Follicular non-Hodgkin lymphoma grades 3A and 3B have a similar outcome and appear incurable with anthracycline-based therapy

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Background: The revised World Health Organization (WHO) classification maintains a histological grading system (grades 1–3) for follicular lymphoma (FL) and subdivides grade 3 into 3A (FL3A) and 3B (FL3B) subtypes. Optimal therapy of FL grade 3 and its potential curability with anthracycline-based chemotherapy remain uncertain.

Patients and methods: We carried out a retrospective population-based analysis evaluating the clinical characteristics and outcome of FL3A and FL3B as strictly defined by WHO diagnostic criteria. Using the BC Cancer Agency Lymphoid Cancer Database, 161 patients with FL grade 3 were identified and, following detailed pathology review, composed of 139 with FL3A and 22 with FL3B.

Results: Patients with FL3B had a higher overall International Prognostic Index (IPI) score than FL3A patients ($P = 0.03$), though no significant difference in individual IPI risk factor frequencies was noted. More patients with FL3B received front-line anthracycline-containing chemotherapy (82% versus 36%, $P \leq 0.001$). With median follow-up of 45 months, no difference in disease-specific survival ($P = 0.74$) or overall survival (OS) ($P = 0.87$) was found between FL3A and FL3B and no survival curve plateau was observed. Analysis limited to FL3A patients showed no OS advantage with front-line anthracycline use ($P = 0.33$).

Conclusion: Using strict diagnostic criteria, there appears to be no difference in outcome between patients with FL3A and FL3B and no evidence of curability with anthracycline-based therapy.

Key words: anthracyclines, follicular lymphoma, follicular lymphoma grade 3, prognosis
The discrepant findings among the various reports of FL grade 3 are likely due to several factors. Most studies have been based on small patient cohorts that have been variably treated. Some studies predate the WHO numerical grading system and utilize the prior pathological definition of ‘follicular large cell lymphoma’ (FLCL) [3–7]. A lack of reproducibility among pathologists when grading FL has been demonstrated and may contribute significantly to the heterogeneity between study populations [16, 17]. Finally, some reports have included cases with a substantial diffuse component of centroblasts on biopsy, which more appropriately should be considered DLBCL [3–8].

Using a retrospective population-based study design, we identified a large cohort of unselected patients with pure FL grade 3. Initial biopsies on all patients were reviewed and classified according to the updated WHO diagnostic criteria [2]. Our objectives were to characterize the clinical features and outcome of patients with FL3A and FL3B and to assess the survival impact of anthracycline-containing chemotherapy within these subtypes.

**patients and methods**

**study population**

This is a retrospective population-based study employing the British Columbia (BC) Cancer Agency Lymphoid Cancer Database. All patients older than 17 years with a diagnosis of FL grade 3 between the years 1982 and 2008 and with initial biopsies available for review were included in this analysis. All biopsies were centrally reviewed (BS and RDG) and were classified according to the updated WHO diagnostic criteria. Patients with primary cutaneous FL were excluded from this study. Biopsies containing a diffuse component of centroblasts as defined by the WHO were designated as DLBCL with FL and were also excluded.

Clinical data, including baseline patient characteristics, initial therapy and follow-up information, were retrieved through the Lymphoid Cancer Database, electronic patient records and primary physicians’ records. This study was reviewed and approved by the University of British Columbia–BC Cancer Agency Research Ethics Board.

**statistical analysis**

Univariate comparisons of patient characteristics were made using the Student’s t-test for continuous variables and the Pearson’s chi-square test for categorical outcomes. A two-sided P value of <0.05 was considered to be statistically significant. Survival analyses were plotted using the Kaplan–Meier method and compared using the log-rank test [18, 19]. Overall survival (OS) was calculated from time of diagnosis to death from any cause or date last known alive. Disease-specific survival (DSS) was calculated from time of diagnosis to death from lymphoma or treatment-related toxicity or date last known alive; deaths from other causes were censored on the date of death. The Statistical Package for the Social Sciences (SPSS) software was used for statistical analysis (version 11.0; SPSS Inc., Chicago, IL).

