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Review

Genetic variation and risk of chronic lymphocytic leukaemia

Dalemari Crowther-Swanepoel, Richard S. Houlston*

Section of Cancer Genetics, Institute of Cancer Research, Sutton, Surrey SM2 5NG, UK

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ABSTRACT

Chronic lymphocytic leukaemia (CLL) is the most common form of lymphoid malignancy in Western countries, accounting for around a quarter of all leukaemias.

Evidence from epidemiological and family studies have provided evidence for familial clustering of CLL compatible with inherited genetic predisposition to CLL. Direct evidence for genetic susceptibility has been provided by a recent genome wide association study of CLL which has identified common variants at 10 different loci which influence CLL risk. Here we review the current knowledge regarding the allelic architecture of susceptibility to CLL and what the currently identified risk loci are telling us regarding disease aetiology.

Chronic lymphocytic leukaemia (CLL; MIM151400) is an indolent disease resulting from the accumulation of CD5-positive neoplastic B-cells characterised by a low rate of proliferation. The disease accounts for about a quarter of all leukaemias and is the most common form of lymphoid malignancy in Western countries [1]. Family and epidemiological studies have provided evidence for the role of inherited genetic susceptibility to CLL. However, genetic analyses have only recently begun to identify predisposition loci and give insight into the biological basis of disease development.

1. Descriptive epidemiology

Chronic lymphocytic leukaemia is primarily a disease of later life with the median age at diagnosis in the European population being around 72 years [2]. Two key features of the disease have hampered the acquisition of descriptive data on CLL. Firstly, CLL is often encountered as a chance diagnosis, which in turn can reflect on health care provision rather than true differences in disease incidence between countries. Secondly, many epidemiological studies have failed to distinguish B-cell diseases including prolymphocytic leukaemia and possibly lymphocytic lymphomas from CLL. Accepting these caveats it is, however, apparent that CLL is nearly twice as common in men as in women and that incidence rates vary considerably throughout the world.

The incidence of CLL is highest in Europe and populations of European descent elsewhere in the world. Low incidence rates are seen in South and East Asia and sub-Saharan Africa, with the lowest recorded rates being seen in the Japanese population (Table 1). Migrant studies of breast and colon cancer show that individuals acquire the rates of new host countries, implying the involvement of environmental factors in disease aetiology. In contrast, migrant studies of CLL have shown that the incidence of the disease remains low in Asians, even in those born in the United States and in subsequent generations; providing evidence for genetic susceptibility to CLL [3,4].

2. The cellular origin of CLL

Among the several unanswered questions regarding CLL, its cellular origin remains a prominent one. During normal B-cell development, B-cells develop from haematopoietic stem cells in the bone marrow (Fig. 1). Rearrangement of variable (V), diversity (D) and joining (J) immunoglobulin heavy chain (*IGH*) gene segments result in highly effective B-cell receptors (BCR), which mediate B-cell signalling. These BCRs enable interaction with specific antigens [5]. Naïve, immature B-cells exit the marrow and continue their maturation in the spleen where they either become marginal zone B-cells or follicular B-cells [6]. Upon antigenic stimulation, both marginal zone B-cells and follicular B-cells can undergo a rapid burst of proliferation and develop into plasma cells that secrete immunoglobulin. These rapidly-responding plasma cells are generally short-lived and undergo apoptosis *in situ*. Some follicular B-cells can form a germinal centre, and these acquire *IGHV* point mutations. Plasma cells exiting the germinal centre mainly have somatically mutated high-affinity BCRs and have the potential to become long-lived [7,8].

Approximately 50% of CLL cases carry somatic *IGHV* mutations; comparable to the frequency seen in normal B-cell development

* Corresponding author. Tel.: +44 020 8722 4175; fax: +44 020 8722 4059.
E-mail address: richard.houlston@icr.ac.uk (R.S. Houlston).

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Table 1
Familial risks of chronic lymphocytic leukaemia and other B-cell malignancies.

Study	Index case	Familial relative risk (95% CI)	
Cohort studies			
Gunz et al. [32]	Leukaemia	Leukaemia in first-degree degree relatives	2.4 (1.9–3.9)
Giles et al. [33]	LPD	LPD in 1st degree relatives	3.4 (2.4–4.7)
Goldgar et al. [34]	Lymphocytic leukaemia	Lymphocytic leukaemias in 1st degree relatives	5.7 (2.6–10.0)
Case–control studies			
Cartwright et al. [25]	CLL	Lymphocytic leukaemias	4.3 (0.9–19.5)
Linet et al. [30]	CLL	Leukaemia in parents and siblings	2.6 (1.2–5.5)
Pottern et al. [35]	CLL	Leukaemia in parents and siblings	2.3 (1.2–4.4)
Goldin et al. [31]	CLL	CLL in 1st degree relatives	7.5 (3.6–15.6)
Goldin et al. [36]	CLL	CLL in 1st degree relatives	8.5 (6.1–11.7)

[9,10]. The presence of somatic mutations indicates that these CLL cells have undergone antigenic stimulation and are derived from germinal centre cells. Conversely, CLL cells without mutated *IGHV* genes are thus likely to be derived from naïve B-cells. It has, however been shown that all CLL cases, regardless of their *IGHV* mutation status, show the hallmarks of BCR-mediated stimulation [11–13].

Patients with CLL exhibit biased *IGHV* usage and subsets of patients can be identified with so-called stereotyped, or homologous complementarity-determining region 3 (CDR3) sequences [14]. Because the probability of two individual B-cells expressing identical BCRs is extremely low, the observation that around 25% of unrelated CLL cases carry stereotyped BCRs is quite remarkable [15]. This suggests a central role for the recognition of a limited set of structurally similar epitopes in the selection and proliferation of leukaemic clones.

The natural question that follows is if a specific antigen(s) is involved in the pathogenesis of CLL. While the presence of polyreactivity, in at least a proportion of CLL cases underscores the possibility that specific self antigens are involved in the activation of the leukaemic clone, the notion that a specific, foreign antigen may be responsible for the cell stimulation cannot be entirely excluded.

