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REVIEW ARTICLE

Transformation of Indolent B-Cell Lymphomas

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ABSTRACT

Histologic transformation (HT) to an aggressive lymphoma is a well-described event in the natural history and clinical course of patients with so-called indolent lymphomas. This phenomenon has been studied most extensively in patients with follicular lymphoma and subsequent transformation to a diffuse large B-cell lymphoma, with little literature on HT in nonfollicular lymphomas. Despite a considerable body of information on the pathologic and molecular events associated with HT, its pathogenesis has remained elusive and the molecular information available has not been translated into clinical advances. It remains unclear if there is already a predisposition to HT and whether this can be detected at the time of diagnosis. The rituximab era has been characterized by a significant improvement in the prognosis of patients with B-cell lymphomas, but HT remains one of the most important challenges in the management of patients with indolent lymphoma, the difficulties starting with the diagnosis and definition of HT and ending with the appropriate management and treatment of the event. Going forward, it is crucial to incorporate HT as a major end point in clinical trials and to include patients with HT as subject of such studies if we are to see meaningful progress in the future.

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INTRODUCTION

The natural history and the clinical course of patients with B-cell indolent lymphomas are characterized, among other features, by the risk of histologic transformation (HT) to an aggressive lymphoma. This is a well-known phenomenon that has been extensively studied in terms of the incidence, risk factors, and outcome in patients with follicular lymphoma (FL), which transforms to a diffuse large B-cell lymphoma (DLBCL; and, less commonly, to Burkitt lymphoma [BL] or other types of aggressive lymphomas), and the vast majority of clinical data available in the literature therefore refers to transformed FL (tFL).[1-10] Although there are few large published series, HT has also been described in other subtypes of B-cell indolent lymphoma, including small lymphocytic lymphoma (SLL)/chronic lymphocytic leukemia (CLL) transforming to either DLBCL (Richter's syndrome [RS])[11-13] or Hodgkin's lymphoma (HL),[14] mucosa-associated lymphoid tissue (MALT) or lymphoplasmacytic lymphoma (LPL) to DLBCL,[15-19] and nodular-lymphocyte predominant HL (NLP-HL) to DLBCL (Table 1).[20-21] Indolent T-cell disorders can also transform to more aggressive histologic subtypes of lymphoma although this falls outside the scope of this review. Somewhat disappointingly, and despite the fact that our knowledge of the molecular events related to HT has significantly increased in recent years, its pathogenesis remains unclear. In fact, whereas the term transformation implies that there is a direct evolution from the indolent to the aggressive lymphoma, this might not be the case, as will be discussed later. The molecular information available has not answered the crucial clinical questions of why only some patients experience HT and whether it can be prevented. Likewise, many series have analyzed the clinical risk factors at diagnosis that are associated with a higher risk of HT during follow-up, especially with regards to tFL, but, again, the weight of evidence has not been sufficiently compelling to result in a practical approach to either prevent HT or to improve the outcome of patients in whom it occurs. This is of the utmost importance given its poor prognosis. Despite some reports suggesting that the outcome of patients with transformed lymphoma has significantly improved in the rituximab era,[22,23] HT remains one of the major challenges in the management of patients with indolent lymphomas.

DEFINITION, DIAGNOSIS, AND INCIDENCE OF HT

Whereas the definition of HT in patients diagnosed with SLL/CLL, LPL, MALT, or HL who subsequently present a different histologic subtype of lymphoma is straightforward, the definition of tFL

Table 1. Histologic Transformation in B-Cell Lymphoma

Indolent Lymphoma	Transformed Lymphoma	Reference
Follicular lymphoma	Diffuse large B-cell lymphoma	4, 6-9
Follicular lymphoma	Burkitt lymphoma	9,10
Small lymphocytic lymphoma/ chronic lymphocytic leukaemia	Diffuse large B-cell lymphoma	11-13
Small lymphocytic lymphoma/ chronic lymphocytic leukemia	Hodgkin lymphoma	14
Lymphoplasmacytic lymphoma	Diffuse large B-cell lymphoma	15
Mucosa-associated lymphoid tissue lymphoma	Diffuse large B-cell lymphoma	16-19
Nodular lymphocyte-predominant Hodgkin's lymphoma	Diffuse large B-cell lymphoma	20, 21

