The aggressive non-Hodgkin’s lymphomas can be cured in more than half of the cases. However, there has been great variation in the results reported from individual clinical Phase II trials. This variation in result can be attributed to unrecognized heterogeneity in this group of diseases. Recent clinical and molecular studies have enabled us to define more homogenous population in which new therapies can be studied. For patients with advanced stages of diffuse large B cell lymphoma, a new standard of therapy exists. For patients with localized aggressive non-Hodgkin’s lymphomas, heterogeneity in patient selection prevents us from defining a new standard of care. Finally, in mantle cell lymphoma, new opportunities in drug discovery may permit advances in the treatment of this uniformly fatal malignancy.

In Section I, Dr. Richard Fisher reviews the development of combination chemotherapy for patients with advanced stage diffuse large B cell lymphoma. Because of great heterogeneity in patients enrolled in Phase II studies, large randomized Phase III studies were required in the 1980s to define CHOP has the standard of care. This heterogeneity has now been defined carefully in the international prognostic factor index and more recently by gene array studies. It will now need to be incorporated prospectively into studies or retrospectively analyzed to understand clinical trial results. The addition of rituximab to CHOP has now been demonstrated to improve survival in two large Phase III studies in elderly patients. A recently presented study in younger patients suggests a similar benefit. Thus CHOP/rituximab has become the established standard of care for all patients with advanced stage diffuse large B cell lymphoma. Other concepts being evaluated to further improve on these results include: dose intensification; initial treatment with chemotherapy plus allogeneic stem cell transplantation; and infusional chemotherapy. Finally, the status of the treatment for relapsed patients will be defined.

In Section II, Dr. Thomas Miller defines the treatment for limited stage aggressive non-Hodgkin’s lymphoma. Randomized trials have demonstrated the critical importance of initial chemotherapy for treatment of these patients. The amount of chemotherapy given needs to be increased for patients with bulky tumors. In most circumstances radiotherapy after the completion of chemotherapy has been shown to be advantageous. A modification of the international prognostic factor index for patients with early stage disease is presented to permit comparisons among different populations. Recently reported early-stage studies need to be analyzed in terms of the heterogeneity of the patients involved to understand the reported results. The addition of monoclonal antibodies, as well as radioimmunotherapy, are being tested in an effort to improve on the results for the poor prognosis patients.

In Section III, Dr. Owen O’Connor describes the pathology immunophenotype and natural history of mantle cell lymphoma. Conventional treatment strategies with combination chemotherapy achieved objective responses in approximately half of the patients but no significant impact on survival. The addition to rituximab to CHOP chemotherapy or other treatment strategies appears to improve the remission rate; however, no major changes in survival have also been reported. Excellent single institution results have been reported with HyperCVAD plus rituximab regimen, which is currently being tested in a national cooperative group trial. The most excitement in this field currently relates to the variety of new agents which appear to have significant activity in relapsed patients with mantle cell lymphoma. This includes the proteosome inhibitor, bortezomib, which is shown to have approximately a 50% response rate with some CRs and reasonable durability in early single institution Phase II studies. Larger national multi-center trials are ongoing. In addition, agents such as thalidomide, flavopiridol, and piroxantrone will be reviewed.
I. Treatment of Advanced Stage, Diffuse Large B Cell Lymphoma

Richard I. Fisher, MD*

For the majority of patients, diffuse large B cell lymphoma (DLCL-B) is a systemic disease at the time of diagnosis. At the completion of the initial staging evaluation, bulky stage II, stage III or stage IV disease is documented in approximately 75% of all DLCL-B patients. Therefore, chemotherapy is the mainstay of treatment. Although the standard chemotherapy regimen has not significantly changed over 25 years, the relatively recent incorporation of monoclonal antibody therapy into the standard treatment program represents an improvement in overall survival for the majority of patients with DLCL-B.

The study that defined the chemotherapy standard was an Intergroup trial conducted by Southwest Oncology Group (SWOG) and Eastern Cooperative Oncology Group (ECOG). In this study, previously untreated patients with stages II bulky, III, and IV disease with intermediate- or high-grade histology were randomized to one of four treatment arms: CHOP (cyclophosphamide, doxorubicin, vincristine, and prednisone), m-BACOD, ProMACE-CytaBOM, or MACOP-B. Each of the regimens was administered exactly as had been described in the prior Phase II studies. The median age of patients was 54 years, with 25% of the patients being older than 64. Actual 5-year failure-free survival varied between 33% and 38% and 5-year overall survival ranged between 45% and 46% (unpublished data). None of those differences among the treatment arms are significant. However, when fatal and life-threatening reactions were combined, significant differences were found between regimens, with CHOP and ProMACE-CytaBOM being less toxic than m-BACOD and MACOP-B (P < .001). Clinical trials comparing CHOP versus m-BACOD, CHOP versus ProMACE-CytaBOM and CHOP versus MACOP-B reached similar conclusions. These results, along with the fact that CHOP was cheaper and easier to administer than the other regimens, established CHOP as the standard therapy throughout the world. However, with a projected disease-free survival rate of 36%, it is obvious that it is far from ideal therapy, and there is clearly a need for better treatment approaches.

How could such disparate results occur between the single institution Phase II studies and the subsequent national Phase III trials? One key explanation is that DLCL, as identified in the current classification schemes, is not a uniform disease. The International Non-Hodgkin’s Lymphoma Prognostic Factors Index (IPI) utilized pretreatment prognostic factors in a sample of over 5000 patients to develop a predictive model of outcome for DLCL. The majority of patients had received adriamycin-based chemotherapy regimens. Five pretreatment characteristics were found to be independent predictors of death: age (≤ 60 vs > 60), tumor stage I or II (localized) vs. III or IV (advanced), the number of extranodal sites of involvement (≤ 1 vs > 1), patient ECOG performance status (0 or 1 [ambulatory] vs ≥ 2 [not ambulatory]), and serum lactate dehydrogenase (LDH) level (less than or equal to 1 times normal vs > 1 times normal). Each of the individual factors had comparable relative risks and thus could be summed together. The resulting model identified 4 risk groups with associated 5-year survival rates: low risk (0–1, risk factor), 73%; low intermediate risk (2 risk factors), 51%; high intermediate risk (3 risk factors), 43%; and high risk (4–5 risk factors), 26%. The increased risk of death was due to both a lower rate of complete responses and a higher rate of relapse from complete response. The percentage of patients with favorable (low) IPI scores in the National Cancer Institute original trial of ProMACE-CytaBOM (44%) was almost twice that in the subsequent SWOG-8516 Phase III study (22%). The differences in outcome caused by these imbalances in patient prognostic factors can easily exceed any treatment differences.

Currently, attempts are being made to define the prognosis of biologically defined subsets of patients with DLCL-B. The Leukemia-Lymphoma Molecular Profiling Project (LLMPP) has used complementary-DNA (cDNA) microarray techniques to demonstrate two distinct subpopulations of DLCL-B with different prognoses and different genetics. Patients with a germinal center (GC) B cell–like signature have a more favorable course than those with an activated B cell–like profile. These differences exist within each risk group identified by the IPI. Lossos et al subsequently demonstrated that they could develop a new predictor of survival of DLCL-B based on the expression of 6 genes assayed by quantitative real-time polymerase chain reaction (PCR). Knowledge of the genetics of each group may permit the future use of therapy targeted against critical cell pathways.