3. Chromosomal abnormalities

The first cytogenetic abnormality in CLL, trisomy 12, was documented in 1980 [16]. Subsequent studies have shown that cytogenetic abnormalities are present in around 50% of CLL cases [17]. The most frequent chromosome anomalies are 13q14 and trisomy 12, which occur in 10–20% of cases. Abnormalities of chromosomes 6q, 11q, 14q and 17p each occur in less than 5% of cases. Overall, around 50% of patients have gross chromosomal abnormalities, 25% have two abnormalities and the remainder a more complex karyotype.

The genetic consequence of trisomy 12 remains unknown. Investigation of rare cases with translocations, duplications or amplifications of chromosome 12 has suggested a possible role for the *MDM2* gene [18]. However, to date no relationship between *MDM2* and CLL development has been established. The majority of cases with 13q14 abnormalities have a deletion in this region,

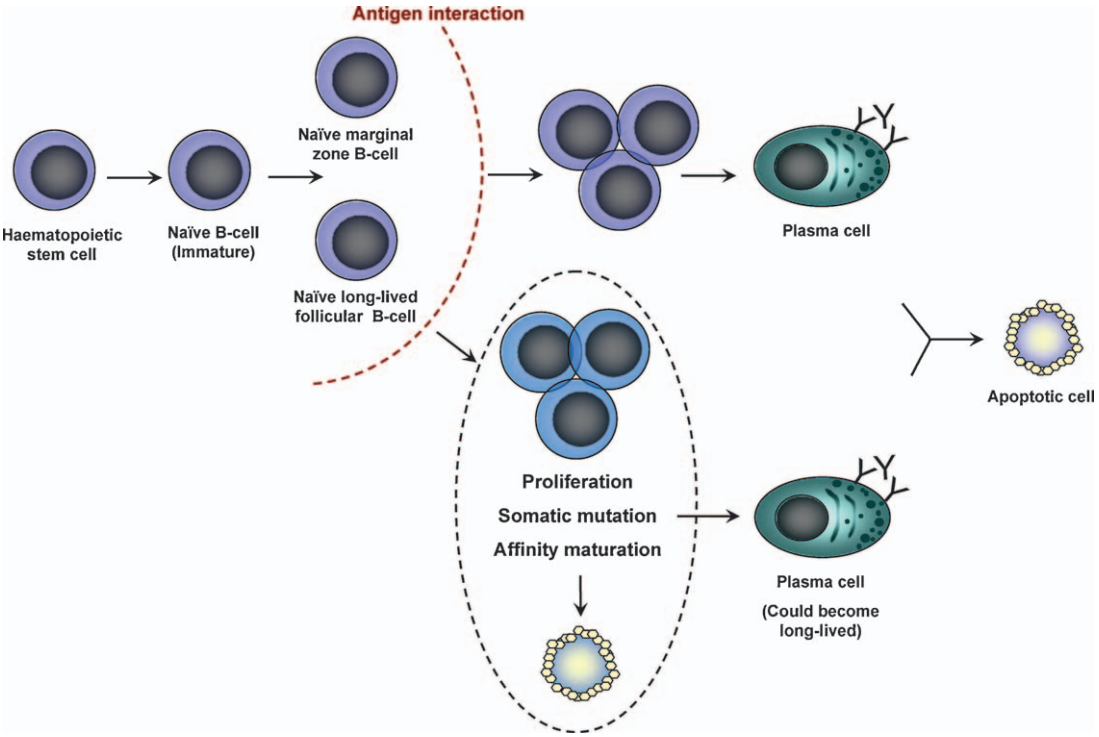

Fig. 1. Normal plasma cell development. Upon antigenic stimulation naïve marginal-zone B-cells and follicular B-cells differentiate into plasma cells. Some activated follicular B-cells form a germinal centre. Plasma cells formed through the germinal centre could become long-lived.

while the remainder have a translocation involving different partner chromosomes. In contrast with other deletions in CLL, loss of homozygosity is frequently seen at 13q14 [19], indicating that one or more genes of importance in CLL pathogenesis map within the region.

Analysis of this region led to the discovery of two physically linked microRNAs, *miR15a* and *miR16-1*. The *miR15a*/*miR16-1* cluster appears to be deleted or down regulated in the majority of CLL [20]. Expression of *miR15a*/*miR16-1* has been shown to be inversely correlated to expression of the anti-apoptotic *BCL2* gene, which is over-expressed in CLL B-cells [21]. Both microRNAs negatively regulate *BCL2* at a post-transcriptional level [22] and thus effectively function as tumour suppressors in CLL.

4. Immune dysfunction

Intuitively, links between genetics and immune dysfunction as a possible basis for CLL development are highly attractive. The clinical course of CLL is dominated by events associated with immune dysfunction, manifesting as susceptibility to infection and autoimmunity. Autoimmune complications occur in 10–25% of patients [23], the most common of which are haemolytic anaemia and immune thrombocytopaenia. The pathogenesis of autoimmunity in CLL is unknown, but may be related to the ability of CLL cells to both process and present antigen.

There is currently no compelling evidence linking infection by human T-cell lymphotropic virus, human immunodeficiency virus or immunosuppression following organ transplantation with CLL pathogenesis [24]. A variety of prior medical conditions have been reported to confer an increased risk of CLL. Scarlet fever, bronchitis and rheumatoid arthritis are some examples [25]. Two studies from Scandinavia [26,27] suggest that the occurrence of one or more episodes of pneumonia within five years of the diagnosis of CLL might serve as a potential trigger for CLL development, although the pneumonia could be a consequence of immune deficiency as an early manifestation of CLL, prior to diagnosis. However, no consistent association has yet emerged and these associations must be considered as unreliable.

5. Genetic susceptibility to CLL

Over the last seven decades more than 100 families have been reported in the literature in which clustering of CLL has been documented [28]. While not exclusively a consequence of genetic predisposition, familial aggregation provides strong evidence for inherited genetic factors playing a role in disease development. In a number of the families reported, CLL co-segregates with other B-cell lymphoproliferative disorders (LPD) such as Hodgkin lymphoma, suggesting that part of the familial predisposition could be mediated through pleiotropic mechanisms [29–31].