varies considerably among different series. Some include patients with cytological progression,[4,24] others consider the existence of a diffuse pattern as the indication of HT,[2] whereas in others the diagnosis of a high-grade lymphoma[1,3-5] or more specifically DLBCL (or BL) defines HT.[8,9] Thus, HT is diagnosed in different series based on cytological samples,[6,7,9] on histologic samples[3,6,8] or in some cases on clinical grounds alone.[6,9] The diagnosis of HT should wherever possible be based on a biopsy sample, rather than relying on cytological samples (ie, fine-needle aspiration) or on clinical criteria.[25] However, it is not always possible to obtain a biopsy, either because of the poor performance status of the patient or because of progression of the disease in inaccessible areas. Al-Tourah et al[9] analyzed the incidence of HT in a population-based study, although it is worth mentioning that the median age in this series was 48 years, with patients older than 60 years having been excluded. Thirty-six percent of patients were diagnosed with HT based on clinical criteria, which included the presence of any of the following: a rise in the lactate dehydrogenase level, a rapid localized nodal growth, new extranodal sites of disease, the presence of B symptoms or hypercalcemia. The survival after transformation for patients diagnosed based on the above clinical criteria was comparable to that in patients diagnosed with HT based on a pathologic sample, supporting the reliability of the clinical criteria to diagnose transformation. It has to be taken into account, however, that whereas there is a clear association between a more aggressive clinical behavior and the diagnosis of HT, this relationship is not absolute. In the St Bartholomew's series, only 25% of the patients with a biopsy demonstrating tFL at the time of first progression had two or more extranodal sites of disease and only 22% had a poor performance status.[8] The results from the British Columbia Cancer Agency (BCCA) study[9] support the notion that patients with an aggressive clinical picture suggesting transformation in which it is not feasible to obtain a biopsy, should be treated aggressively as if they have HT (which is not equivalent to say that these patients have HT). In fact, a not uncommon clinical scenario is that of an unwell patient with clinical features highly suggestive of transformation in which the biopsy shows persistence of the indolent lymphoma with no evidence of HT. This may be a function of the site of the biopsy. Several studies have reported on the clinical utility of an [^{18}F]fluorodeoxyglucose (FDG) positron emission tomography (PET) scan to detect areas suspicious of transformation, demonstrating the correlation between a higher standardized uptake value on a FDG-PET and a more aggressive histology,[26-28] and leading to pilot studies assessing the efficacy of FDG-PET to direct the biopsy to areas highly suspicious of HT.[29] Of note, the original indolent lymphoma subtype can reappear in patients with disease progression after having responded to salvage therapy for HT,[5,30] emphasizing the need of repeat biopsies at each progression, even after transformation.

The considerable variability in the incidence of HT reported in different series may be explained by differences in the population included, the definition of transformation, and the diagnostic method. As HT is a time-dependent event, the length of follow-up also contributes to such variability, as well as the percentage of patients with a repeat biopsy at the time of progression and the inclusion of necropsy data (Table 2).[31,32] Allowing for these differences among series, the risk of transformation for patients with FL is around 20% at 5 years and 30% at 10 years (Fig 1).[6,8,9] In the BCCA study, a continuous risk of 3% per year was reported, with, according to the authors, no evidence of a plateau after a median follow-up of 9 years.[9] However, in the St Bartholomew's series, with a median follow-up of 15 years, no cases of HT were observed after 16 years, at the time of publication, suggesting that there might be a population of patients in whom HT does not occur despite a very prolonged follow-up.[8] Similar results have been reported by Bastion et al[6] in a series with a median follow-up of 9 years, and in patients with CLL.[13] The prevalence of HT in other types of indolent lymphoma is significantly lower. Thus, in a study including 3,986 patients with SLL/CLL at the MD Anderson Cancer Center, RS was demonstrated either by biopsy or fine-needle aspiration in 4% of patients,[12] whereas transformation to HL was even rarer, being detected only in 0.4% of patients.[14] A smaller study reported a 10-year incidence of RS of 16% with a prevalence of 9%.[13] It is important to bear in mind, though, that the distinction between secondary lymphoma (ie, transformed lymphoma) and second lymphoma in a disease like CLL characterized by a high incidence of second malignancies is not always easy and requires molecular studies.[33-35] Two series on the risk of HT in patients with NLP-HL have been published recently. The French registry reported a 10-year risk of transformation of 12% in 164 patients,[20] whereas the BBCA found the risk to be 7% at 10 years.[21] The largest series on the risk of HT in patients with LPL reported the development of DLBCL during follow-up in 11% of patients.[15]