The monoclonal antibody rituximab has been combined with CHOP chemotherapy in an attempt to improve the therapeutic results. The GELA group randomized 399 previously untreated patients with DLCL-B, 60 to 80 years old, to receive either 8 cycles of CHOP

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every 3 weeks or 8 cycles of CHOP plus rituximab given on day 1 of each cycle. Complete response rates of 76% and 63% (P = 0.005) and 2-year overall survivals of 70% and 57% (relative risk for CHOP-R versus CHOP: 0.64, P = 0.007) were achieved by CHOP-R and CHOP, respectively. The incidence of severe or serious side effects was similar in the two treatment arms.

In patients with a low-risk, age-adjusted IPI (0 or 1 adverse prognostic factors), 1-year event-free survival (EFS) was 81% and 57% for the CHOP-R and CHOP arms, respectively (P < 0.001). For those with high-risk disease (2 or 3 adverse factors), 1-year EFS was 61% and 47% for the CHOP-R and CHOP arms, respectively (P = 0.01). These results suggest that addition of rituximab therapy to standard CHOP may lead to significant prolongation of event-free and overall survival in elderly patients with both high-risk and low-risk disease with no increase in toxicity. In an early analysis, CHOP-R appeared to be more effective than CHOP in bcl-2–positive, but not in bcl-2–negative, patients, suggesting that the benefit of addition of rituximab might overcome bcl-2 associated chemotherapy resistance.

A larger (N = 632) intergroup US study randomized a similar population of elderly patients to receive initial therapy with either CHOP or CHOP with rituximab. The rituximab was given on a differently described in follicular lymphoma. Responding patients then were randomized to receive either rituximab maintenance therapy (4 doses q 6 months × 2 years) or no maintenance. Preliminary results suggest a progression-free survival benefit for the group initially randomized to CHOP plus rituximab (P = 0.059); however, no overall survival benefit was apparent. One obvious difference between this trial and the GELA trial that might account for the failure to show a survival benefit is the fact that approximately 40% of patients on the CHOP induction arm received maintenance rituximab. While rituximab maintenance therapy benefited all patients, a subset analysis demonstrated that there was no benefit of maintenance rituximab for patients who received initial treatment with CHOP plus rituximab. The trial is difficult to therefore difficult to analyze for overall survival because of this demonstrable interaction between the induction and maintenance therapies. When a weighted analysis was performed to mathematically model two groups being treated with CHOP alone or CHOP plus rituximab, an overall survival benefit was apparent when induction therapy consisted of CHOP combined with rituximab. Thus this study shows a benefit of combining rituximab with CHOP chemotherapy as either induction therapy or maintenance therapy but not both. Ongoing analysis of this trial is being conducted to determine whether the benefit of rituximab is restricted to bcl-2–positive patients as suggested in the GELA trial. The results of these three pivotal studies are compared in Table 1.

However, until recently, there were no Phase III data addressing the value of rituximab in younger patients with DLCL-B. Preliminary results from the MiNIT Trial (Mab Thera International Trial), a trial evaluating the combination of CHOP and rituximab in patients younger than age 60, were presented this year. Eligibility criteria included CD20+ DLCL-B, 18–60 years, IPI 0 or 1, stages II–IV or stage I with bulky. Patients received 6 cycles of any one of several CHOP-like regimens followed by radiation therapy (30–40 Gray to bulky disease or E lesions). Rituximab was administered in the same schedule used by the previously described GELA trial. The preliminary report analyzed the first 326 patients. Median age of patients was 48 years, and 50% had bulky disease. Stage distribution was as follows: stage I, 19%, stage II, 57%; stage III, 12%, and stage IV, 12%. Thus, this trial mixed early and advanced stage DLCL-B patients. In fact, since almost 80% of patients had stage I and II disease and only 50% had bulky disease, one can estimate that almost 30% of the patients had low bulk, early-stage disease, i.e., they had an excellent prognosis. Patients receiving rituximab with chemotherapy had a significantly longer 2-year time to treatment failure (81% vs 58%) compared with those receiving chemotherapy alone. In addition, the 2-year OS significantly favored chemotherapy plus rituximab (95% vs 85%, P = 0.0026). This is the first randomized trial supporting the use of rituximab in younger patients, albeit a selected subset of younger patients; it did not deal with the poor-prognosis subset of younger patients.

### Table 1. Results of CHOP with or without rituximab in elderly diffuse large B cell lymphoma (DLCL-B).

<table>
<thead>
<tr>
<th>Study</th>
<th>Therapy</th>
<th>2-Year TTF</th>
<th>2-Year OS</th>
</tr>
</thead>
<tbody>
<tr>
<td>SWOG-8516</td>
<td>CHOP</td>
<td>49%</td>
<td>60%</td>
</tr>
<tr>
<td>GELA</td>
<td>CHOP</td>
<td>40%</td>
<td>60%</td>
</tr>
<tr>
<td></td>
<td>CHOP-Rituxim</td>
<td>58%</td>
<td>72%</td>
</tr>
<tr>
<td>ECOG-4494</td>
<td>CHOP</td>
<td>55%</td>
<td>65%</td>
</tr>
<tr>
<td></td>
<td>CHOP-Rituxim</td>
<td>65%</td>
<td>71%</td>
</tr>
</tbody>
</table>

Abbreviations: CHOP, cyclophosphamide, doxorubicin, vincristine, prednisone; TTF, time to treatment failure; OS, overall survival; SWOG, Southwest Oncology Group; GELA, Groupe d’Etude des Lymphomes de l’Adulte; ECOG, Eastern Cooperative Oncology Group.
Based largely upon the published results from GELA, CHOP with rituximab therapy (all therapy administered on day 1) has emerged to become the standard initial treatment for advanced stage DLCL-B in the US. Ongoing research will better define groups of patients with large cell lymphoma, if any, who do not benefit from the addition of monoclonal antibody therapy to chemotherapy.

A second concept aimed at improving the treatment of advanced stage DLCL-B is dose intensity. Two recent studies suggest possible benefit to dose intensification strategies. The SWOG conducted a pilot study evaluating dose-intensified CHOP (CHOP-DI: cyclophosphamide 1,600 mg/m², doxorubicin 65 mg/m², and vincristine 1.4 mg/m²) with filgrastim support, every 14 days for 6 planned courses. Treatment with CHOP-DI was safely administered in the cooperative group setting and resulted in survival 14% better compared with historical SWOG controls. Moreover, the NHL-B1 trial from Germany randomized patients to 6 cycles of CHOP-21, CHOP-14, CHOEP-21 (CHOP plus etoposide 100 mg/m² d1-d3), or CHOEP-14 in a 2 × 2 factorial study design. Patients in the 2-weekly regimen received granulocyte colony-stimulating factor (G-CSF) starting from day 4. Patients in this trial also received radiotherapy (36 Gy) to sites of initial bulky disease and extranodal disease. In patients older than age 60, 5-year overall survival rates were 40.6% for CHOP-21 and 53.3% for CHOP-14, suggesting a benefit to intensified therapy in this group of patients. There is no question that toxicity is also significantly increased in these dose-intensive regimens. A modification of this approach combines infusional therapy with dose escalation to maximal patient tolerance. While additional trials incorporating monoclonal antibody therapy into intensified programs are ongoing, the routine use of dose-intensive regimens is not recommended outside of a clinical trial setting.