Eight epidemiological case–control and cohort studies have systematically enumerated the risk of relatives of CLL patients developing CLL or other LPDs [25,30–36] (Table 1). All have reported elevated risks of CLL in relatives of cases. The largest and most comprehensive of these was based on an analysis of 9717 CLL cases and 38,159 controls ascertained through the Swedish Cancer Registry. Findings underscored CLL being characterised by a high familial relative risk (FRR); the FRR of CLL in first-degree relatives of cases was increased 8.5-fold. Furthermore, the risk of other non-Hodgkin lymphoma (NHL) was increased 1.9-fold. Evaluating NHL subtypes revealed a striking excess of indolent B-cell NHL, specifically lymphoplasmacytic lymphoma/Waldenström macroglobulinemia and hairy cell leukaemia [36]. These findings substantiate a relationship between the risk of CLL and other LPDs which has previously been noted in case reports of single families and that may reflect the pleiotropic effects of an inherited predisposition.

6. Characteristics of familial CLL

The phenotype of earlier age of onset and increased risk of second tumours is a classical feature of many familial cancers. A survey of 28 CLL families suggests that familial cases presents ~10 years earlier than sporadic cases, implying a more aggressive clonal expansion [37], however more recent studies provide little support for such an assertion [38]. Anticipation, the phenomenon of intensified clinical severity and earlier age of onset with each successive generation, has been reported for CLL, with mean declines between parents and offspring being as many as 22 years [39–41]. However, findings were based on data from families ascertained for genetic studies, which are enriched for younger cases. Bias could therefore be introduced through censoring or cohort effects. In a study using Swedish registry data where corrections were made for possible sources of bias there was little evidence to support anticipation in CLL [42].

It is possible that CLL development may be influenced by antigenic recognition or selection through the BCR. Thus familial CLL would be associated with a more restricted phenotype with respect to ontogenic development than sporadic disease. Two studies have compared CLL phenotypes between familial and sporadic CLL [38,43]. Stage at diagnosis, need for treatment and overall survival were reported to be comparable [43]. However, the sex ratio of familial CLL was found to be more equal than that of sporadic CLL [38,43]. Females affected by CLL might therefore have more predisposition alleles. The relatives of affected females probably share the same predisposition genes, which increase their genetic liability, accounting for the higher proportion of familial cases among females compared to males. The frequency of mutated CLL was higher among familial CLL cases and there was evidence of intrafamilial concordance in mutation status. The repertoire and frequency of *IGHV* usage was, however, not significantly different between familial and sporadic CLL and *IGHV* usage was not correlated between affected members of the same family [38]. These observations provide evidence that familial CLL is essentially indistinguishable from sporadic CLL and favours a multi-factorial basis to disease development in general.

Notwithstanding these data the repertoire of *IGHV* genes expressed by B-cells in CLL patients is, however, biased when compared to that of normal B-cells [44]. Asymmetric usages of the immunoglobulin genes have been well characterised in CLL, with notable overrepresentation of various genes, including *IGHV1-69* and *IGHV4-34* [45]. Such preferential usage of certain *IGHV* genes could indicate selective drive on a B-cell population via a superantigen and lends support to the hypothesis that selection by a common antigen could contribute to disease pathogenesis. Preferential stimulation of B-cells expressing the *IGHV4-34* gene occurs in a number of infections, including those caused by Epstein-Barr virus (EBV) and cytomegalovirus (CMV) [46]. The first evidence linking latent or persistent infection by EBV and CMV with CLL cases expressing *IGHV4-34* was recently published [47], signifying the possible involvement of these pathogens in the aetiology of CLL.

7. Models of inherited predisposition in CLL

The magnitude of the familial risk of CLL is compatible with a wide range of genetic models of inheritance. Although rare, large families with multiple affected individuals support a role for dominantly acting mutations that confer a high risk of CLL. To date five linkage scans of CLL families have been performed [48–52]. The largest of these, certainly in terms of number of families geno-

Table 2
The ten CLL risk loci identified to date.

SNP	Chromosome	Risk allele	Nearest gene(s)	Odds ratio
rs17483466	2q13	G	*ACOXL*, *BCL2L11*	1.39
rs13397985	2q37.1	G	*SP140*, *SP110*	1.41
rs757978	2q37.3	A	*FIR*	1.39
rs872071	6p25.3	G	*IRF4*	1.54
rs2456449	8q24.21	G	–	1.26
rs735665	11q24.1	A	*GRAMD1B*	1.45
rs7169431	15q21.3	A	*RFX7*, *NEDD4*	1.36
rs7176508	15q23	A	–	1.37
rs305061	16q24.1	T	*IRF8*	1.22
rs11083846	19q13.32	A	*PRKD2*, *STRN4*	1.35

typed, was the scan conducted by Sellick et al. [49], based on an analysis of 206 CLL pedigrees. The best evidence for a single disease locus in this study was attained at 2q21 under a recessive model.

Genetic heterogeneity inevitably erodes study power and failure to unambiguously identify a disease locus in this and two other scans may have been a consequence of limited power. To circumvent the issue of heterogeneity two research groups have performed linkage searches on single, large CLL families. The first was based on analysis of a family comprising 11 affected members [51]. The second study involved the analysis of a multigenerational family in which 7 members had been diagnosed with CLL [52]. Neither study provided statistically significant evidence for a single major locus conferring susceptibility to CLL.

Failure to identify a disease-causing gene through linkage has led to a reappraisal of the assumption of Mendelian predisposition to CLL and a polygenic model of inheritance seems more likely. Under this model, multiple low risk variants, with effect sizes of ~1.1–1.5, could make an important contribution to the overall familial risk. Although such alleles have small effects individually, they could contribute significantly to disease susceptibility in the general population. Furthermore, by acting in concert they could generate a high risk of CLL in a subset of the population [53].