PATHOGENESIS OF HISTOLOGIC TRANSFORMATION

Despite our best efforts, little progress has been made in defining a consensus regarding the genetic basis of FL transformation. This is in sharp contrast with the considerable amount of information available on the molecular events related to this process, which has been the subject of two recent reviews.[25,36] This lack of success reflects the difficulty in obtaining biopsy-proven paired FL and tFL, the long clinical evolution of the disease and the requirement for life-long follow-up, all of which has led to a reliance on diagnostic samples as means of generating a molecular predictor of an event that may arise several years from diagnosis. Given the allegedly inherent nature of FL cells to transform, an assumption that some tFL programming is already imprinted at diagnosis is reasonable. Whereas pretreatment

Table 2. Incidence of Histologic Transformation in Patients With Indolent Lymphoma

Series	No.	Population	Diagnosis	Criteria	Bx at Relapse (%)	Median Follow-Up (years)	Prevalence (%)	Actuarial Risk (%)
Risdall[31]	35	Nodular NHL	PM Bx	Diffuse pattern (Rappaport)	—	—	63	—
Cullen[1]	64	Low-grade NHL	Bx	High-grade (Kiel)	75	—	27 (of Bx)	—
Hubbard[2]	205	Nodular NHL	Bx	Diffuse pattern (Rappaport)	No. = 63/205 (31)	—	30 (of Bx)	—
Acker[3]	150	Favorable histology	Bx	Unfavorable incl NH (Rappaport)	62	—	29 (of relapses)	5 year: 20 8 year: 60
Garvin[32]	56	Nodular NHL	PM Bx	Diffuse pattern	—	—	64	—
Horning[24]	83/131*	Low-grade NHL (advanced stage)	Bx	Diffuse pattern large cell FL	45/67*	4	12/18* (of relapses) 44/42* (of Bx)	8 year: 19/23*
Ersbøll[4]	127	FL (F-SC, F-M)	Bx	Intermediate/high-grade (WF)	65	11	24 38 (of relapses)	5 year: 30 (relapses) 10 year: 56 (relapses)
Bastion[6]	220	FL	Bx, Cx, Cl	Aggressive diffuse lymphoma	—	9	24 37 (of relapses)	5 year: 22 10 year: 31
Giné[7]	276	FL	Bx, Cx	High-grade diffuse lymphoma	NS	6.5	—	10 year: 15 15 year: 22
Montoto[8]	325	FL	Bx	DLBCL	70	15	27	10 year: 28 15 year: 37
Al-Tourah[9]	600	FL < 60 years	Bx, Cx, Cl	DLBCL or BL	NS	9	28	10 year: 30 15 year: 45

Abbreviations: Bx, biopsy; NHL, non-Hodgkin's lymphoma; PM, post-mortem; Cx, cytology; Cl, clinical; NH, nodular histiocytic; NS, not specified; F-SC, follicular small cell; F-M, follicular mixed; WF, Working Formulation; DLBCL, diffuse large B-cell lymphoma; BL, Burkitt lymphoma.
*Expectant management/initially treated, respectively.

biopsies are the obvious starting point to focus attention on the 15% to 20% of patients with FL who develop early progressive or transformed disease, their use for inferring later transformation is questionable. Gene-expression profiling studies in FL diagnostic cases have been successful in predicting immediate clinical behavior, but its use to discriminate between long-term survivors[37] or risk of transformation has been somewhat disappointing.[38]