A third novel treatment approach involves the use of autologous stem cell transplantation (ASCT) as part of the initial treatment regimen. Although it is beyond the scope of this paper to review all the literature involving ASCT, several studies have now concluded that not all patients with aggressive lymphoma benefit when stem-cell transplantation is incorporated into their initial treatment strategy compared with patients who are treated with conventional strategy of initial chemotherapy followed by stem-cell transplant at first relapse. Thus there seems to be no indication to add ASCT to the initial combination chemotherapy treatment for all patients with aggressive lymphoma. However, all trials that retrospectively have shown a benefit to ASCT in subset analyses have incorporated a standard course of induction therapy (rather than an abbreviated course) prior to consolidative transplantation and focused on patients with high clinical risk. An international consensus conference reached the conclusion that high-dose therapy and autotransplantation in patients with high-risk IPI scores seemed to provide benefit, and this is the subject of an ongoing intergroup randomized trial in the US. At the present time, we do not recommend routine use of ASCT as consolidative therapy for newly diagnosed large cell lymphoma outside a clinical trial.

Additionally, it is important to note that, regardless of the progress that has been made to date, a significant number of patients are still not cured of this disease. Multiple new agents are being considered for incorporation into the initial treatment regimens of patients with advanced stage DLCL-B. These include radioimmunotherapy, protein kinase C (PKC) beta inhibitors, epatuzumab, gallium nitrate, genasense, and anti-VEGF agents. Hopefully, these more targeted agents will not only increase the cure rate but also decrease toxicity.

Finally, we will briefly discuss the treatment of the relapsed patient with DLCL-B. The initial step in planning salvage chemotherapy is to determine the goal of treatment. Some patients who fail to achieve an initial remission or relapse from complete remission can be cured. This is less likely in elderly patients, those with extensive disease, and those with a poor performance status. In such patients less intensive, palliative systemic treatments, with single agent vincristine, cytarabine, alkylating agents, or anthracyclines might be better pursued. Responses to single agent rituximab occur approximately 30% of the time, and are generally of brief duration. Radiotherapy can also be used to alleviate the symptoms at a particular site of involvement in patients with relapsed diffuse large B-cell lymphoma. Most younger patients receive second-line combination chemotherapy regimens. These regimens usually incorporate drugs such as cisplatin, ifosfamide, etoposide, and cytarabine, often in combination with rituximab.

An international randomized trial referred to as the PARMA study defined the role of bone marrow transplant in relapsed DLCL. In this trial, 109 patients who had relapsed from complete remission and responded to two cycles of DHAP (dexamethasone, cytarabine, cisplatin) were randomly allocated to high-dose chemotherapy or continued treatment with DHAP. Bone marrow transplantation was associated with a superior failure-free survival (51% vs 12% at 5 years) and overall survival (53% vs 32% at 5 years). This trial enrolled only young patients at first relapse who remained chemo-sensitive. Thus salvage ABMT, as currently utilized will result in survival of approximately 50% of all
patients who actually receive transplants; however, only a minority of all patients meet all the strict selection criteria for ideal outcome following transplantation. For these patients, however, high-dose therapy and autologous bone marrow transplantation are the treatments of choice.

Allogeneic bone marrow transplantation has been used less frequently for patients with diffuse large B cell lymphoma. While occasional patients failing autologous transplantation can have prolonged survival with allogeneic transplantation, overall results have favored autologous transplantation, due to toxicity associated with allogeneic transplantation. Ongoing studies in high-risk patients are evaluating nonmyeloablative allogeneic transplantation.

Summary
Even with recent advances, many patients with advanced-stage DLCL-B are not cured with conventional therapy. Hence, each treating physician must recognize the inadequacy of current therapy and urge all eligible patients to participate in well-designed clinical trials. The best therapy remains to be defined, and therefore the best approach for the patient is an experimental approach designed to improve our ability to cure the disease. A list of active national protocols for treatment of DLCL-B will be presented during this educational session. If a patient is not eligible or does not wish to participate in a clinical trial, CHOP with rituximab is now the gold standard against which all new therapy must be compared.

II. LIMITED STAGE LYMPHOMA:
TREATMENT FOR AGGRESSIVE HISTOLOGIES

Thomas P. Miller, MD*

The term limited disease, as it refers to aggressive histologies of non-Hodgkin lymphoma (NHL), requires definition. “Limited disease,” also referred to as “early-stage” disease, “localized disease” and “low-stage” disease, has been given unique definitions in every published study to date. There is extreme heterogeneity within the group of patients described as having “limited-stage” lymphoma, making comparisons of outcome across this heterogeneous population difficult. Differences in outcome commonly ascribed to treatment strategies may be more accurately explained by patient selection. Understanding how these selection criteria influence the outcome of clinical trials is essential to choosing the optimal treatment for patients.

Modern therapy for limited-stage NHL was developed shortly after the doxorubicin-containing chemotherapy regimen CHOP was shown to be effective in advanced disease. At that time, patients with limited disease treated with radiotherapy (RT) alone routinely relapsed at distant sites (outside the initial areas of RT). CHOP chemotherapy was tested in patients with limited disease to determine if systemic therapy could prevent the growth potential of micrometastatic disease and thereby reduce the relapse rate and increase survival. Early pilot studies included patients with stage I or stage II disease and tested variable numbers of treatment cycles of CHOP, with and without consolidative RT.1,2 Subsequently, Connors and colleagues focused on the concept of a brief course of chemotherapy (3 cycles of CHOP) followed by involved-field RT [CHOP(3) plus RT] for patients with limited disease.3 Limited disease was carefully defined. Subsequently, the SWOG compared the concept of brief chemotherapy followed by RT [CHOP(3) plus RT] to 8 cycles of CHOP [CHOP(8)] in patients with localized aggressive histologies of NHL and found that CHOP(3) plus RT was superior to CHOP(8) through the first 5 years of follow-up.4 Localized disease was defined as stage I and non-bulky stage II disease. Bulky disease was defined as any mass having a maximum diameter greater than 10 cm or any mediastinal mass exceeding 1/3 the maximum trans-thoracic diameter.

Patients with bulky-stage II disease are known to have a prognosis similar to patients with advanced disease and were excluded from trials of limited-stage disease.5 Patients with bulky-stage II disease accrued to SWOG studies over the past 30 years have had a 5-year survival of 49%. Patients with stage III or IV disease have had a 5-year survival of 46%; therefore, bulky-stage II disease and any stage III or stage IV disease are considered advanced disease. Further, if patients with bulky-stage II disease are included in trials designed for patients with limited disease and are, consequently, treated with a short-course of chemotherapy plus RT (a treatment strategy designed for patients with lesser tumor burdens), they have an inferior survival compared to similar patients treated with aggressive chemotherapy designed for advanced disease. Reyes and colleagues for the GELA (Groupe d’Etude des Lymphomes de l’Adulte) have recently reported the results of a randomized trial comparing CHOP(3) plus RT to an aggressive combination-chemotherapy program consisting of doxorubicin, cyclophosphamide, vincristine, bleomycin and prednisone induction (ACVBP) at 2-week intervals, followed by high-dose methotrexate,
etoposide and cytarabine consolidation (LNH93-1). The ACVBP chemotherapy regimen was originally designed for advanced disease. In the subgroup of bulky-stage II disease, treatment with ACVBP was superior to CHOP(3) plus RT with 5-year survival estimates of 82% and 50%, respectively ($P = 0.03$). Although data on sites of relapse were not provided, one might surmise that most of the treatment failures on CHOP(3) plus RT were at distant sites of disease outside the RT field. One conclusion of this trial might be that the micrometastatic tumor burden in bulky-stage II disease is too high to be eradicated by 3 cycles of CHOP. Patients with bulky-stage II disease do not appear to have limited disease and seem better served receiving treatment designed for advanced disease.

