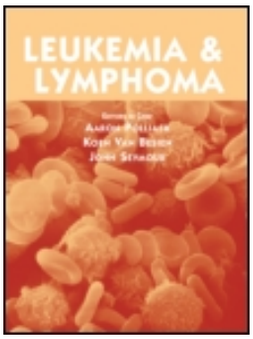

Leukemia & Lymphoma

ISSN: 1042-8194 (Print) 1029-2403 (Online) Journal homepage: informahealthcare.com/journals/ilal20

FISHing for answers in proliferation centers of chronic lymphocytic leukemia lymph nodes

Clive S. Zent, Aaron Polliack & Tamar Tadmor

To cite this article: Clive S. Zent, Aaron Polliack & Tamar Tadmor (2011) FISHing for answers in proliferation centers of chronic lymphocytic leukemia lymph nodes, Leukemia & Lymphoma, 52:6, 946-947, DOI: 10.3109/10428194.2011.565099

To link to this article: https://doi.org/10.3109/10428194.2011.565099

Leukemia & Lymphoma, June 2011; 52(6): 946–947

informa
healthcare

COMMENTARY

FISHing for answers in proliferation centers of chronic lymphocytic leukemia lymph nodes

CLIVE S. ZENT[1], AARON POLLIACK[2], & TAMAR TADMOR[3]

[1]*Division of Hematology, Mayo Clinic, Rochester, MN, USA,* [2]*Department of Hematology, Hadassah University Hospital, Hebrew University Medical School, Jerusalem, Israel, and* [3]*Hematology Unit, Bnai-Zion Medical Center, Haifa, Israel*

Chronic lymphocytic leukemia (CLL) is a dynamic disease involving multiple interacting cellular compartments, with most cell proliferation occurring in the pathognomonic proliferation centers (PCs) of the lymph nodes (LNs) [1,2]. Until recently the importance of the PCs was not fully appreciated [3], but it is now apparent that they play a major role in CLL pathogenesis. CLL cells in the PCs have a higher proliferative rate, as shown by increased expression of CD71, Ki-67, and co-expression of BCL2 together with survivin, compared to CLL cells in the non-PC component of the LN [1]. Increased PC size and cellular proliferation rates within LNs can be used to predict patient outcome by identifying a subgroup with CLL who have more aggressive disease, which has been termed 'accelerated CLL' [1].

Recent gene expression profiling studies have also identified the LN as an important site for up-regulation of gene signatures indicative of B-cell receptor (BCR) signaling and subsequent nuclear factor κB (NFκB) activation [2,4]. In addition, LN derived CLL cells had higher levels of BCR pathway-associated gene expression than CLL cells from the peripheral blood [2]. This difference was more accentuated in patients with unmutated imunoglobulin heavy chain variable gene (IGHV) associated with more aggressive CLL [2]. LN derived CLL cells also had higher levels of expression of the proliferation markers c-MYC, E2F, and Ki-67 [2]. All these data support a central role for the tissue microenvironment of the LN PC in the control of tumor proliferation in CLL [1–4].

Factors that could potentially increase the size and number of PCs include changes in the PC microenvironment and genetic defects in CLL cells that increase CLL cell proliferation or decrease apoptosis. The chromosomal defects associated with aggressive CLL include deletion of 17p13 (17p13−) and 11q22 (11q22−), which result in loss of one allele of TP53 or ATM, respectively. This suggests that defective DNA damage responses in CLL cells could result in a poorer outcome, an association that is further supported by the findings that almost all patients with 17p13− with progressive CLL have a dysfunctional mutation in the remaining TP53, and that patients with bi-allelic loss of function of TP53 in the absence of 17p13− also have a very poor prognosis [5]. In addition, patients with CLL with 11q22− and loss of function of the remaining ATM allele have a very poor prognosis [6]. These TP53 and ATM defects can be acquired in the course of CLL, and this clonal evolution is often associated with disease progression [7]. However, it is not yet known whether clonal evolution is a late event in CLL that is more likely to occur because of increased cell division or if the emergence of a subclone of CLL cells with a new mutation is the result of selection of one of multiple pre-existing clones by selective pressure or growth advantage. The supportive microenvironment of the PC could have a role in either of these possible mechanisms.

The PC microenvironment provides supportive stromal cells, access to antigen via dendritic cells, T cell help, and cytokines [1] which could have a role in either the generation of new subclones or the selection of existing more aggressive clones. The role of the PC in clonal evolution and expansion would ideally be studied in a large prospective study

Correspondence: Dr Clive S. Zent Mayo Clinic, 200 First St. SW, Rochester, MN 55905, USA. Tel: 507-284-2511. Fax: 507-266-4972.
E-mail: zent.clive@mayo.edu

This commentary accompanies an article to be published in *Leukemia & Lymphoma*. Please refer to the table of contents of the print issue in which this commentary appears.