The search for low risk alleles for CLL has centred on association studies of candidate genes, where the frequencies of polymorphic variants, usually single nucleotide polymorphisms (SNPs), are compared in cases and controls. Most studies have evaluated only a restricted number of polymorphisms, such as those influencing methylation or carcinogen metabolism and no definitive susceptibility alleles have been identified from candidate gene analyses. The inherent statistical uncertainty of case–control studies involving just a few hundred cases and controls seriously limits the power of such studies to reliably identify genetic determinants conferring modest but potentially important risks. Furthermore, without a clear understanding of the biology of predisposition the definition of suitable genes for the disease is inherently problematic, making an unbiased approach to loci selection highly desirable.

With the completion of the Human Genome Project, more than 10 million SNPs have been documented in addition to smaller numbers of insertion/deletion and copy number variations. The high resolution LD maps and comprehensive sets of tagging SNPs available through the HapMap, coupled with the development of highly efficient analytical platforms allow genome wide association (GWA) studies to be conducted in a more economical way. This approach is unbiased and does not depend on prior knowledge of the function or involvement of any gene in disease causation. Furthermore, important variants in previously unstudied genes or even in non-coding regions could be identified in this way.

The recent genome wide association (GWA) study of CLL [54,55] has vindicated the assertion of common susceptibility to CLL, identifying ten novel CLL risk loci at 2q13, 2q37.1, 2q37.3, 6p25.3, 8q24.21, 11q24.1, 15q21.3, 15q23, 16q24.1 and 19q13.32 (Table 2). Six of these variations have been replicated in further independent case–control series, thereby providing compelling evidence for the associations [56,57]. None of the genes implicated by the GWA scan have previously been evaluated in targeted association studies, emphasising that the candidate gene approach was severely limited by inadequate knowledge of tumour biology.

While the risks of CLL associated with these 10 SNPs are modest, with relative risks of 1.2–1.7 per allele, their contribution to CLL incidence is high as a large proportion of the population is carriers of risk alleles. Moreover, the risk increases with increasing numbers of variant alleles for the ten loci and for the 2% of the population who carry 13 or more risk alleles the risk of CLL is increased ~8-fold [55].

8. Monoclonal B-cell lymphocytosis

The recognition that common variants influence the risk of CLL raises the possibility that, while clinical CLL may be relatively uncommon in the population, progenitor lesions may be far more common. Intriguingly this assertion is supported by the observation that CLL-phenotype B-cells (CD5+, CD23+, $CD20_{low}$, $sIgM_{low}$) of monoclonal B-cell lymphocytosis (MBL) are detectable in ~3% of adults in the general population. These cells are essentially indistinguishable from CLL B-cells in terms of chromosomal abnormalities and *IGHV* mutation status [58].

Studies have shown that MBL develops into CLL at a rate of 1.1% per year [59]. These data coupled with the observation that approximately 15% of relatives of familial CLL patients have MBL [60,61] supports the notion that MBL is a surrogate marker of genetic predisposition.

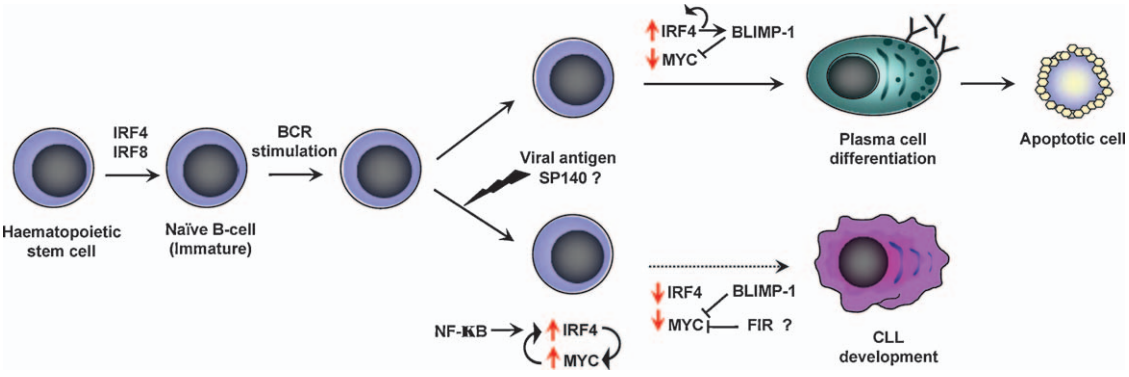

Fig. 2. Model of normal B-cell versus CLL development. Established roles for IRF4, IRF8 and MYC are indicated, along with a possible role for FIR and SP140. BCR, B-cell receptor [8,62,86].

9. Integrating genetics and biology

Findings from the GWA study of CLL provide evidence that variation in *SP140*, *IRF4*, and *FIR* (Fig. 2) influence the risk of developing the disease [54,55]. *IRF4* is a strong candidate gene for CLL susceptibility *a priori*, being a key regulator of lymphocyte development and proliferation. Moreover, *IRF4* expression is involved in the development of CLL and multiple myeloma. Through interaction with transcription factors including PU.1, IRF4 controls the termination of pre-BCR signalling and promotes the differentiation of pro-B cells to small B-cells. Furthermore, IRF4 controls the transition of memory B-cells, thought to be the precursor cell type for CLL, to plasma cells [8,62,63]. The observation that genotype is associated with *IRF4* expression in a dose-dependent fashion in EBV-transformed lymphocytes is consistent with a model in which the causal variant influences risk by arresting transition of memory B-cells through decreased *IRF4* expression [54]. Interestingly, it has also been found that EBV transformation of human B-cells *in vitro* requires the presence of high levels of *IRF4* [64].

Sunlight has immunosuppressive properties to lymphocytes and a link between malignant melanoma and CLL has been reported, raising the possibility of a common biological basis to the two diseases [65,66]. Such an assertion is supported by the observation that genetic variation in *IRF4* has been associated with skin pigmentation [67] as well as risk of CLL [54] and melanoma [68].