By excluding patients with bulky-stage II disease one might presume that the remaining patients with limited disease comprise a homogeneous group with regard to prognosis and choice of optimal therapy. That is clearly not the case as 10-year survival can vary from 90% to 10% within subgroups of limited-stage patients. Predicting such variable outcome is easily accomplished using a minor modification of the IPI termed the stage-modified IPI (Table 2). The IPI is familiar to most clinicians and uses five risk factors to predict outcome including age, stage, serum LDH, performance status (PS), and the number of extranodal sites of disease. Likewise, age, stage, serum LDH and PS each predict significant outcome differences for patients with limited disease (the number of extranodal sites of disease is not applicable to this group of patients because the adverse risk is associated with two or more extranodal sites, a presentation which by definition indicates stage IV disease). However, stage must be redefined as stage I versus stage II in order to have utility. This modification of the IPI has proven utility as tested in two different series of limited-disease patients. These four risk factors (stage, age, serum LDH and PS) can be assessed for each patient and the prognosis determined based on the number of adverse risk factors (Table 3). Patients having no adverse risk factors (fully ambulatory patients less than 60 years with stage I disease and a normal serum LDH) have an outstanding prognosis when treated with initial doxorubicin-containing chemotherapy. Five- and 10-year survival estimates exceed 90% (Table 4 summarizes outcome by stage and risk group and proposes that this category be called “very limited disease”). These exceptional results can be achieved regardless of the treatment strategy chosen, CHOP(3) plus RT, CHOP(8), or ACVBP. Most clinicians would, therefore, select the initial treatment for patients with very limited disease based on toxicity profiles. In that regard CHOP(3) plus RT is clearly superior to ACVBP (25% hospitalization rate with each of 3 induction cycles of ACVBP), and probably superior to 8 cycles of CHOP (based on a limited but real increased risk of cardiomyopathy and neutropenic fevers associated with 8 cycles of CHOP). Further, patients with very limited disease have such a good prognosis that investigators will be hard-pressed to show a statistical improvement in outcome in randomized trials as there are so few events that the numbers of patients required to reach significance would be unrealistic. However, these patients, if included in trials, will have a positive influence on the outcome of any trial. Consequently, it is imperative to know the proportion of very

<table>
<thead>
<tr>
<th>Risk Factor</th>
<th>SWOG 5-Year OS</th>
<th>BCCA 5-Year OS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stage</td>
<td></td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>82%</td>
<td>–</td>
</tr>
<tr>
<td>II</td>
<td>68%</td>
<td>–</td>
</tr>
<tr>
<td>Age</td>
<td></td>
<td></td>
</tr>
<tr>
<td>≤ 60</td>
<td>85%</td>
<td>90%*</td>
</tr>
<tr>
<td>&gt; 60</td>
<td>70%</td>
<td>75%*</td>
</tr>
<tr>
<td>LDH</td>
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<td></td>
</tr>
<tr>
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<td>–</td>
</tr>
<tr>
<td>&gt; Normal</td>
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<td>0–1</td>
<td>79%</td>
<td>–</td>
</tr>
<tr>
<td>Status</td>
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<td></td>
</tr>
<tr>
<td>2</td>
<td>50%</td>
<td>–</td>
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<tr>
<td>0</td>
<td>94%</td>
<td>97%</td>
</tr>
<tr>
<td>1 or 2</td>
<td>–</td>
<td>77%</td>
</tr>
<tr>
<td>≥ 1</td>
<td>70%</td>
<td>–</td>
</tr>
<tr>
<td>3</td>
<td>50%</td>
<td>–</td>
</tr>
<tr>
<td>3 or 4</td>
<td>–</td>
<td>58%</td>
</tr>
</tbody>
</table>

* Progression-free survival

Abbreviations: LDH, lactate dehydrogenase; PS, performance status

### Table 2. Adverse risk factors for the International Prognostic Index (IPI) and stage-modified IPI compared (for use in limited stage lymphoma).

<table>
<thead>
<tr>
<th>Adverse Risk Factor</th>
<th>IPI</th>
<th>Stage-Modified IPI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stage</td>
<td>III, IV</td>
<td>Non-bulky II</td>
</tr>
<tr>
<td>Age</td>
<td>&gt; 60</td>
<td>&gt; 60</td>
</tr>
<tr>
<td>LDH</td>
<td>&gt; normal</td>
<td>&gt; normal</td>
</tr>
<tr>
<td>PS</td>
<td>≥ 2</td>
<td>≥ 2</td>
</tr>
<tr>
<td>“E” sites</td>
<td>≥ 2</td>
<td>Not applicable</td>
</tr>
</tbody>
</table>

Abbreviations: LDH, lactate dehydrogenase; PS, performance status

### Table 3. Five-year overall survival rates (OS) for patients treated with CHOP(3) plus radiotherapy in the Southwest Oncology Group (SWOG) or at the British Columbia Cancer Agency (BCCA), according to risk factors using a stage-modified International Prognostic Index (IPI) system.
limited disease patients in a study group to correctly interpret the results.

So who are the patients with limited disease and how should they be treated? Patients with limited disease (see Table 4) have stage I disease and at least one adverse risk factor or non-bulky stage II disease. The benchmark for comparison is CHOP(3) plus RT and the expected results depend on the number of adverse risk factors (see Table 4). Claims have been made that new standard therapies have been established. Investigators from the GELA have concluded that ACVBP (as discussed above) is superior to CHOP(3) plus RT. However, at the American Society of Hematology meetings in 2002, Reyes and colleagues demonstrated that the overall effect was largely (or wholly) due to the subgroup of patients having bulky-stage II disease. There was no perceptible difference between treatment arms (except for toxicity) in the subgroup of stage I and non-bulky stage II patients (limited disease). At those same meetings another GELA study was reported by Fillet and colleagues comparing 4 cycles of CHOP (CHOP(4)) to CHOP(4) plus 40 Gy involved-field RT in patients older than 60 years with no other adverse risk factors according to the age-adjusted IPI. Stage II and bulky-stage II patients were included. The results are intriguing. There was no difference in outcome as measured by event-free survival (EFS) or overall survival at 5 years (69% for CHOP alone versus 64% for CHOP plus RT for EFS). The involved-field RT seemed only to add to toxicity. This study implies that for patients with stage I and stage II (including bulky stage II), 4 cycles of chemotherapy is not only effective at eliminating microscopic sites of disease, but is effective at controlling even bulky sites of visible disease. That is remarkable because Horning and colleagues had recently reported that 8 cycles of CHOP plus RT had no effect on survival compared to 8 cycles of CHOP alone, but the addition of RT did significantly influence disease-free survival at 5 years. In this ECOG study only 215 of 352 eligible patients achieved a CR (61%) and were subsequently randomized to observation or RT alone. On the other hand, Fillet and colleagues had 69% of elderly patients free of disease at 5 years with 4 cycles of CHOP alone. The GELA trial piques interest, but many details need careful scrutiny before widespread application of such limited treatment.

