Brief report

SNP rs6457327 in the HLA region on chromosome 6p is predictive of the transformation of follicular lymphoma

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Inherited risk determinants for follicular lymphoma (FL) have recently been described in the immune gene-rich human leukocyte antigen region on chromosome 6p. The known importance of host immune response to FL survival led us to evaluate these germline factors in FL outcome. We confirm the association of single nucleotide polymorphisms rs10484561 (P = 3.5 × 10−9) and rs6457327 (P = .008) with risk of FL and demonstrate that rs6457327 predicts both time to (P = .02) and risk of (P < .01) FL transformation independently of clinical variables, including the Follicular Lymphoma International Prognostic Index. (Blood. 2011;117(11):3147-3150)

Introduction

The first genome-wide association studies of follicular lymphoma (FL) recently identified susceptibility loci, rs10484561 (upstream of HLA-DQB1) and rs6457327 (near C6orf15), in the immune gene-rich human leukocyte antigen (HLA) region on chromosome 6p21.2 (supplemental Figures 1-2, available on the Blood Web site; see the Supplemental Materials link at the top of the online article). The role of these single nucleotide polymorphisms (SNPs) and HLA allele types3,4 in FL risk and the importance of the immune response to FL outcome5 led us to investigate the clinical significance of rs10484561 and rs6457327 in a series of FL cases from the United Kingdom.

Methods

Patient samples

DNA from fresh frozen uninvolved bone marrow or peripheral blood samples was obtained from 218 patients diagnosed with FL between 1977 and 2005 and managed at Barts and the London NHS Trust. Sample collection followed informed, written consent in accordance with the Declaration of Helsinki, and the study was performed under approval 05/Q0605/140 from East London and the City Health Authority Local Research Ethics Committee. A case-control analysis evaluated FL risk in the United Kingdom for SNPs rs6457327 and rs10484561 by comparing patient genotypes against those of control populations (2691 and 1570 persons for the respective SNPs) established from the Wellcome Trust Case-Control Consortium 2 (1958) birth cohort (www.wtccc.org.uk; Table 1). Clinical outcome was assessed on a subset of cases (n = 2684 persons for the respective SNPs) established from the Wellcome Trust Case-Control Consortium 2 (1958) birth cohort (www.wtccc.org.uk; Table 1). The publication costs of this article were defrayed in part by page charge payment. Therefore, and solely to indicate this fact, this article is hereby marked “advertisement” in accordance with 18 USC section 1734.

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Submitted October 26, 2010; accepted December 16, 2010. Prepublished online as Blood First Edition paper, January 13, 2011; DOI 10.1182/blood-2010-10-315382.

The online version of this article contains a data supplement.
Table 1. SNPs rs10484561 and rs6457327 association with risk of developing FL in the United Kingdom

| SNP/genotype | Genotype count | MAF (cases/control) | Variant allele OR (95% CI) | Allelic OR (95% CI) | Allelic P | Trend P | Genotypic P |
|--------------|----------------|---------------------|---------------------------|--------------------|------------|---------|-------------|
| rs10484561   | GG             | 0.21/0.11           | 2.40 (1.80-3.19)         | 2.07 (1.61-2.63)   | 3.502 × 10⁻³ | 2.743 × 10⁻³ | 6.342 × 10⁻³ |
|              | GT             | 0.32/0.39           | 0.77 (0.58-1.01)         | 0.75 (0.61-0.93)   | .008       | .008    | .0190       |
|              | TT             | 0.32/0.39           | 0.77 (0.58-1.01)         | 0.75 (0.61-0.93)   | .008       | .008    | .0190       |
| rs6457327    | AA             | 0.32/0.39           | 0.77 (0.58-1.01)         | 0.75 (0.61-0.93)   | .008       | .008    | .0190       |
|              | AC             | 0.32/0.39           | 0.77 (0.58-1.01)         | 0.75 (0.61-0.93)   | .008       | .008    | .0190       |
|              | CC             | 0.32/0.39           | 0.77 (0.58-1.01)         | 0.75 (0.61-0.93)   | .008       | .008    | .0190       |

MAF indicates minor allele frequency; HWE, Hardy-Weinberg equilibrium; and CI, confidence interval.