So are we any closer to determining why transformation occurs only in some patients? The reanalysis of available FL and paired FL-tFL gene-expression profiles by the Stanford group offers some encouragement.[39] Employing a modular network approach they identified an embryonic stem cell (ESC) signature at diagnosis that predicts HT. This does not imply that tFL arises from a hematopoietic stem cell but rather from a germinal center B-cell with enhanced stem cell expression features. The emergence of this ESC signature may reflect early epigenetic remodeling within the B-cell population with t(14;18), with recent studies demonstrating specific hypermethylation of polycomb repressor complex two target genes in these tumors. This suggests that some programmed global change in gene expression may be a prerequisite for onset of FL or tFL.[40,41] These hypotheses come at a time when the previous assumptions that transformation merely reflects the emergence of a more aggressive subclone of cells from an existing FL population are under review, pointing out the inaccuracy of the term transformation. This idea is further supported by clinical data, with evidence of recurrences after HT as the initial indolent histologic subtype.[5,30] The current notion that tFL (and indeed relapsed FL) may arise from a more immature cell, referred to as a common ancestor cell[42] or common progenitor cell (CPC; Fig 2)[43] is based on several lines of genetic evidence,[43-50] all of which have traced the genetic changes in sequential biopsy samples taken over the clinical course of a patient's disease. Taken together, the evidence suggests that in some patients the transformed tumor originates from a more undifferentiated B-cell population rather than reflecting clonal evolution from a previous episode of the disease.

The acquisition of an ESC signature within this CPC pool of cells in FL may very well mark patients for later transformation, and our ability to predict transformation might depend on the quantity of such cells within biopsies and the corresponding strength of the HT expression signature (Fig 2). These cells may be reminiscent of t(14;18) bearing cells detected in blood of healthy individuals,[49,50] which survive long-term in the bone marrow niche, and acquire the additional genomic lesions or perhaps lay dormant for the correct host microenvironment to provide the optimal selective pressure to thrive. Although this germinal center B-cell population has not been characterized, multiplexed flow cytometry approaches have been developed by Irish et al.[51,52] Their identification of B-cell receptor insensitive cells that appear to increase in number at the time of tumor progression is an exciting new observation and may offer a technical approach for sampling rare cell populations within FL biopsy, in search of a formal characterization of this proposed CPC population.

The application of high throughput sequencing approaches in FL, pioneered by the BCCA group,[53] is likely to identify many new mutational targets, some of which might be linked directly with a risk of HT. Irrespective of the pace of these new advances, it is practical in light of the evolutionary complexity of FL to monitor the genetic and microenvironment characteristics of FL both at initial diagnosis and during follow-up in order to improve our understanding and management of relapsed FL and HT.

RISK FACTORS AND CLINICAL CHARACTERISTICS AT TRANSFORMATION

Several studies have analyzed the clinical characteristics at diagnosis associated with a higher risk of HT during follow-up in an attempt to