SP140 is the lymphoid-restricted homolog of *SP100* expressed in all mature B cells and plasma cell lines, as well as some T-cells [69,70]. SP100 is a major mediator of EBV-encoded nuclear antigen leader protein co-activation, which is important for establishment of latent viral infections and B-cell immortalisation [71]. As *SP140* expression has been implicated in host response to immunodeficiency virus type 1 [72] it is possible that *SP140* influences CLL risk by affecting response to viral challenge.

The association signal at 2q37.3 provides evidence for a role of the *FARP2* gene in the aetiology of CLL. *FARP2*, also known as FBP-interacting repressor (*FIR*) was originally isolated as a poly(U) binding splicing factor, that together with the splicing factors p54 and U2AF, promotes RNA splicing [73]. FIR is also an important regulator of *MYC* gene activity which, by interacting with far upstream element (FUSE) and FUSE binding protein (FBP), represses *MYC* transcription [74,75].

The 19q13.32 association implicates variation in *PRKD2* in CLL. Low levels of *PRKD2* expression and autophosphorylation have been reported to be a feature of a number of B-cell tumours including mantle cell and Burkitt's lymphoma, and ~50% of CLL/small lymphocytic lymphomas [76]. Variation in *IRF8* is a strong candidate for the association with CLL risk as IRF8 regulates α (alpha)- and β (beta)-interferon response. Moreover, *IRF8* is involved in B-cell lineage specification, immunoglobulin rearrangement and regulation of the germinal centre reaction [77]. The association signal at 15q21.3 is flanked by *NEDD4* and *RFX7*. Although there is no evidence for a direct role of *NEDD4* in CLL, it represents a credible candidate gene because of its role in regulating viral latency and pathogenesis of EBV. Specifically, NEDD4 regulates EBV-LMP2A, which mimics signalling induced by the BCR, thereby altering B-cell development [78].

The probable basis for the 2q13, 11q24.1 and15q23 associations may be less straightforward than a regulatory effect on candidate gene expression, perhaps favouring a position effect through long-range linkage disequilibrium with a variant mapping outside the gene locus.

The 8q24.21 association is intriguing. GWA studies of other cancers have shown that the 128–130 Mb genomic interval at 8q24.21 harbours multiple independent loci with different tumour specificities namely prostate [79], breast [80], colorectal-prostate [81,82], prostate [83] and bladder cancer [84]. The genomic regions defining these loci are however distinct from the 8q24.21 CLL association signal. The 8q24.21 region to which the cancer associations map is bereft of genes and predicted transcripts. The colorectal-prostate cancer association has been shown to affect TCF4 binding to an enhancer for *MYC*, providing a mechanistic basis for this 8q24.21 association [85]. It is possible that the effect of the other 8q24.21 cancer risk loci is via *MYC* through similar long-range *cis*-acting mechanisms. If the 8q24.21 locus influences risk through differential *MYC* expression, the association is highly relevant because MYC is a direct target of IRF4 in activated B-cells (Fig. 2) [86]. This, together with the fact that FIR plays an important regulatory role in *MYC* expression, might indicate a central role for MYC in CLL development.

Collectively, these data show that common low-penetrance susceptibility alleles contribute to the risk of developing CLL and implicate genes involved in transcriptional regulation and differentiation of B-cell progenitors as the biological basis of predisposition. Moreover recent data shows a strong association between genetic variation at the 10 loci and MBL risk, providing support for these loci impacting on early development of CLL rather than disease progression (unpublished data).

10. Conclusions

Our knowledge of predisposition to CLL is rapidly developing. It is now well established that CLL has a strong familial basis. Moreover, the observation that MBL represents a progenitor lesion for CLL offers considerable opportunities for understanding the key events in the early development of CLL.

The advent of analytical platforms, which allow comprehensive interrogation of the genome, is enabling researchers to identify variants that influence an individual's susceptibility to develop CLL. Identifying the sequence changes responsible for causal associations identified should thus provide further insight into the biology of CLL and this may lead to the development of aetiological hypotheses regarding non-genetic risk factors. Presently, there is increasing data from genetic associations to implicate a viral basis to CLL development. Finally, a greater understanding of the biological basis of the disease should lead to the development of novel therapeutic interventions.

Conflict of interest statement

The authors declare that there are no conflicts of interest.

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References

[1] Ries LA, Wingo PA, Miller DS, Howe HL, Weir HK, Rosenberg HM, et al. The annual report to the nation on the status of cancer, 1973–1997, with a special section on colorectal cancer. Cancer 2000;88:2398–424.
[2] Horner MJ, Ries LAG, Krapcho M, Neyman N, Aminou R, Howlader N, et al. In: Edwards BK, editor. SEER cancer statistics review, 1975–2006. Bethesda; 2009.
[3] Gale RP, Cozen W, Goodman MT, Wang FF, Bernstein L. Decreased chronic lymphocytic leukemia incidence in Asians in Los Angeles County. Leuk Res 2000;24:665–9.
[4] Haenszel W, Kurihara M. Studies of Japanese migrants. I. Mortality from cancer and other diseases among Japanese in the United States. J Natl Cancer Inst 1968;40:43–68.
[5] Cobb RM, Oestreich KJ, Osipovich OA, Oltz EM. Accessibility control of V(D)J recombination. Adv Immunol 2006;91:45–109.
[6] Chung JB, Silverman M, Monroe JG. Transitional B cells: step by step towards immune competence. Trends Immunol 2003;24:343–9.