We validated that variant alleles for rs10484561 (allelic P = 3.5 × 10⁻⁷) and rs6457327 (allelic P = .008) predict increased and reduced risk of FL, respectively, in our United Kingdom cohort (Table 1). Allele frequencies were similar to those reported in other populations. Further, we demonstrate, for the first time, that rs6457327 predicts clinical outcome of FL with the variant allele (AA + AC) predicting a shorter time from diagnosis to transformation (P = .01; Figure 1A) and higher risk of transformation occurrence (P = .006; Figure 1B). These effects were independent of rs10484561, which showed no clinical correlation (supplemental Table 1).

Potential confounders for the associations with transformation are detailed in supplemental Table 2. After their incorporation in multivariate analyses, only rs6457327 genotype retained its predictive value for time to transformation (P = .02, hazard ratio = 2.25; 95% CI, 1.16-4.36). For risk of transformation only rs6457327 AA + AC genotype (P < .01, adjusted OR = 5.48; 95% CI, 1.94-15.51) and progression after first therapy (P = .01, adjusted OR = 8.63; 95% CI, 1.56-47.74) remained predictive (Figure 1C).

rs6457327 is in a 26-kb segment of high linkage disequilibrium that includes only one coding locus, C6orf15. This locus is not, however, a promising candidate as we found that C6orf15 expression was restricted to tonsils (3 of 4) and was not detected in other samples, including B-cell non-Hodgkin lymphoma cell lines and primary tumors (n = 24). Furthermore, DNA sequencing of C6orf15 in 50 diagnostic FL tumor samples revealed no mutational events.

Although more than 30 studies have reported SNPs associated with FL risk, only the recent genome-wide association studies were validated in multiple patient cohorts. Our study is the first to provide independent confirmation of the association between SNPs rs10484561 and rs6457327 and FL risk. Moreover, these findings further support the role of rs10484561 as a major susceptibility locus for FL. There are fewer reports regarding SNP associations with FL outcome, and only one identified an association with transformation. Similar to that study, the association of rs6457327 genotype with transformation did not translate into prediction of survival. It seems unlikely that this is the result of limitations of the study cohort, as the transformation frequency (35%) and its association with overall survival (P < .001, supplemental Figure 3) are similar to those of larger FL series. Nonetheless, it will be important to validate our observations in independent case cohorts, particularly those that accrue prolonged follow-up from the current era of combination immunochemo therapy.

The direct role of C6orf15 in FL remains to be established and, because rs6457327 is also in linkage disequilibrium with HLA-C alleles, further studies are needed to determine the functional locus influencing transformation. However, this study represents a step forward in the characterization of inherited predictors of clinical outcome which, together with recently described acquired
predictors, represent a growing pool of molecular outcome markers in FL.

Acknowledgments

The authors thank Dr George Wright from the National Cancer Institute, National Institute of Health, Bethesda, MD, for providing guidance regarding C6orf15 in the previous genome-wide expression profiling study of FL.

This work was supported by Cancer Research UK (program grant C1574/A6806; D.W., J.F., J.G.G., and T.A.L.), the National Cancer Institute (grants CA122663 and CA104682), the National Institutes of Health (C.F.S.), the Wellcome Trust (grant 075491/Z/04; J.-B.C.), the Partner Fellowship (2009/01) awarded by the European Hematology Association (C.B.), Olivia Walduck’s family (S.M.), and the Mark Ridgwell Family trust (S.M.).

This study makes use of data generated by the Wellcome Trust Case-Control Consortium. A full list of the investigators who contributed to the generation of the data is available from www.wtccc.org.uk. Funding for the project was provided by the Wellcome Trust (award nos. 076113 and 085475).

Authorship

Contribution: D.W. designed the study, performed research, analyzed data, and wrote the paper; P.L., C.F.S., J.-B.C., and L.C. analyzed data; J.M. provided data; S.I., E.C., and C.B. provided samples; M.C. and S.M. collected data; and J.G.G., T.A.L., and J.F. designed the study and wrote the paper.

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

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