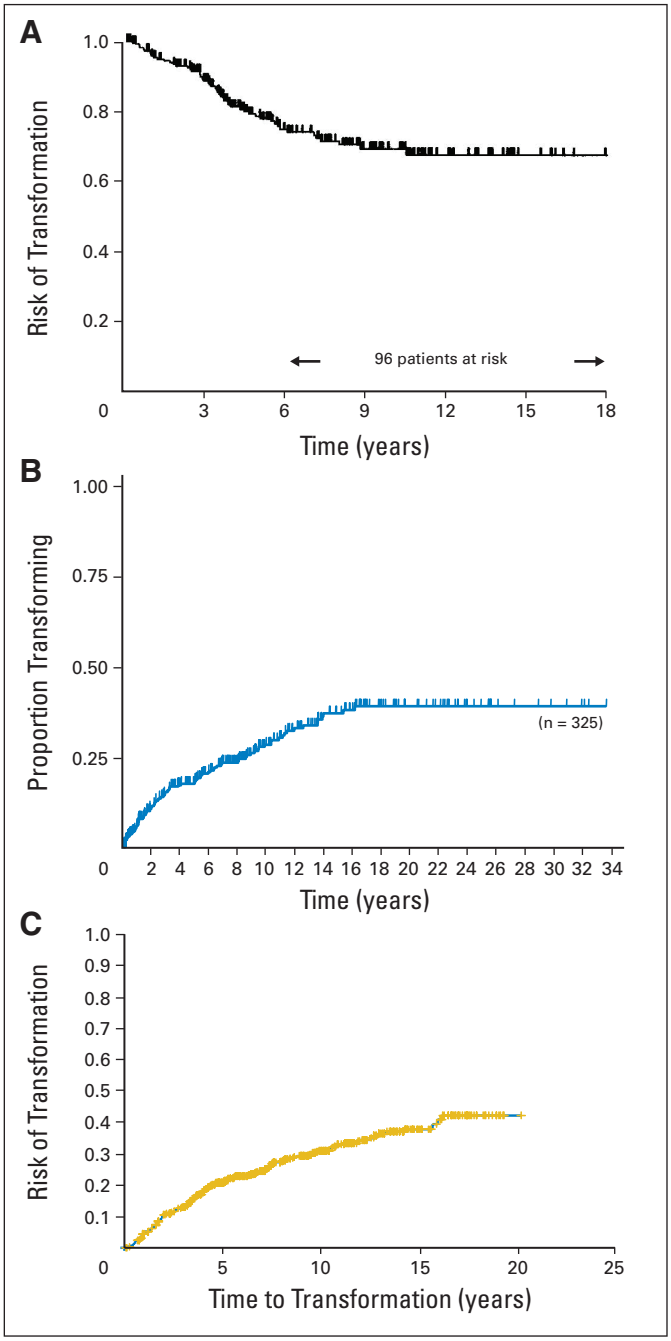

Fig 1. Risk of histologic transformation in patients with follicular lymphoma in series with a long follow-up. Adapted from (A) Bastion et al,[6] (B) Montoto et al,[8] (C) Al-Tourah et al.[9]

identify the population at risk. Most information comes from studies in FL. In two large series, the risk of HT was higher in those presenting with advanced-stage disease,[8,9] although this was not seen in an older study.[2] The Follicular Lymphoma International Prognostic Index[7,8] and the International Prognostic Index for aggressive lymphomas[8] at diagnosis have demonstrated their predictive value for the risk of HT at some point during the course of the disease. This is not surprising since these scores are merely indicators of a poor outcome, reflecting both tumor load and characteristics of the patients; however, they are not markers of susceptibility or predisposition to transformation. Therefore, its association with HT has not resulted in any practical approach to improve the outcome of patients with transformation. More controversial, but with potentially more important clinical implications, are the data on the risk of HT according to the initial management. The impact of expectant management at diagnosis on the risk of HT is unclear: it was unexpectedly associated with a higher risk of transformation in the St Bartholomew's series,[8] but not in another retrospective study.[24] Only one randomized study comparing expectant management at diagnosis with other approaches has reported on the risk of HT. A Groupe d'Etude des Lymphomes de l'Adulte study compared expectant management versus prednimustine versus interferon and found no differences in the risk of HT among the three arms.[54] In contrast, there was a suggestion of a higher risk of HT for patients randomly assigned to watchful waiting in a National Cancer Institute study,[55] however, this study has never been fully published. There is also some controversy regarding to the influence of the type of initial chemotherapy on the likelihood of transformation. Two cohorts of patients treated on serial phase II trials at BCCA were compared.[9] Those that had received combination chemotherapy including bleomycin, cisplatin, etoposide, doxorubicin, cyclophosphamide, vincristine, and prednisone had a significantly lower risk of transformation than the group treated with an alkylator plus a purine analog (10-year risk of transformation: 18% *v* 30%). However, in another retrospective study, no differences in the risk of transformation were detected according to whether patients had received doxorubicin as part of the initial treatment or not.[7] The decision to use doxorubicin-containing regimens was based on the local policy at the time and varied over the years. The lack of impact of the use of anthracyclines on the risk of HT is further supported by an older randomized study comparing PCOP (cyclophosphamide, procarbazine, vincristine, and prednisone) with PACOP (cyclophosphamide, procarbazine, vincristine, and prednisone with doxorubicin), which did not show any differences between the two arms in terms of histologic progression.[56] However, it is difficult to draw firm conclusions from this small study and, hence, there is not strong evidence to decide on the initial management based on a hypothetical subsequent risk of transformation. Unfortunately, no recent randomized studies on first-line treatment in FL have included the risk of HT as an end point. There has also been some suggestion that specific treatments influence the risk of transformation in patients with CLL after a series of 101 patients treated with fludarabine that reported a higher risk of RS (10%) than that published previously.[57] In this regard, the incidence of RS in a randomized study was 7% and 2% in patients receiving fludarabine or chlorambucil, respectively, in the older population,[58] but with no differences observed in younger patients.[59] The Leukemia Research Fund CLL4 trial including 777 patients treated with fludarabine, chlorambucil, or fludarabine plus cyclophosphamide did not find any differences in the frequency of RS among the three arms,[60] although follow-up remains short.