[7] McHeyzer-Williams LJ, Driver DJ, McHeyzer-Williams MG. Germinal center reaction. Curr Opin Hematol 2001;8:52–9.
[8] Shapiro-Shelef M, Calame K. Regulation of plasma-cell development. Nat Rev Immunol 2005;5:230–42.
[9] Fais F, Ghiotto F, Hashimoto S, Sellars B, Valetto A, Allen SL, et al. Chronic lymphocytic leukemia B cells express restricted sets of mutated and unmutated antigen receptors. J Clin Invest 1998;102:1515–25.
[10] Schroeder Jr HW, Dighiero G. The pathogenesis of chronic lymphocytic leukemia: analysis of the antibody repertoire. Immunol Today 1994;15:288–94.
[11] Damle RN, Ghiotto F, Valetto A, Albesiano E, Fais F, Yan XJ, et al. B-cell chronic lymphocytic leukemia cells express a surface membrane phenotype of activated, antigen-experienced B lymphocytes. Blood 2002;99:4087–93.
[12] Stevenson FK, Caligaris-Cappio F. Chronic lymphocytic leukemia: revelations from the B-cell receptor. Blood 2004;103:4389–95.
[13] Klein U, Tu Y, Stolovitzky GA, Mattioli M, Cattoretti G, Husson H, et al. Gene expression profiling of B cell chronic lymphocytic leukemia reveals a homogeneous phenotype related to memory B cells. J Exp Med 2001;194:1625–38.
[14] Murray F, Darzentas N, Hadzidimitriou A, Tobin G, Boudjogra M, Scielzo C, et al. Stereotyped patterns of somatic hypermutation in subsets of patients with chronic lymphocytic leukemia: implications for the role of antigen selection in leukemogenesis. Blood 2008;111:1524–33.
[15] Stamatopoulos K, Belessi C, Moreno C, Boudjograh M, Guida G, Smilevska T, et al. Over 20% of patients with chronic lymphocytic leukemia carry stereotyped receptors: pathogenetic implications and clinical correlations. Blood 2007;109:259–70.
[16] Gahrton G, Robert KH, Friberg K, Zech L, Bird AG. Nonrandom chromosomal aberrations in chronic lymphocytic leukemia revealed by polyclonal B-cell-mitogen stimulation. Blood 1980;56:640–7.
[17] Oscier DG. Cytogenetic and molecular abnormalities in chronic lymphocytic leukaemia. Blood Rev 1994;8:88–97.
[18] Dierlamm J, Michaux L, Criel A, Wlodarska I, Van den Berghe H, Hossfeld DK. Genetic abnormalities in chronic lymphocytic leukemia and their clinical and prognostic implications. Cancer Genet Cytogenet 1997;94:27–35.
[19] Gardiner AC, Corcoran MM, Oscier DG. Cytogenetic, fluorescence in situ hybridisation, and clinical evaluation of translocations with concomitant deletion at 13q14 in chronic lymphocytic leukaemia. Genes Chromosomes Cancer 1997;20:73–81.
[20] Calin GA, Dumitru CD, Shimizu M, Bichi R, Zupo S, Noch E, et al. Frequent deletions and down-regulation of micro-RNA genes miR15 and miR16 at 13q14 in chronic lymphocytic leukemia. Proc Natl Acad Sci USA 2002;99:15524–9.
[21] Kitada S, Andersen J, Akar S, Zapata JM, Takayama S, Krajewski S, et al. Expression of apoptosis-regulating proteins in chronic lymphocytic leukemia: correlations with In vitro and In vivo chemoresponses. Blood 1998;91:3379–89.
[22] Cimmino A, Calin GA, Fabbri M, Iorio MV, Ferracin M, Shimizu M, et al. miR-15 and miR-16 induce apoptosis by targeting BCL2. Proc Natl Acad Sci USA 2005;102:13944–9.
[23] Hamblin TJ. Autoimmune complications of chronic lymphocytic leukemia. Semin Oncol 2006;33:230–9.
[24] Analo HI, Akanmu AS, Akinsete I, Njoku OS, Okany CC. Seroprevalence study of HTLV-1 and HIV infection in blood donors and patients with lymphoid malignancies in Lagos, Nigeria. Cent Afr J Med 1998;44:130–4.
[25] Cartwright RA, Bernard SM, Bird CC, Darwin CM, O'Brien C, Richards ID, et al. Chronic lymphocytic leukaemia: case control epidemiological study in Yorkshire. Br J Cancer 1987;56:79–82.
[26] Landgren O, Pfeiffer RM, Stewart L, Gridley G, Mellemkjaer L, Hemminki K, et al. Risk of second malignant neoplasms among lymphoma patients with a family history of cancer. Int J Cancer 2007;120:1099–102.
[27] Landgren O, Rapkin JS, Caporaso NE, Mellemkjaer L, Gridley G, Goldin LR, et al. Respiratory tract infections and subsequent risk of chronic lymphocytic leukemia. Blood 2007;109:2198–201.
[28] Yuille MR, Matutes E, Marossy A, Hilditch B, Catovsky D, Houlston RS. Familial chronic lymphocytic leukaemia: a survey and review of published studies. Br J Haematol 2000;109:794–9.
[29] Jonsson V, Houlston RS, Catovsky D, Yuille MR, Hilden J, Olsen JH, et al. CLL family 'Pedigree 14' revisited: 1947–2004. Leukemia 2005;19:1025–8.
[30] Linet MS, Van Natta ML, Brookmeyer R, Khoury MJ, McCaffrey LD, Humphrey RL, et al. Familial cancer history and chronic lymphocytic leukemia. A case–control study. Am J Epidemiol 1989;130:655–64.
[31] Goldin LR, Pfeiffer RM, Li X, Hemminki K. Familial risk of lymphoproliferative tumors in families of patients with chronic lymphocytic leukemia: results from the Swedish Family-cancer database. Blood 2004;104:1850–4.
[32] Gunz FW, Gunz JP, Veale AM, Chapman CJ, Houston IB. Familial leukaemia: a study of 909 families. Scand J Haematol 1975;15:117–31.
[33] Giles GG, Lickiss JN, Baikie MJ, Lowenthal RM, Panton J. Myeloproliferative and lymphoproliferative disorders in Tasmania, 1972–80: occupational and familial aspects. J Natl Cancer Inst 1984;72:1233–40.
[34] Goldgar DE, Easton DF, Cannon-Albright LA, Skolnick MH. Systematic population-based assessment of cancer risk in first-degree relatives of cancer probands. J Natl Cancer Inst 1994;86:1600–8.
[35] Pottern LM, Linet M, Blair A, Dick F, Burmeister LF, Gibson R, et al. Familial cancers associated with subtypes of leukemia and non-Hodgkin's lymphoma. Leuk Res 1991;15:305–14.
