confirmation that variation near *GATA3* confers risk of childhood ALL. Furthermore, *GATA3* risk alleles contribute to the increased ALL incidence rate observed in Hispanics relative to Europeans and may underlie their poorer outcomes. Fine-mapping via SNP imputation localized the association peak in Hispanics to an ~15-kb stretch in the shared promoter region of *GATA3* and the *GATA3* antisense RNA (*GATA3-AS1*).[8] Although the *GATA3* risk variants identified by Migliorini et al do not affect protein coding, a significant imputed SNP in our data (rs1244185) is predicted to affect transcription factor binding in *GATA3-AS1* and may influence *GATA3* regulation.[9]

Kyle M. Walsh
Division of Neuroepidemiology, Department of Neurological Surgery and Program in Cancer Genetics, Helen Diller Family Comprehensive Cancer Center, University of California, San Francisco, San Francisco, CA

Adam J. de Smith
Department of Epidemiology and Biostatistics, University of California, San Francisco, San Francisco, CA

Anand P. Chokkalingam
School of Public Health, University of California, Berkeley, Berkeley, CA

Catherine Metayer
School of Public Health, University of California, Berkeley, Berkeley, CA

William Roberts
Department of Pediatric Hematology/Oncology, Rady Children's Hospital, University of California, San Diego, San Diego, CA

Lisa F. Barcellos
School of Public Health, University of California, Berkeley, Berkeley, CA

Joseph L. Wiemels
Department of Epidemiology and Biostatistics, University of California, San Francisco, San Francisco, CA

Patricia A. Buffler
School of Public Health, University of California, Berkeley, Berkeley, CA

K.M.W. and A.J.d.S. are joint first authors.

Acknowledgments: We wish to acknowledge Professor Mel Greaves for his collaborative spirit and helpful discussion about the *GATA3* locus. The authors gratefully acknowledge the clinical investigators at the following collaborating hospitals for help in recruiting patients: University of California, Davis Medical Center (Dr Jonathan Ducore), University of California, San Francisco (Drs Mignon Loh and Katherine Matthay), Children's Hospital of Central California (Dr Vonda Crouse), Lucile Packard Children's Hospital (Dr Gary Dahl), Children's Hospital Oakland (Dr James Feusner), Kaiser Permanente Roseville (formerly Sacramento) (Drs Kent Jolly and Vincent Kiley), Kaiser Permanente Santa Clara (Drs Carolyn Russo, Alan Wong, and Denah Taggart), Kaiser Permanente San Francisco (Dr Kenneth Leung), and Kaiser Permanente Oakland (Drs Daniel Kronish and Stacy Month). We also recognize the assistance of Yang Wang and Ling-i Hsu at the University of California, Berkeley. We mourn the unexpected passing of Pat Buffler, our consummate leader, whose life brought joy to those she knew and whose work brought health to countless more.

Contribution: K.M.W. and A.J.d.S. conceived and designed the study; L.B., P.B., A.P.C., C.M., W.R., K.M.W., and J.L.W. assisted in assembling the data; K.M.W. analyzed and interpreted the data; A.J.d.S. and K.M.W. wrote the manuscript; and all authors critically reviewed and edited the manuscript for intellectual content and gave final approval of the manuscript.

Conflict-of-interest disclosure: The authors declare no competing financial interests.

Correspondence: Kyle M. Walsh, University of California, San Francisco Helen Diller Family Comprehensive Cancer Center, Box 0520, 1450 3rd St, HD276, San Francisco, CA 94143-0520; e-mail: kyle.walsh@ucsf.edu.