**results**

**clinical characteristics and initial therapy**

A total of 161 patients met criteria for study inclusion and were subclassified as FL3A (n = 139) or FL3B (n = 22). Representative images of the lymph node histology are shown in Figure 1. Baseline clinical characteristics and initial treatment details according to the subtype are compared in Table 1. Median age of the entire cohort was 63 years (range 18–88 years) and was not different between the subgroups. No significant difference in frequencies of individual International Prognostic Index (IPI) risk factors was noted between FL3A and FL3B subgroups, though there was a trend toward worse performance status in the FL3B subgroup. Overall, the proportion of patients with high-intermediate or high-risk IPI scores (IPI 3–5) was greater in the FL3B subgroup (36% versus 17%, P = 0.03).

Initial treatment information was available for 154 of the 161 patients. Patients received various initial therapies based on individual physicians’ discretion, including anthracycline-containing chemotherapy, non-anthracycline-containing chemotherapy, radiation therapy alone, and expectant management. A larger proportion of patients with FL3B received an anthracycline-containing regimen as initial therapy (82% versus 36%, P < 0.001). Approximately one-third (29%) of the entire cohort received rituximab as part of their initial treatment, with a trend toward greater usage in the FL3B subgroup.

**clinical outcome**

Median follow-up duration for the entire cohort was 45 months (range 1–239 months). Among 139 patients with FL3A, there were 36 lymphoma-related deaths and 12 deaths from other causes, with 89 patients alive at the time of last follow-up and 2 patients lost to follow-up. Among 22 FL3B patients, there were 8 lymphoma-related deaths and 1 unrelated death, with 13 patients alive at the time of last follow-up. Kaplan–Meier

![Figure 1](original_article Figure 1. Representative histology of follicular lymphoma grades 3A and 3B. (A) Grade 3A. There are >15 centroblasts per high-power field, with admixed centrocytes. (B) Grade 3B. Solid sheets of centroblasts are present.  )
analyses of OS ($P = 0.87$) and DSS ($P = 0.74$) demonstrated no significant difference in outcome between FL3A and FL3B subgroups (Figure 2). The 5-year OS (72% versus 71%) and median OS (10.1 versus 8.3 years) were similar for FL3A and FL3B, respectively. Kaplan–Meier analyses of OS ($P = 0.33$) and DSS ($P = 0.83$) limited to the FL3A cohort revealed no difference between those who received an initial anthracycline-based regimen and those who did not (Figure 3). IPI score did not differ significantly between FL3A patients who did and did not receive an anthracycline-containing regimen as initial therapy ($P = 0.09$). No survival curve plateau was seen either for the FL3A or FL3B subgroups or for the subset of FL3A patients receiving an anthracycline-containing regimen as initial therapy.

comparison with FL grades 1 and 2

In order to compare outcomes between patients with FL grade 3 and FL grades 1–2, a historical cohort of patients with FL grades 1–2 diagnosed within the same time frame (1982 to 2008) was identified. Patients were included if they had a pathological diagnosis of FL grades 1–2 previously centrally reviewed at the BC Cancer Agency and had available clinical information. A total of 1885 patients were identified (1268 FL grade 1 and 617 FL grade 2). Interestingly, there was no difference in OS ($P = 0.11$) or DSS ($P = 0.17$) between patients with FL grades 1–2 and FL grade 3 (Figure 4).

discussion

The correlation between histological grade and clinical behavior in FL is a subject of ongoing controversy. Within the earlier Working Formulation, FL with increased centroblast numbers was designated FLCL and classified as an ‘intermediate-grade’ lymphoma [20]. Subsequently reclassified as FL grade 3, this entity has been reported to behave more aggressively than FL grades 1–2 and has been treated most commonly with anthracycline-based DLBCL-appropriate regimens [3–8]. FL grading has been complicated by problems with diagnostic reproducibility. In both the earlier Working Formulation and the Revised European-American Lymphoma classification [20, 21], exact centroblast numbers were not specified for FL grading. In studies of diagnostic concordance in FL grading, significant interobserver variability has been described [16, 17]. To a great extent, this inability to achieve consensus in the grading of FL results from a lack of agreement on the specific cytological features for distinguishing large centroblasts, small centroblasts, large centrocytes and follicular dendritic cells. This lack of diagnostic consistency likely contributes to differences in outcome between previous clinical studies, preventing a clear determination of survival in FL grade 3.