CHOP(3) plus RT as initially proposed by investigators at the British Columbia Cancer Agency is still the benchmark for future comparisons, giving patients with very limited disease a 90% probability of cure at 10 years and giving patients with limited disease a 70% probability for cure at 10 years. CHOP(3) plus RT was not designed for use in advanced disease and is inadequate treatment for bulky-stage II disease patients. Minor variations in the schedule and simple substitutions of other drugs have not made a noticeable impact on outcome. Increased dose intensity has seemingly only added to toxicity, and increased treatment duration has not been directly compared to CHOP(3) plus RT. Future trials should probably confine patient selection to limited disease as defined in Table 4.

Future treatment strategies are likely to follow the paradigm of what works in advanced disease should also work in limited disease. In that regard it seems likely that investigators will combine a short course of doxorubicin-containing chemotherapy with targeted monoclonal antibodies. The results of at least one such pilot study have been submitted (Annual Meeting of the American Society of Hematology, 2004). Investigators for the SWOG have tested the efficacy of combining 4 infusions of rituximab with 3 cycles of CHOP followed by involved-field RT in patients with limited-stage diffuse aggressive B-cell histologies of NHL. Sixty-two patients in the experimental treatment group were compared to 68 patients from a historical group treated with CHOP(3) plus RT using identical selection criteria. PFS was compared at 2 years and was 94% for the rituximab-containing treatment arm and 85% for the historically treated CHOP(3) plus RT arm. Only 6 of the patients treated with rituximab-containing chemotherapy and 15 of the patients treated in the selected historical group have relapsed. The number of deaths

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**Table 4. The relationship between clinical stage, risk factors, and outcome for patients with limited disease.**

<table>
<thead>
<tr>
<th>Clinical Stage</th>
<th>Stage-Modified IPI Risk Factors*</th>
<th>Treatment</th>
<th>5-Year Median Survival</th>
<th>Limited Stage</th>
<th>Proposed Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>I, IE</td>
<td>0</td>
<td>CHOP(3) + RT</td>
<td>&gt; 90%</td>
<td>Yes</td>
<td>Very limited</td>
</tr>
<tr>
<td>I, IE, II, IIE (no bulky II)</td>
<td>≥ 1</td>
<td>CHOP(3) + RT</td>
<td>70%</td>
<td>Yes</td>
<td>Limited</td>
</tr>
<tr>
<td>Bulky II, Bulky IIE</td>
<td>≥ 1</td>
<td>CHOP(8)</td>
<td>50%</td>
<td>No</td>
<td>Advanced</td>
</tr>
</tbody>
</table>

* For stage-modified IPI risk factors see Table 2.

Abbreviations: IPI, International Prognosis Index; CHOP, cyclophosphamide, doxorubicin, vincristine, prednisone
in the first 2 years has also been reduced by 50% in the rituximab-containing treatment arm. These preliminary findings in a pilot study echo the results of previously published studies in advanced disease and provide a lead for future comparative trials.\textsuperscript{12,13}

Although the majority of patients with limited disease can be cured using a brief course of doxorubicin-containing chemotherapy followed by involved-field RT, there is room for improvement. Widely available studies combining targeted drugs with standard treatment offer patients a very real possibility of improved outcome.

### III. Emerging New Strategies in the Treatment of Mantle Cell Lymphoma

\textit{Owen A. O’Connor, MD, PhD*}

**Distinguishing Features of Mantle Cell Lymphoma**

The lymphomas represent one of the most heterogeneous group of malignancies known to medicine. Underneath the single rubric of lymphoma exist some of the fastest growing cancers known to science (Burkitt’s lymphoma, lymphoblastic lymphoma/leukemia), as well as some of the most indolent (small lymphocytic lymphoma, follicular lymphoma, and marginal zone lymphoma). This remarkable diversity of biology imposes significant challenges on pathologists who seek to understand the cell of origin and differentiate what are sometimes subtle differences between the related subtypes of disease. This process relies heavily on both standard immunohistochemistry approaches and the latest techniques in molecular cytogenetics to make the best diagnosis. Understanding these basic biological differences is beginning to afford us many new opportunities to tailor and develop specific treatments for these diseases that go well beyond simple CHOP-based chemotherapy, a paradigm that is now having significant ramifications in the treatment of even mantle cell lymphoma (MCL). This disease has moved rapidly from being an only recently described entity, to now being the subject of enormous basic science research. This new understanding has made MCL an inviting target for an incredible panoply of new and exciting drugs with promising activity. The goal of this section is to present these promising therapies in the context of the unique biology that defines the mantle cell lymphomas.

One of the major liabilities of the older lymphoma classification schemes is that they relied heavily on morphology and nodal architecture coupled in some cases to the clinical course, to classify the different subtypes of lymphoma. As a result, MCL was confused with other distinctly different types of lymphoma because of this resemblance in cellular and nodal features to other lymphomas. Historically, MCL had been referred to as “lymphocytic lymphoma of intermediate differentiation,” because some of the cells had well-rounded nuclei (i.e., small non-cleaved cell lymphomas), while others appeared to have indented or cleaved nuclei like those of small cleaved cell lymphoma. In 1982, Weisenburger\textsuperscript{1} and Palutke\textsuperscript{2} described a distinctive type of “follicular lymphoma” characterized by the proliferation of atypical small lymphoid cells in the wide mantles around benign germinal centers.\textsuperscript{3} Believing this represented the follicular counterpart of diffuse intermediate lymphocytic lymphoma, they coined the term \textit{mantle zone} lymphoma to describe this entity. In the early 1990s, it became evident there was a subset of diffuse-small cleaved cell lymphomas and small lymphocytic lymphomas that behaved very differently from other diseases with similar morphology. These patients often carried a worse prognosis with a distinctly different natural history. The development of new monoclonal antibodies used in immunohistochemistry, coupled with new techniques in cytogenetics, allowed pathologists to make important distinctions between these sub-types of diseases that transcended simple morphologic descriptors. As a result, in the early 1990s, Raffeld and Jaffe\textsuperscript{4} and Banks\textsuperscript{5} coined the term “mantle cell lymphoma” to describe a subset of small lymphocytic lymphomas that carried a unique translocation that results in the transpositioning of the BCL1 gene (11q13) to a site downstream of the immunoglobulin heavy-chain gene promoter (14q32). This balanced translocation leads to the upregulation of BCL1 [the so called t(11;14)(q13;q32)]. BCL1 (B-cell lymphoma 1), also known as PRAD1 because it has been identified in parathyroid adenoma, is a normal gene that codes for the protein cyclin D1. The juxtaposition of PRAD 1 downstream of the immunoglobulin heavy chain gene promoter leads to the constitutive overproduction of cyclin D1, theoretically resulting in a markedly dysregulated pattern of growth.