References

1. Migliorini G, Fiege B, Hosking FJ, et al. Variation at 10p12.2 and 10p14 influences risk of childhood B-cell acute lymphoblastic leukemia and phenotype. *Blood*. 2013. In press.
2. Yang JJ, Cheng C, Yang W, et al. Genome-wide interrogation of germline genetic variation associated with treatment response in childhood acute lymphoblastic leukemia. *JAMA*. 2009;301(4):393-403.
3. Yamamoto JF, Goodman MT. Patterns of leukemia incidence in the United States by subtype and demographic characteristics, 1997-2002. *Cancer Causes Control*. 2008;19(4):379-390.
4. Goggins WB, Lo FF. Racial and ethnic disparities in survival of US children with acute lymphoblastic leukemia: evidence from the SEER database 1988-2008. *Cancer Causes Control*. 2012;23(5):737-743.
5. Yang JJ, Cheng C, Devidas M, et al. Ancestry and pharmacogenomics of relapse in acute lymphoblastic leukemia. *Nat Genet*. 2011;43(3):237-241.
6. Walsh KM, de Smith AJ, Chokkalingam AP, et al. Novel childhood ALL susceptibility locus BMI1-PIP4K2A is specifically associated with the hyperdiploid subtype. *Blood*. 2013;121(23):4808-4809.

7. Walsh KM, Chokkalingam AP, Hsu LI, et al. Associations between genome-wide Native American ancestry, known risk alleles and B-cell ALL risk in Hispanic children [published online ahead of print April 25, 2013]. *Leukemia*.

8. Zhang H, Nestor CE, Zhao S, et al. Profiling of human CD4(+) T-cell subsets identifies the TH2-specific noncoding RNA GATA3-AS1. *J Allergy Clin Immunol*. 2013;132(4):1005-1008.

9. Boyle AP, Hong EL, Hariharan M, et al. Annotation of functional variation in personal genomes using RegulomeDB. *Genome Res.* 2012;22(9): 1790-1797.

To the editor:

Imatinib therapy in a patient with suspected chronic neutrophilic leukemia and FIP1L1-PDGFRA rearrangement

A 54-year-old man with no significant past medical history presented with skin rash. Complete blood count showed a white blood cell count of 64×10^9/L (63% neutrophils, 2% eosinophils), hemoglobin 14 g/dL, and platelet count 166×10^9/L. Bone marrow evaluation showed a hypercellular marrow with marked granulocytic hyperplasia (Figure 1). There was no increase in marrow eosinophils or fibrosis. No dysplasia was identified. Conventional cytogenetics showed constitutional inv(9). Testing for *BCR-ABL* rearrangement and *JAK2V617F* mutation was negative. The patient was diagnosed with chronic neutrophilic leukemia (CNL) and treatment with hydroxyurea was initiated. Subsequently, array comparative genomic hybridization demonstrated monoallelic interstitial deletion of chromosome 4q12. This was confirmed by interphase fluorescence in situ hybridization (FISH) analysis using a probe set that detects loss of *CHIC2* (surrogate marker of the fusion of the factor interacting with PAP [Fip1]-like 1 [*FIP1L1*] gene to the platelet derived growth factor receptor-α [*PDGFRA*] gene) and by reverse-transcription polymerase chain reaction. The diagnosis was revised to myeloproliferative neoplasm with *FIP1L1-PDGFRA* fusion Imatinib (100 mg daily) was prescribed. Hydroxyurea was discontinued. The white blood cell count normalized within 2 weeks and follow-up testing for *FIP1L1-PDGFRA* fusion by FISH at 3 months was negative. The patient continues to be treated with imatinib 100 mg daily for >1 year with no recurrence of skin rash or leukocytosis.