The 2001 WHO diagnostic criteria standardized histological grading in FL, separating FL into grades 1–3 based on the number of centroblasts within neoplastic follicles. This diagnostic system also addressed the problem of composite histology, in which coexisting diffuse and FL components are present in a diagnostic biopsy. According to the WHO definitions, separate diagnoses of FL and DLBCL are to be reported in this situation, which likely represents early
A histological transformation of FL to DLBCL [1]. In the newly updated 2008 WHO classification, the FL grading system described in the previous edition is maintained and the separate reporting of diffuse and follicular components within the biopsy is emphasized.

The 2001 WHO classification system also introduced the subdivision of FL grade 3 into FL3A and FL3B [1, 2]. Recently, cytogenetic and molecular studies have suggested that FL grades 1–3A form a spectrum of similar biology, while FL3B is more closely related to de novo DLBCL. Ott et al. [12, 13] have reported that FL3B exhibits a lower frequency of CD10 antigen expression and t(14;18) chromosomal translocation, both markers seen in a high proportion of FL grades 1–3A. By contrast, cytoplasmic immunoglobulin expression and the presence of chromosome band 3q27 rearrangement, features associated more commonly with DLBCL, were seen more commonly in FL3B than in FL3A. Despite these molecular insights, the clinical relevance of the FL3A/FL3B subdivision remains unclear.

Our objective was to determine clinical outcome in a large retrospective cohort of patients with FL3A/3B, strictly defined according to the current WHO diagnostic criteria. In keeping with these criteria, our study population excluded all patients with a diffuse lymphoma component at initial diagnostic biopsy. Our findings indicate a survival pattern in FL3A/FL3B similar to that of FL grades 1–2, with continuous relapses and no evidence of curability even after anthracycline-based chemotherapy. This pattern of continuous relapse occurred despite the first-line use of anthracycline-based chemotherapy in >80% of FL3B patients and in ~40% of FL3A patients. These results are consistent with an earlier study by Miller et al. [9], which examined clinical outcome in 53 patients with FLCL treated on various anthracycline-based Southwest Oncology Group protocols and found an ongoing risk of late relapse with a long median follow-up duration of 17 years.

To date, no studies have demonstrated a difference in outcome between FL3A and FL3B, although FL3B cohort sizes have typically been small due to the rarity of this entity [8, 10, 11, 15]. In our study, no difference in either OS or DSS was noted, although significantly fewer patients with FL3A received anthracycline-containing chemotherapy. However, an analysis limited to the FL3A cohort showed no survival advantage in association with first-line anthracycline use. This absence of anthracycline benefit is in agreement with several smaller prior analyses [10, 11]. Although this retrospective study could not control for differences in clinical features between treatment groups, our findings suggest that FL3A/3B patients may be managed with strategies similar to those of FL grades 1–2.
Table 2. Prior clinical studies of FL grade 3