The MCL phenotype (Table 5) is characterized by the expression of pan B-antigens (CD20\textsuperscript{+}, CD22\textsuperscript{+}, though typically CD23 negative), with monotypic immunoglobulin (IgM\textsuperscript{+}D\textsuperscript{+}) and co-expression of the pan T antigen CD5. In addition, a λ-light chain restriction predominates, which is different from the typically κ-light chain restricted pattern commonly seen in other B

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cytochemical and cytogenetic features help to distinguish it from the marginal zone B cell lymphomas, with which these diseases are often confused. The nodal architecture in patients with MCL usually consists of atypical small lymphoid cells that generally display a nodular or diffuse pattern of growth, sometimes with elements of each. Focal areas of nodularity are evident in about 30% of cases on initial presentation. In nodular MCL, some of the nodules may consist of follicles with reactive germinal centers surrounded by broad expansile mantles of small lymphoid cell. As the disease progresses, there is a gradual invasion and obliteration of the interfollicular nodular areas by the neoplastic cells, leading to a diffuse pattern of growth, and a variant known as diffuse MCL. Morphologically, the lymph nodes consist of a monotonous population of atypical small to medium sized lymphocytes with irregular and indented nuclei. In about 20% of cases, the neoplastic cells of MCL are larger than those typically seen in the nodular variant, and the nuclei seem to have finely dispersed nuclear chromatin and prominent nucleoli. These cases have been referred to as the “blastic variant” or “anaplastic centrocytic” form of MCL. These blastic variants are often associated with a higher mitotic rate, a more aggressive clinical course, and an unfavorable natural history. Histologic progression from a nodular pattern to a diffuse pattern may be evident in repeat biopsy specimens obtained from the same patient, as may progression from the predominantly small lymphocytic forms of MCL to blastic cytology. Some reports suggest that histologic transformation to blastic cytology on re-biopsy can occur in up to 17% of cases, and may be as high as 70% at autopsy. Histologic transformation of MCL to DLCL-B, like that seen in patients with follicular lymphoma or small lymphocytic lymphoma, is considered a rare event.

The clinical separation of MCL from the other subtypes of diseases with which it is often lumped was first clarified in a landmark paper reported by Fisher, based upon an analysis of patient tissue obtained from three sequential randomized clinical trials conducted by the SWOG between 1972 and 1983. These data re-evaluated the tissue diagnosis from over 376 patients with Working Formulation diagnoses encompassing categories A through E (Table 6). They reported that 6 of 70 patients with small lymphocytic/diffuse well-differentiated lymphoma (category A) in fact had MCL, while 9 of 171 patients with follicular small cleaved/nodular poorly differentiated lymphoma (category B) had MCL, and 21 of 66 patients with diffuse small cleaved/diffuse lymphoma poorly differentiated lymphoma (category E) had MCL. They also demonstrated that the failure-free survival (FFS) and OS of patients with MCL was significantly worse than that of patients with Working Formulation diagnoses encompassing categories A through E (Table 6).

Table 6. Consensus pathology review of Southwest Oncology Group (SWOG) lymphoma cases (1972–1983).*

<table>
<thead>
<tr>
<th>IWF Diagnosis</th>
<th>Total (N)</th>
<th>MCL</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>70</td>
<td>6</td>
</tr>
<tr>
<td>B</td>
<td>171</td>
<td>9</td>
</tr>
<tr>
<td>C</td>
<td>40</td>
<td>0</td>
</tr>
<tr>
<td>D</td>
<td>29</td>
<td>0</td>
</tr>
<tr>
<td>E</td>
<td>66</td>
<td>21</td>
</tr>
<tr>
<td>Total Reviewed</td>
<td>376</td>
<td>36 (10%)</td>
</tr>
</tbody>
</table>

Abbreviations: IWF, International Working Formulation; MCL, mantle cell lymphoma

* Fisher et al. Blood 85:1075
Formulation (WF) diagnoses from categories A and E ($P = 0.0001$ and $0.0001$, respectively). In fact, the OS at 10 years for patients with MCL was only 8% compared to 35% for the WF categories A through E. In addition, separating the different histologic variants of MCL into the nodular, diffuse and blastic variants, the SWOG report demonstrated that the overall survival at 10 years for these MCL subtypes was 14%, 10% and 0% respectively. Clinically, MCL has an approximately 2:1 male to female predominance, with a median age of occurrence of about 58 years. It presents with generalized adenopathy in 71%–90% of cases, and with bone marrow positivity in 53%–90% of cases. Involvement of the gastrointestinal tract is thought to be nearly universal at the time of diagnosis. Today we know that the median survival of patients with MCL is only about 3 years, and that the median FFS from up-front conventional CHOP based treatment is only 15 to 18 months. These points are poignantly underscored in the very last sentence from the 1995 SWOG analysis,8 where it was emphasized that “...patients with MCL do not have an indolent lymphoma and are candidates for innovative therapy.” A recommendation that has been heeded over the past decade with some promising developments.

Despite our ability to distinguish MCL from the other forms of small lymphocytic lymphoma, it is clear that even this new entity of disease represents a spectrum of diseases, a spectrum of biology that is now being reorganized based on recent advancements in molecular analysis and gene expression array technologies. While a detailed analysis of the defining molecular events seen in MCL is beyond the scope of this presentation, it is clear that there are a number of molecular derangements beyond the t(11:14)(q13;q32) translocation that characterize the disease. Derangements in p27 and p53 for example appear to interact almost synergistically to yield a subtype of MCL with a particularly poor prognosis.9,10 In addition, Rosenwald et al11 reported on the use of gene expression profiling to establish a molecular basis for stratifying different subtypes of MCL. Such techniques are focused on developing a quantitative approach to elucidate the underlying pathogenesis and risk stratification of all patients. These investigators showed that the measurement of tumor cell proliferation determined by the identification of a set of “proliferation signature genes” was able to risk-stratify patients, based to a large extent on the differences in cyclin D1 mRNA abundance and the presence or absence of the cdk inhibitor INK4aARF.12 Collectively, these data have begun to reveal potential targets in these different subsets of MCL that may be appropriate substrates for novel drug discovery. At the least, this genetic based risk stratification could lead to the tailoring of innovative new treatments based on the particular subtype of MCL.

Conventional Treatment Strategies for MCL
Because MCL demonstrates some of the poorest long-term survival of all lymphoma subtypes, upfront treatment is typically indicated for most patients, although there appears to exist a small fraction of elderly patients whose MCL can assume a somewhat more indolent course, justifying in these otherwise frail and elderly patients a closely monitored “watch-and-wait” approach. In general, the upfront care of patients with MCL usually revolves around four basic questions: (1) Which combination chemotherapy regimen offers the patient the best chance for a durable remission of their disease? (2) Is an anthracycline based treatment regimen necessary? (3) What are the benefits of high dose chemotherapy in first remission? and (4) Is there a benefit to some consolidation based therapy in the form of either maintenance rituximab or a course of radioimmunotherapy. To date, while many studies are actively addressing these issues, the jury is largely still adjourned regarding the definitive answers to these questions.

Anthracycline-based regimens have become the cornerstone of upfront MCL therapy, despite conflicting evidence regarding the real benefits of the anthracycline component. Complete response rates from 13% to 51% for standard CHOP based chemotherapy have been published in a number of series.8,13-17 Only one randomized study to date comparing COP (n = 37) versus CHOP (n = 26) has been reported in MCL.13 This study could not demonstrate any statistically significant difference between the two arms with regard to complete (41% vs 58%) or partial remission (43% vs 31%), median OS (32 vs 37 months), relapse-free survival (10 vs 7 months), rates of relapse (73% vs 67%) or death. Teodorovic et al14 conducted a small randomized study through the European Organisation for Research and Treatment of Cancer (EORTC) from 1985 through 1992. In this study, 64 patients were randomized to one of four different regimens, including: (1) a cyclophosphamide, doxorubicin, teniposide, prednisone, vincristine, bleomycin regimen; (2) a modified ProMACE-MOPP regimen; (3) a CVP (cyclophosphamide, vincristine, prednisone) based regimen; or (4) CVP followed by one year of maintenance α-interferon. Although the overall (83% vs 52%) and complete remissions (60% vs 40%) rates were better with the more aggressive regimens in (1) and (2) above, the median survival was 45 months in all arms, with no benefit seen in the maintenance α-IFN arm.