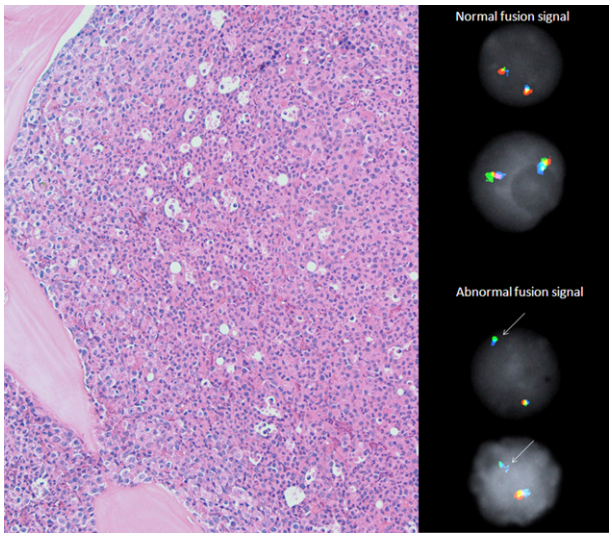

Figure 1. Bone marrow morphology and *CHIC2* FISH. The bone marrow biopsy (left panel) demonstrated hypercellularity and marked predominance of maturing neutrophilic granulocytes. Megakaryocytes were decreased, and no features of myelofibrosis or myelodysplasia were present. Notably, eosinophils and blasts were rare. By fluorescence in situ hybridization (right panel) using a tricolor probe set (Abbott Molecular), normal cells had two intact tricolor (green/orange/aqua) fusion signals whereas abnormal cells (90% in this sample) with monoallelic loss of *CHIC2* (orange pseudocolor) had one green/aqua fusion signal.

Imatinib is approved for treatment of patients with *FIP1L1-PDGFRA* fusion-positive myeloid neoplasms.[1] These patients typically present with peripheral blood eosinophilia. Cools et al[2] described this entity as an interstitial deletion of chromosome 4q12 that leads to the juxtaposition of the *FIP1L1* gene to the *PDGFRA* gene. The resultant fusion product, *FIP1L1-PDGFRA*, results in constitutive activation of the tyrosine kinase PDGFRA and is amenable to therapy with imatinib. Baccarani et al[3] reported achievement of complete hematologic response with imatinib in all patients with *FIP1L1-PDGFRA* fusion and the responses were durable.[4]

Our patient presented with characteristic diagnostic features of patients with CNL (neutrophilic leukocytosis, hypercellular bone marrow with granulocytic hyperplasia, and absence of dysplasia).[5] Although the patient did not present with eosinophilia, *FIP1L1-PDGFRA* fusion was tested because of the presence of skin rash and, remarkably, the test was positive. To the best of our knowledge, this is the first case of a *FIP1L1-PDGFRA* fusion without eosinophilia that responded to tyrosine kinase inhibitor therapy. There is a recent report of *FIP1L1-PDGFRA* fusion without eosinophilia in a patient with monoclonal gammopathy; however, the response to imatinib was not reported.[6] Imatinib response in a patient with CNL has also been reported previously; however, the molecular mechanism for the response was not elucidated.[7] The diagnosis could easily have been missed in our patient because he did not present with eosinophilia. Establishing the correct diagnosis significantly altered the treatment management because *FIP1L1-PDGFRA* fusion is extremely sensitive to imatinib. The 2008 World Health Organization classification lists "myeloid neoplasms associated with *PDGFRA* rearrangement" under "myeloid neoplasms associated with eosinophilia and abnormalities of *PDGFRA,"* indicative of a strong emphasis on the presence of eosinophilia.[8] This is a report of a single patient, but because of the enormous therapeutic implications, we recommend that evaluation for *FIP1L1-PDGFRA* fusion should be considered for all patients with nonclassical myeloproliferative neoplasms. We evaluated 7 additional CNL patients who had stored samples for *FIP1L1-PDGFRA* fusion by FISH; however, all were negative. Recently, activating *CSF3R* mutations were identified in majority of patients with CNL.[9,10] It is possible that CNL patients without *CSF3R* mutations (17% to 41% of cases) likely have alternate molecular pathogenic lesions leading to constitutive activation of other tyrosine kinases such as *PDGFRA*.

Nitin Jain
Department of Leukemia, MD Anderson Cancer Center, Houston, TX

Joseph D. Khoury
Department of Hematopathology, MD Anderson Cancer Center, Houston, TX