<table>
<thead>
<tr>
<th>Study</th>
<th>N</th>
<th>Median follow-up duration, years</th>
<th>Median overall survival, years</th>
<th>Overall survival % (number of years)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anderson et al. [3]</td>
<td>107</td>
<td>2</td>
<td>NR</td>
<td>70 (3)</td>
</tr>
<tr>
<td>Bartlett et al. [4]</td>
<td>96</td>
<td>5.2</td>
<td>NR</td>
<td>75 (5)</td>
</tr>
<tr>
<td>Wendum et al. [5]</td>
<td>89</td>
<td>5.4</td>
<td>NR</td>
<td>59 (5)</td>
</tr>
<tr>
<td>Miller et al. [9]</td>
<td>53</td>
<td>17</td>
<td>6.9</td>
<td>57 (10)</td>
</tr>
<tr>
<td>Rodriguez et al. [6]</td>
<td>100</td>
<td>5.6</td>
<td>11.8</td>
<td>72 (5)</td>
</tr>
<tr>
<td>Rodriguez et al. [7]</td>
<td>62</td>
<td>14.7</td>
<td>5.1</td>
<td>31 (10)</td>
</tr>
<tr>
<td>Chau et al. [10]</td>
<td>55</td>
<td>6.7</td>
<td>22.2</td>
<td>78 (5)</td>
</tr>
<tr>
<td>Hsi et al. [11]</td>
<td>45</td>
<td>2.</td>
<td>3.7</td>
<td>FL3A 65 (3); FL3B 22 (3)</td>
</tr>
<tr>
<td>Ganti et al. [8]</td>
<td>227</td>
<td>9</td>
<td>9.4</td>
<td>45 (10)</td>
</tr>
<tr>
<td>Current study</td>
<td>161</td>
<td>3.8</td>
<td>FL3A 10.1; FL3A 72 (5); FL3B 8.3; FL3B 71 (5)</td>
<td></td>
</tr>
</tbody>
</table>

NR, Not reported.

subgroups, comparison of baseline prognostic features between anthracycline- and non-anthracycline-treated FL3A patients did not indicate any significant treatment bias.

Table 2 lists results from recent studies focusing on clinical outcome in FL grade 3. Our findings contrast with those of several previous analyses, which have indicated potential curability of FL grade 3 following intensive chemotherapy as in DLBCL [3–6, 8]. Differences in pathological criteria used for patient inclusion may account for these disparate outcomes. Investigators from Nebraska examined outcomes in 227 patients with FL grade 3, including 136 patients with ‘pure FL grade 3’, 41 patients with ‘FL grade 3 and a diffuse component <50%’ and 50 patients with ‘FL grade 3 and a diffuse component >50%’ [8]. A survival curve plateau indicating prolonged relapse-free survival was described in the FL grade 3 cohort, with all patients receiving an anthracycline-based regimen as initial therapy. However, in this study, patients with ‘pure FL grade 3’ and ‘FL grade 3 with a diffuse component <50%’ are analyzed as a single entity for survival patterns, potentially introducing significant heterogeneity into this cohort. In an MD Anderson study of anthracycline-treated patients between the years 1984–1998, with a median follow-up duration of 5.5 years, prolonged failure-free survival was described in a subset of FL grade 3 patients [6]. However, in an earlier cohort from the same institution, with a longer median follow-up duration of 14.7 years, a pattern of late relapse and disease-related mortality continued to be observed even after 15 years [7].

Anthracycline-containing regimens are commonly used as first-line treatment in patients with symptomatic FL, though no survival benefit has been demonstrated in prospective FL trials. Our results suggest that anthracyclines may be withheld from first-line therapy in FL grade 3A, allowing these agents to be reserved for the event of aggressive histological transformation, without compromising outcome. While the value of anthracyclines in FL3B cannot be determined from our cohort, the lack of a plateau on the survival curve also calls into question the benefit of anthracyclines in this subgroup. However, the role of anthracyclines in FL grade 3 must be reevaluated in light of the anti-CD20 monoclonal antibody rituximab, which has demonstrated OS benefit in combination with conventional chemotherapy regimens in randomized trials in FL [22, 23]. In our cohort, approximately one-third of patients received rituximab as part of their initial therapy, although due to short follow-up duration of this subset, our survival results likely do not fully reflect the impact of this agent.

In summary, our analyses indicate that FL3A and FL3B follow a similar indolent relapsing course, with outcomes that are similar to FL grades 1–2. As our data are retrospective in nature, these findings should be confirmed prospectively within the context of rituximab-based therapeutic trials, including correlative studies of pure FL grade 3 cohorts.

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disclosure

The authors declare no conflict of interest.

references


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