The addition of rituximab to different chemotherapy
regimens appears to improve overall response, and in select cases, even overall survival, in patients with MCL. For example, integration of rituximab into a purine analog–based treatment in patients with MCL and FL was recently reported by Forstpointner and colleagues, who recently published the results of a large randomized Phase 3 study comparing the regimen of fludarabine, cyclophosphamide and mitoxantrone (FCM) alone versus FCM plus rituximab (R).\textsuperscript{18} Though the study included patients with both MCL and follicular lymphoma (FL), a subset analysis of patients with MCL was performed. The overall response rate (ORR) for all patients on study receiving the R-FCM regimen was 79\% (33\% CR; 45\% PR) compared to 58\% for FCM alone (13\% CR; 45\% PR; \textit{P} = 0.01), with an overall response rate (ORR) of 58\% versus 45\% in the subset of patients with MCL, respectively. Interestingly, in FL the PFS was significantly longer in the R-FCM arm (\textit{P} = 0.0139) while in the MCL patients a significantly longer OS was noted (\textit{P} = 0.0042), both in favor of the rituximab containing arm. Hence, in this well-balanced randomized study, it appears rituximab favorably benefited patients with MCL receiving FCM. Howard and colleagues also reported on the addition of rituximab to CHOP in patients with newly diagnosed MCL.\textsuperscript{19} Of 40 patients enrolled on study, 48\% achieved a CR or CRu (CR unconfirmed), while another 48\% experienced a PR, which is still in the range of responses seen with CHOP alone. The median PFS was about 16 months; again, not too dramatically different from CHOP alone. Interestingly, though, 9 of 25 patients who achieved molecular remissions (i.e., no evidence of PCR detectable BCL-1/IgH or clonal IgH products) of their disease in peripheral blood or bone marrow had a median PFS that was not statistically different from those patients who did not achieve a molecular remission (18.8 vs 16.5 months; \textit{P} = 0.51). Collectively, these data appear to suggest some role for the use of rituximab in patients with MCL, though the definitive value of rituximab remains to be clarified in future randomized studies.

Recently, the HyperCVAD regimen has gained a lot of attention in the upfront treatment of MCL, though there are only limited published data on the regimen for this disease.\textsuperscript{20-22} In one of the earlier reports, Romaguera et al\textsuperscript{23} reported on the use of the regimen in 25 patients older than 65 who received HyperCVAD followed by methotrexate/ cyctarbaine, where they reported an overall response rate of 92\% and a complete remission rate of 68\%. At a median follow up of 17 months, the FFS was 15 months. An update of these data by Romaguera et al\textsuperscript{23} with the addition of rituximab (375 mg/m\textsuperscript{2} preceding each of the first 6 cycles of therapy by 24 hours) reported on 92 patients with a median age of 61. The response rate was nearly 100\%, with 91\% complete remission, and 3 toxic deaths. For those patients younger than 65, the median FFS at 2 years was 80\%, compared to 50\% for patients older than 65 years of age. Khouri et al\textsuperscript{20,21} examined the merits of autologous stem cell transplantation following Hyper CVAD–MTX/Ara-C. They reported that at 3 years, the OS and EFS for previously untreated patients was 92\% and 72\%, respectively. For those patients who were previously treated, the results were much worse, with the OS and EFS being 25\% and 17\%, respectively. Based on very short follow-up of these studies, the MD Anderson Cancer Center (MDACC) group reported that the addition of rituximab to the HyperCVAD–MTX/Ara-C regimen produced results nearly identical to that seen with a consolidative ASCT.\textsuperscript{21,23} SWOG (Study S0213) is currently conducting a pilot trial of HyperCVAD followed by MTX/Ara-C with rituximab in patients with mantle cell lymphoma with the goal of accruing 50 patients.

Clearly, despite what would be considered very aggressive therapy, curative intent may not yet be possible for patients with MCL. For this reason, new strategies which seek to exploit some of the unique biology underlying MCL are warranted.

**Novel Agents in Development for MCL**

Over the last several years, there has been an explosion in the number of new drugs being tested in early phase clinical trials. Several of these agents have already shown promising activity in MCL, justifying more detailed advanced phase studies. While there are many agents that deserve attention here, I am able to discuss only a few select compounds for the purpose of this review.

**Proteasome inhibitors: bortezomib**

The proteasome is one component of a larger intracellular pathway responsible for the degradation of more than 90\% of all cytoplasmic protein, a pathway commonly referred to as the ubiquitin-proteasome pathway. The first step in the degradation of such proteins involves the highly regulated and coordinated cascade of enzymatic reactions that leads to the polyubiquitination of intracellular proteins targeted for degradation. The second major component of the pathway is the proteasome proper. The proteasome itself is composed of two components, one commonly referred to as the 20S proteasome, the second referred to as the 19S regulatory subunit. Collectively, they combine to form the 26S proteasome, which internally houses a number of different proteases responsible for degrading proteins into smaller irrelevant fragments. Many theories abound...
regarding the potential mechanism through which proteasome inhibition may lead to cell death or cell cycle arrest. One line of evidence has shown that inhibition of the proteasome leads to the accumulation of several cell cycle regulatory proteins, including the cyclins, and cyclin dependent kinase inhibitors p21 and p27. Another potentially important mechanism revolves around the potential direct induction of apoptosis through the modulation of anti- and proapoptotic proteins, namely bax and bik. To date, the most extensively studied mechanism revolves around the inhibition of NF-κB by inhibiting the degradation of its natural inhibitor, IκB. In normally quiescent cells, NF-κB exists in an inactivated form bound to IκB. In malignant cells, and in cells stimulated or stressed through exposure to various cytokines, cytotoxic drugs, viruses, oxidative triggers, or other mitogenic factors, IκB is phosphorylated by IκB kinase and then ubiquitinated, leading to its eventual degradation, and liberation of active free NF-κB.

On the Phase I study of bortezomib in patients with advanced hematologic malignancies, one heavily treated patient with MCL achieved a durable partial remission. Subsequently, at least three single agent Phase II studies with significant experiences in MCL have been reported. The first of these was reported by our group at Memorial Sloan-Kettering Cancer Center (MSKCC). Eligible patients had small lymphocytic lymphoma/chronic lymphocytic leukemia, FL, marginal zone lymphoma, or MCL. All patients were treated with 1.5 mg/m² on the typical day 1, 4, 8 and 11 schedule. Based on an update of the original data set, over 11 evaluable patients with MCL had been treated, with 6 of them achieving major partial remissions, and 5 achieving stable disease. Of the patients that achieved a PR, the shortest duration of that response has been 6 months, and the longest duration has been over 19 months. The latter patient has since gone on to receive 3 courses of therapy, now with over 27 months of disease-free survival, despite only a 6 month duration of response to CHOP followed by rituximab. A second study, recently reported by Goy et al from the MDACC, designed exactly the same as the MSKCC study except for a broader set of inclusion criteria for the types of NHL allowed, recently reported on 29 evaluable patients with MCL, 12 of whom achieved a major response to bortezomib (41% response rate). Interestingly, 6 of these patients had achieved a CR, 6 achieved a PR, and 6 attained stable disease. Though the followup in both studies is short, the median duration of response thus far in the MDACC study is about 6 months, in what would be considered a very heavily treated population of patients.