HT is accompanied in the majority of the patients by a change in the pace of the disease to a more aggressive clinical behavior and in most of the series it is associated with clinical characteristics typical of aggressive lymphomas, such as a declining performance status,[7,13,21] rapidly growing or asymmetric growth of lymphadenopathy,[13] involvement of extranodal sites,[13,15,21] hypercalcemia,[9] and raised lactate dehydrogenase level.[6-8,12,15,21]

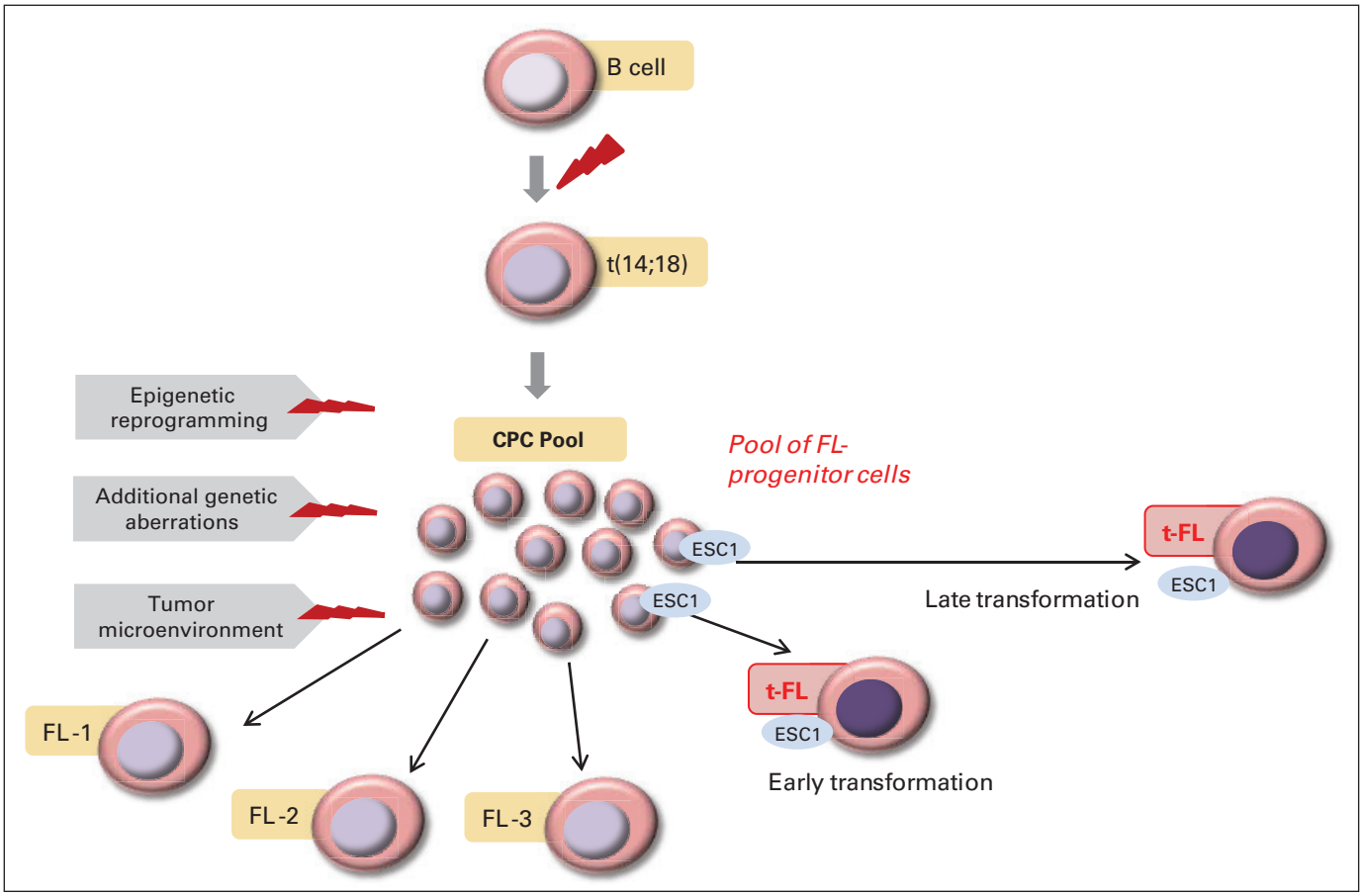

Fig 2. Model of histologic transformation of follicular lymphoma (FL). t(14;18) positive B-cell undergoes an epigenetic reprogramming, which in conjunction with the acquisition of additional genetic events leads to the development of an FL-initiating pool of common progenitor cells (CPCs). The emergence of a particular FL-CPC clone at diagnosis (FL-1) or relapse (FL-2, FL-3) is dependent on the selective pressure of these acquired genetic mutations and the cellular microenvironment that may differ between CPCs. Cells that have acquired a particular embryonic stem cell 1 (ESC1) modular expression profile rise to transformed FL (tFL). This model is distinct from a process of direct evolution where FL1, FL2, FL3, and tFL reflect the sequential acquisition of events from a prior episode of disease.

TREATMENT AND OUTCOME

The prognosis of patients with FL who develop HT is extremely poor, with a median survival after transformation of around 1 to 2 years,[2,3,5,7-9] although it may be improving. Several long follow-up studies have demonstrated that the overall survival (OS) of patients who develop transformation at any point during the course of the disease is significantly shorter than that in patients never diagnosed with transformation.[8,9] Patients who present at transformation with limited stage have a better outcome than the rest, and it has been suggested that they might achieve a relatively long survival.[5,7,9] Not surprisingly, a crucial factor that determines outcome after HT is the response to therapy, so that patients who achieve a complete response do significantly better than the remainder.[2,3,5-7] Being chemotherapy naive before HT has been reported to be associated with a better outcome in one study.