[36] Goldin LR, Bjorkholm M, Kristinsson SY, Turesson I, Landgren O. Elevated risk of chronic lymphocytic leukemia and other indolent non-Hodgkin's lymphomas among relatives of patients with chronic lymphocytic leukemia. Haematologica 2009;94:647–53.
[37] Ishibe N, Sgambati MT, Fontaine L, Goldin LR, Jain N, Weissman N, et al. Clinical characteristics of familial B-CLL in the National Cancer Institute Familial Registry. Leuk Lymphoma 2001;42:99–108.
[38] Crowther-Swanepoel D, Wild R, Sellick G, Dyer MJ, Mauro FR, Cuthbert RJ, et al. Insight into the pathogenesis of chronic lymphocytic leukemia (CLL) through analysis of IgVH gene usage and mutation status in familial CLL. Blood 2008;111:5691–3.
[39] Wiernik PH, Ashwin M, Hu XP, Paietta E, Brown K. Anticipation in familial chronic lymphocytic leukaemia. Br J Haematol 2001;113:407–14.
[40] Horwitz M, Goode EL, Jarvik GP. Anticipation in familial leukemia. Am J Hum Genet 1996;59:990–8.
[41] Yuille MR, Houlston RS, Catovsky D. Anticipation in familial chronic lymphocytic leukaemia. Leukemia 1998;12:1696–8.
[42] Daugherty SE, Pfeiffer RM, Mellemkjaer L, Hemminki K, Goldin LR. No evidence for anticipation in lymphoproliferative tumors in population-based samples. Cancer Epidemiol Biomarkers Prev 2005;14:1245–50.
[43] Mauro FR, Giammartini E, Gentile M, Sperduti I, Valle V, Pizzuti A, et al. Clinical features and outcome of familial chronic lymphocytic leukemia. Haematologica 2006;91:1117–20.
[44] Brezinschek HP, Brezinschek RI, Lipsky PE. Analysis of the heavy chain repertoire of human peripheral B cells using single-cell polymerase chain reaction. J Immunol 1995;155:190–202.
[45] Hamblin TJ, Davis Z, Gardiner A, Oscier DG, Stevenson FK. Unmutated Ig V(H) genes are associated with a more aggressive form of chronic lymphocytic leukemia. Blood 1999;94:1848–54.
[46] Mockridge CI, Rahman A, Buchan S, Hamblin T, Isenberg DA, Stevenson FK, et al. Common patterns of B cell perturbation and expanded V4-34 immunoglobulin gene usage in autoimmunity and infection. Autoimmunity 2004;37:9–15.
[47] Kostareli E, Hadzidimitriou A, Stavroyianni N, Darzentas N, Athanasiadou A, Gounari M, et al. Molecular evidence for EBV and CMV persistence in a subset of patients with chronic lymphocytic leukemia expressing stereotyped IGHV4-34 B-cell receptors. Leukemia 2009;23:919–24.
[48] Goldin LR, Ishibe N, Sgambati M, Marti GE, Fontaine L, Lee MP, et al. A genome scan of 18 families with chronic lymphocytic leukaemia. Br J Haematol 2003;121:866–73.
[49] Sellick GS, Goldin LR, Wild RW, Slager SL, Ressenti L, Strom SS, et al. A high-density SNP genome-wide linkage search of 206 families identifies susceptibility loci for chronic lymphocytic leukemia. Blood 2007;110:3326–33.
[50] Sellick GS, Webb EL, Allinson R, Matutes E, Dyer MJ, Jonsson V, et al. A high-density SNP genomewide linkage scan for chronic lymphocytic leukemia-susceptibility loci. Am J Hum Genet 2005;77:420–9.
[51] Fuller SJ, Papaemmanuil E, McKinnon L, Webb E, Sellick GS, Dao-Ung LP, et al. Analysis of a large multi-generational family provides insight into the genetics of chronic lymphocytic leukemia. Br J Haematol 2008;142:238–45.
[52] Raval A, Tanner SM, Byrd JC, Angerman EB, Perko JD, Chen SS, et al. Downregulation of death-associated protein kinase 1 (DAPK1) in chronic lymphocytic leukemia. Cell 2007;129:879–90.
[53] Houlston RS, Peto J. The future of association studies of common cancers. Hum Genet 2003;112:434–5.
[54] Di Bernardo MC, Crowther-Swanepoel D, Broderick P, Webb E, Sellick G, Wild R, et al. A genome-wide association study identifies six susceptibility loci for chronic lymphocytic leukemia. Nat Genet 2008;40:1204–10.
[55] Crowther-Swanepoel D, Broderick P, Di Bernardo MC, Dobbins SE, Torres M, Mansouri M, et al. Common variants at 2q37.3, 8q24.21, 15q21.3 and 16q24.1 influence chronic lymphocytic leukemia risk. Nat Genet 2010;42:132–6.
[56] Slager SL, Goldin LR, Strom SS, Lanasa MC, Spector LG, Rassenti L, et al. Genetic susceptibility variants for chronic lymphocytic leukemia. Cancer Epidemiol Biomarkers Prev 2010;19:1098–102.
[57] Crowther-Swanepoel D, Mansouri M, Enjuanes A, Vega A, Smedby KE, Ruiz-Ponte C, et al. Verification that common variation at 2q37.1, 6p25.3, 11q24.1, 15q23, and 19q13.32 influences chronic lymphocytic leukaemia risk. Br J Haematol 2010;150:473–9.
[58] Rawstron AC, Green MJ, Kuzmicki A, Kennedy B, Fenton JA, Evans PA, et al. Monoclonal B lymphocytes with the characteristics of "indolent" chronic lymphocytic leukemia are present in 3.5% of adults with normal blood counts. Blood 2002;100:635–9.
[59] Rawstron AC, Bennett FL, O'Connor SJ, Kwok M, Fenton JA, Plummer M, et al. Monoclonal B-cell lymphocytosis and chronic lymphocytic leukemia. N Engl J Med 2008;359:575–83.
[60] Marti GE, Carter P, Abbasi F, Washington GC, Jain N, Zenger VE, et al. B-cell monoclonal lymphocytosis and B-cell abnormalities in the setting of familial B-cell chronic lymphocytic leukemia. Cytometry B Clin Cytom 2003;52:1–12.
[61] Rawstron AC, Yuille MR, Fuller J, Cullen M, Kennedy B, Richards SJ, et al. Inherited predisposition to CLL is detectable as subclinical monoclonal B-lymphocyte expansion. Blood 2002;100:2289–90.
[62] Busslinger M. Transcriptional control of early B cell development. Annu Rev Immunol 2004;22:55–79.
[63] Klein U, Casola S, Cattoretti G, Shen Q, Lia M, Mo T, et al. Transcription factor IRF4 controls plasma cell differentiation and class-switch recombination. Nat Immunol 2006;7:773–82.
[64] Xu D, Zhao L, Del Valle L, Miklossy J, Zhang L. Interferon regulatory factor 4 is involved in Epstein-Barr virus-mediated transformation of human B lymphocytes. J Virol 2008;82:6251–8.