ISSN 1042-8194 print/ISSN 1029-2403 online © 2011 Informa UK, Ltd.
DOI: 10.3109/10428194.2011.565099

enrolling patients with early stage disease soon after diagnosis and before they receive any treatment. These patients would need to undergo multiple serial lymph node biopsies to examine for acquisition of new genetic defects using global genomic analysis. These assays, which include microarray single nucleotide polymorphism (SNP), array comparative genomic hybridization (aCGH), and next generation sequencing, could detect multiple genetic defects and thus accurately asses the incidence and consequences of clonal evolution in PCs and other CLL cell compartments. However, a more feasible initial study could use the more limited but readily available technique of fluorescence *in situ* hybridization (FISH) analysis for known genetic defects in a retrospective analysis of fixed tissue from lymph nodes.

In this issue of *Leukemia and Lymphoma*, Balogh and colleagues report results of a study that uses a small cohort of patients with accelerated disease to determine whether 'PCs play a role in the accumulation of genetic alterations in CLL' [8]. The results reported here are not definitive because of the small sample size and the limited repertoire of the FISH probes used. On the other hand, the data are novel, and could be useful in designing future studies to understand the relationship between the PC and clonal evolution. This study does not provide the answer to the central question of whether the PC increases the risk of clonal evolution or, alternatively, if the increase in the size and numbers of lymph node PCs in patients with more aggressive CLL is a result of clonal evolution. If the microenvironment of the PC increases CLL cell proliferation and survival, thereby increasing the generation and persistence of adverse mutations, the PCs could be expected to contain cells with multiple different genetic defects. However, the failure to demonstrate this convincingly in this study could have been the result of the limited repertoire of the probes used in the FISH analysis or related to the fact that only a limited subset of mutant CLL cells generated in the PC will survive and proliferate. The latter possibility would be more in line with the recently reported SNP analysis data from a large population of patients with progressive CLL requiring treatment, showing only a limited number of recurrent genetic defects [9]. In contrast, mutation in a single CLL cell at any anatomical location conferring a survival advantage could be followed by blood borne distribution to many different LN PCs. This in turn may result in a higher accumulation and concentration of CLL cells with genetic defects in the PCs because of the intrinsic properties of these cells to proliferate rather than the propensity of the PCs to induce clonal evolution. The data presented in this study are thus insufficient to confirm or refute the tested hypothesis.

The PC and clonal evolution have important roles in the etiology and progression of CLL. A better understanding of any role of the PC in the mechanism of clonal evolution could have important implications for improving the management of CLL. Tissue data from this and other studies [1,8] show that it could be important to do a lymph node biopsy in CLL in patients who have a rapidly accelerated disease course. While some of these patients will have transformation to diffuse large B-cell lymphoma, examination of the lymph nodes in those patients without transformation using standard laboratory techniques could also provide valuable prognostic information [1,8]. The clinical utility of these findings will need to be investigated further. In addition, a better understanding of the regulation of cell growth in the PC will be important in determining how the PC can be targeted to improve treatment outcomes in CLL.

Potential conflict of interest: Disclosure forms provided by the authors are available with the full text of this article at www.informahealthcare.com/lal.

References

1. Gine E, Martinez A, Villamor N, et al. Expanded and highly active proliferation centers identify a histological subtype of chronic lymphocytic leukemia ('accelerated' chronic lymphocytic leukemia) with aggressive clinical behavior. Haematologica 2010;95:1526–1533.
2. Herishanu Y, Pérez-Galán P, Liu D, et al. The lymph node microenvironment promotes B-cell receptor signaling, NF-{kappa}B activation, and tumor proliferation in chronic lymphocytic leukemia. Blood 2011;117:563–574.
3. Schmid C, Isaacson PG. Proliferation centres in B-cell malignant lymphoma, lymphocytic (B-CLL): an immunophenotypic study. Histopathology 1994;24:445–451.
4. Soma LA, Craig FE, Swerdlow SH. The proliferation center microenvironment and prognostic markers in chronic lymphocytic leukemia/small lymphocytic lymphoma. Hum Pathol 2006;37:152–159.
5. Mohr J, Helfrich H, Fuge M, et al. DNA damage induced transcriptional programme in CLL: biological and diagnostic implications of functional p53 testing. Blood 2011;117:1622–1632.
6. Austen B, Skowronska A, Baker C, et al. Mutation status of the residual ATM allele is an important determinant of the cellular response to chemotherapy and survival in patients with chronic lymphocytic leukemia containing an 11q deletion. J Clin Oncol 2007;25:5448–5457.
7. Shanafelt TD, Witzig TE, Fink SR, et al. Prospective evaluation of clonal evolution during long-term follow-up of patients with untreated early-stage chronic lymphocytic leukemia. J Clin Oncol 2006;24:4634–4641.
8. Balogh Z, Reiniger L, Rajnai H, et al. High rate of neoplastic cells with genetic abnormalities in proliferation centers of chronic lymphocytic leukemia. Leuk Lymphoma 2011;52:1080–1084.
9. Zenz T, Busch R, Fink A, et al. Genetics of patients with F-refractory CLL or early relapse after FC or FCR: results from the CLL8 trial of the GCLLSG. Blood 2010;116(Suppl. 1): Abstract 2427.