[5] The outcome of patients with CLL who experience HT is universally poor, with a median OS of 8 to 16 months, regardless of whether they transform to DLBCL[12,13] or HL,[14] and a similar dismal prognosis is reported for patients with LPL in whom the disease transforms to DLBCL.[15] In contrast, the outcome of patients with NLP-HL with transformation to DLBCL seems to be significantly better, with the 10-year survival after transformation being reported of 60%.[20,21] Despite this, the most frequent cause of death among patients with NLP-HL in the BCCA study was secondary NHL.[21]

The question of transformation at diagnosis or the diagnosis of discordant (ie, different histologies diagnosed simultaneously in different samples: most commonly DLBCL in a lymph node and FL in the bone marrow [BM]) or composite (different histologies in the same sample) lymphoma and its potential prognostic implications is less clear, given the small size of the series reported and their heterogeneity. Earlier studies compared the outcome of patients with DLBCL and BM involvement according to its histology, and demonstrated that the prognosis of patients with large cell lymphoma in the BM was significantly worse than that of patients with low-grade lymphoma or with no BM involvement (the outcome in the latter two situations being similar).[61,62] These results have been confirmed in a recent study,[63] but not in another (only presented in abstract format).[64] In a series of patients with DLBCL not restricted to the population with BM infiltration, the presence of a low-grade component was associated with a clinical outcome more akin to

that of patients with indolent lymphoma, with a lower complete response rate and shorter progression-free survival (PFS) although similar OS,[65] whereas it is clear that patients with a diffuse component in a biopsy showing grade 3 FL (which, according to the WHO classification, should be reported and considered as DLBCL) have a poorer outcome than those with pure grade 3 FL and similar to those with DLBCL.[66] The largest and most homogeneous studies on this issue are on gastric lymphoma and the impact on outcome of the existence of a high-grade component in biopsies showing MALT lymphoma. The proportion of patients in whom DLBCL is detected at the time of MALT diagnosis varies from 17% to 36%.[16-19,67] In the two studies in which it was reported, the presence of a DLBCL component in patients with gastric MALT was not associated with a worse outcome.[18,67]