[65] Adami J, Frisch M, Yuen J, Glimelius B, Melbye M. Evidence of an association between non-Hodgkin's lymphoma and skin cancer. BMJ 1995;310:1491–5.
[66] Swerdlow AJ, Storm HH, Sasieni PD. Risks of second primary malignancy in patients with cutaneous and ocular melanoma in Denmark, 1943–1989. Int J Cancer 1995;61:773–9.
[67] Han J, Kraft P, Nan H, Guo Q, Chen C, Qureshi A, et al. A genome-wide association study identifies novel alleles associated with hair color and skin pigmentation. PLoS Genet 2008;4:e1000074.
[68] Pho LN, Leachman SA. Genetics of pigmentation and melanoma predisposition. G Ital Dermatol Venereol 2010;145:37–45.
[69] Bloch DB, de la Monte SM, Guigaouri P, Filippov A, Bloch KD. Identification and characterization of a leukocyte-specific component of the nuclear body. J Biol Chem 1996;271:29198–204.
[70] Dent AL, Yewdell J, Puvion-Dutilleul F, Koken MH, de The H, Staudt LM. LYSP100-associated nuclear domains (LANDs): description of a new class of subnuclear structures and their relationship to PML nuclear bodies. Blood 1996;88:1423–6.
[71] Ling PD, Peng RS, Nakajima A, Yu JH, Tan J, Moses SM, et al. Mediation of Epstein-Barr virus EBNA-LP transcriptional coactivation by Sp100. EMBO J 2005;24:3565–75.
[72] Madani N, Millette R, Platt EJ, Marin M, Kozak SL, Bloch DB, et al. Implication of the lymphocyte-specific nuclear body protein Sp140 in an innate response to human immunodeficiency virus type 1. J Virol 2002;76:11133–8.
[73] Page-McCaw PS, Amonlirdviman K, Sharp PA. PUF60: a novel U2AF65-related splicing activity. RNA 1999;5:1548–60.
[74] Duncan R, Bazar L, Michelotti G, Tomonaga T, Krutzsch H, Avigan M, et al. A sequence-specific, single-strand binding protein activates the far upstream element of c-myc and defines a new DNA-binding motif. Genes Dev 1994;8:465–80.
[75] Liu J, He L, Collins I, Ge H, Libutti D, Li J, et al. The FBP interacting repressor targets TFIIH to inhibit activated transcription. Mol Cell 2000;5:331–41.
[76] Kovalevska LM, Yurchenko OV, Shlapatska LM, Berdova GG, Mikhalap SV, Van Lint J, et al. Immunohistochemical studies of protein kinase D (PKD) 2 expression in malignant human lymphomas. Exp Oncol 2006;28:225–30.
[77] Miyamoto Y, Yamauchi J, Itoh H. Src kinase regulates the activation of a novel FGD-1-related Cdc42 guanine nucleotide exchange factor in the signaling pathway from the endothelin A receptor to JNK. J Biol Chem 2003;278:29890–900.
[78] Ikeda M, Longnecker R. The c-Cbl proto-oncoprotein downregulates EBV LMP2A signaling. Virology 2009;385:183–91.
[79] Gudmundsson J, Sulem P, Manolescu A, Amundadottir LT, Gudbjartsson D, Helgason A, et al. Genome-wide association study identifies a second prostate cancer susceptibility variant at 8q24. Nat Genet 2007;39:631–7.
[80] Easton DF, Pooley KA, Dunning AM, Pharoah PD, Thompson D, Ballinger DG, et al. Genome-wide association study identifies novel breast cancer susceptibility loci. Nature 2007;447:1087–93.
[81] Tomlinson I, Webb E, Carvajal-Carmona L, Broderick P, Kemp Z, Spain S, et al. A genome-wide association scan of tag SNPs identifies a susceptibility variant for colorectal cancer at 8q24.21. Nat Genet 2007;39:984–8.
[82] Yeager M, Orr N, Hayes RB, Jacobs KB, Kraft P, Wacholder S, et al. Genome-wide association study of prostate cancer identifies a second risk locus at 8q24. Nat Genet 2007;39:645–9.
[83] Amundadottir LT, Sulem P, Gudmundsson J, Helgason A, Baker A, Agnarsson BA, et al. A common variant associated with prostate cancer in European and African populations. Nat Genet 2006;38:652–8.
[84] Kiemeney LA, Thorlacius S, Sulem P, Geller F, Aben KK, Stacey SN, et al. Sequence variant on 8q24 confers susceptibility to urinary bladder cancer. Nat Genet 2008;40:1307–12.
[85] Pomerantz MM, Ahmadiyeh N, Jia L, Herman P, Verzi MP, Doddapaneni H, et al. The 8q24 cancer risk variant rs6983267 shows long-range interaction with MYC in colorectal cancer. Nat Genet 2009;41:882–4.
[86] Shaffer AL, Emre NC, Lamy L, Ngo VN, Wright G, Xiao W, et al. IRF4 addiction in multiple myeloma. Nature 2008;454:226–31.