There is no standard treatment for HT, the choice of therapy depending mostly on the previous therapy. Unfortunately, patients with HT are often excluded both from indolent lymphoma studies and from DLBCL studies, so objective data on which to base decisions are scarce, and frequently the optimal management of transformed lymphoma is extrapolated from available data on DLBCL. Thus, most patients are treated with an anthracycline-containing regimen, if they have not previously received one and with a second-line regimen for DLBCL otherwise. The advent of rituximab has also made a difference in the outcome of patients with HT. Survival after transformation was significantly longer in patients that had received rituximab at the time of HT in a retrospective study,[22] whereas the group from Stanford reported an improvement in the outcome of patients diagnosed with HT in the rituximab era.[23] By analogy with the treatment of relapsed DLBCL, the response is frequently consolidated with high-dose therapy (HDT) and autologous stem cell rescue. However, the evidence to support or disregard such an approach is limited. The European Group for Blood and Marrow Transplantation registry reported that the outcome of patients who receive HDT for transformed lymphoma is similar to that seen in patients having HDT either for FL or for DLBCL.[68] Whether this proves the benefit of HDT in HT or it is only a consequence of the selection of patients that actually get HDT (ie, those with a good response to salvage treatment) remains to be answered. After the advent of reduced-intensity conditioning (RIC) regimens, several groups have explored the use of RIC-allogeneic transplant in patients with HT. The results are not as good as those in patients with FL,[69] but they are not as bad as those reported in patients with DLBCL treated with RIC-allogeneic transplants.[70] However, published series are extremely heterogeneous including only small numbers of patients with HT so the results are difficult to compare, ranging from a 3-year OS and PFS around 20% to a 4-year OS and PFS of 60%.[69,70] Of note, around half the patients included in these series had previously received HDT with autologous stem cell rescue. As mentioned, most investigational studies exclude patients with HT so there are scarce data on the potential effectiveness of new drugs in the management of HT. Notwithstanding, there are some promising data reporting response rates of 57% to 79% and median PFS/response duration of around 1 year with iodine-131 tositumomab[71,72] and lenalidomide.[73] There is no information available on maintenance with rituximab after HT.

CONCLUSIONS

HT remains one of the major challenges in the management of patients with indolent lymphoma, despite a significant improvement in the outcome of patients with transformed lymphoma in the rituximab era. Even with the available clinical and molecular data, we are no closer to identifying at diagnosis those patients who are destined to transform. Indeed we cannot be certain if predisposition to HT exists at the time of diagnosis or whether this tendency is confined to a rare population of FL cells that may be below the threshold of current detection methods, making the identification at diagnosis of patients at high risk impossible. Better clinical and molecular predictors of HT need to be identified, perhaps as a first step restricting the focus to distinguish patients with risk of early HT, particularly when information available is limited to the time of diagnosis. Not least important, we also lack the tools to treat this complication, either at diagnosis in susceptible patients or when HT arises.

The way ahead must include obtaining a biopsy not only when HT is clinically suspected but at each recurrence, since an accurate histologic diagnosis will contribute to a better understanding of the pathogenesis of HT and to unravel the mechanism(s) leading to transformation. Ideally, this should result in the development of targeted therapies either to prevent HT in patients at high risk or to improve on current salvage approaches. In any event, if molecular data are to be integrated into routine clinical practice, our current research tools must be made amenable for use on formalin-fixed, paraffin-embedded material, the current standard preparation of tissue biopsies. Not only would this increase the number of FL and paired samples available for biomarker discovery projects, it would also serve as a driver for more careful evaluation of existing markers. In the meantime, rituximab should be part of the treatment of HT (although there are not strong evidence-based data to support this) and, most importantly, the current climate of excluding patients with HT from clinical trials must be re-evaluated to allow testing the potential efficacy of new drugs in this poor prognosis population.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

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