In addition to these studies, another smaller study being conducted by the National Cancer Institute of Canada (NCIC) in patients with MCL recently reported on their results. As of the most recent report on these data, 13 of 17 patients were evaluable for response. The NCIC study differed from the ones reported by MSKCC and MDACC in that they employed a lower dose of bortezomib (1.3 mg/m²). Partial responses were seen in 38% of patients, with one of these patients achieving a complete remission of all nodal and non-nodal based disease, though their bone marrow response was not fully evaluated (hence one CRu). Of those patients achieving a PR, the median time to response was about 2 cycles, with a duration of remission that ranged from 2.4 to 6.7 months (as of this report, and with an unclear period of follow-up, the median duration of response had not been attained). Presently, based on the encouraging results from these Phase II studies, a large industry sponsored multicenter study evaluating the time to progression and overall response rate for MCL patients receiving bortezomib is underway.

**Thalidomide-based treatments**

Thalidomide, originally developed as a sedative in the 1950s, was found to have marked teratogenic properties when administered to pregnant women, which eventually led to the widespread restriction of its use. Most recently, the drug has been resurrected based on its promising activity in both multiple myeloma and in combination with clarithromycin (Biaxin) and dexamethasone in Waldenström’s macroglobulinemia. Thalidomide is an immunomodulatory agent whose exact mechanism of action in both liquid and solid tumors remains to be clarified. In general though, it is known to have a number of pleiotropic effects on cells, ranging from angiogenic to anti-inflammatory effects, presumably by altering cytokine production. One of the properties that may explain its efficacy in multiple myeloma revolves around its potential to modify the stromal environment in the bone marrow, leading to a perturbation in the signaling pathways responsible for plasma cell survival. Damaj et al published a case report on two patients with very heavily pretreated MCL who achieved major durable partial remissions on thalidomide, which in both cases was maintained for over a year on the maintenance therapy. Recently, Kaufmann et al conducted a Phase II study of rituximab (R; 375 mg/m² IV weekly times four weekly doses) given concomitantly with thalidomide (T; 200 mg by mouth daily, with a incremental dose increases to 400 mg on day 15). The therapy was administered until progression or relapse. Remark-
ably, 13 patients (81%) experienced an objective response, with 5 complete responders (31%). The median PFS was 20.4 months, and the estimated 3-year survival was 75%. In those patients attaining a response, the PFS after R+T was longer than that achieved with the preceding chemotherapy. Overall the regimen was well tolerated, and the major adverse effects included 2 thromboembolic events and one grade 4 neutropenia.

Based on the anticancer properties of thalidomide, a significant research effort has been dedicated to the generation of new small molecules that are structurally similar though functionally distinct from thalidomide. These novel molecules, known as IMiDs, are orally bioavailable compounds with a safety profile that so far seems more favorable compared to thalidomide. One of these compounds, Revlimid, has now completed Phase I/II evaluation in multiple myeloma. These studies have demonstrated that Revlimid is not associated with the same spectrum of toxicities associated with thalidomide (sedation, thromboembolic events, constipation, neuropathy) and has produced significant reductions in paraproteins in most myeloma patients. Many studies are now underway to evaluate the activity of Revlimid in combination with rituximab in patients with lymphoma (Cancer and Leukemia Group B; CALGB). Clearly, the IMiDS are a class of molecules that warrant further attention in patients with MCL, especially in combination with other active agents like rituximab, traditional chemotherapy programs, and possibly bortezomib.

**Other promising agents**

**Flavopiridol:** One promising agent in development which may make perfect sense in targeting a disease characterized by gross dysregulation of a cyclin, is the use of flavopiridol. Flavopiridol is a large multicyclic compound originally derived from a plant indigenous to India known as *Dysoxylum bincectariferum*. Flavopiridol is a pan cyclin-dependent kinase (cdk) inhibitor that binds directly to the ATP-binding site at nanomolar concentration of most cyclin-dependent kinases.35 It is a potent inhibitor of the Cyclin D1, D2, D3-ckd4/6 complex, the Cyclin E/ckd2 complex, the Cylin B/ckd1 complex, and the Cyclin A/ckd1 complex. As such, it has been found to be a potent inducer of apoptosis when used with traditional chemotherapy drugs in a schedule dependent manner.36,37

Recently, Kouroukis et al38 published their experience with a Phase II study of flavopiridol in 30 patients with MCL, of whom 11 had no prior therapy. The response rate was about 11% (3 of 28 patients), with a median duration of response of only about 3 months (range 3 to 13 months). While these data suggested only modest activity, it is clear from the solid tumor experience to date that the merit of flavopiridol may not exist in its single agent use, but rather when combined rationally, and in a schedule dependent manner, with other chemotherapeutic agents with known single agent-activity.35,36

**Pixantrone (BBR 2778):** Anthracyclines are among the most active drugs in the treatment of aggressive large cell lymphoma. These compounds however, are often associated with cardiotoxicity, especially when used in high cumulative doses. In preclinical models pixantrone, a novel aza-anthracenedione, was shown to have greater cytotoxicity against P388 and L1210 leukemia cell lines compared to mitoxantrone and doxorubicin, and an in vivo murine model.39,40 These studies reported a more favorable therapeutic index with less cardiotoxicity. Recently, a very small Phase II experience in lymphoma was published on 33 patients with either DLCL-B (n = 24), MCL (n = 7), and 2 patients with transformed lymphoma.41 The overall response rate in this heavily treated population of patients was 27%. Of the 7 patients with MCL, one patient achieved a CR that was durable for over 15 months. Five of the 7 patients experienced a transient tumor reduction of more than 50%, which was unfortunately not durable. While pixantrone may have some suggestion of activity in MCL, its most promising venue for development may reside in the treatment of aggressive lymphomas and MCL. Within this context, it is becoming clear that understanding the levels of certain biochemical determinants, such as glutathione S-transferase π (GST-π) and topoisomerase IIα (topoIIα), for example, may help predict the patients and subtypes of disease more or less likely to respond.

Recent reports have shown that topoIIα correlated very strongly with OS in patients with MCL.42 Patients with low topoIIα expression (i.e., 0%–10%) had a median OS of 49 months, while those patients with levels > 10% had a median survival of only 17 months. A multivariate Cox regression analysis revealed the expression of topoIIα as the most important prognostic factor in MCL (P < 0.001), superior to the IPI. In addition, Bennaceur-Griscelli et al reported that the level of GST-π expression in MCL was significantly higher that that typically seen in DLCL-B or FL.43 Interestingly, GST-π is located at 11q13 and is co-amplified with the cyclin D1 gene in the same amplicon, which may account for some of the intrinsic chemotherapy resistance in MCL. Given that pixantrone produces its cytotoxic effects through the inhibition of topoisomerase II, by inducing double-strand breaks and intercalating into DNA, interpreting these results in the context of...
these molecular markers may afford new opportunities to tailor some of these new drugs to an individual’s particular subtype of MCL.

m-TOR inhibitors: The phosphoprotein kinase, TOR (target of rapamycin) is an important downstream component in the phosphoinositol-3 kinase (PI3K)/Akt pathway, playing an important role in the regulation of protein translation. Following mitogenic stimulation, activation of these kinases led to the proliferation of both T and B cells. In fact, increased PI3K activity in transgenic mice induces a T cell lymphoproliferative disorder that leads to the early development of T cell lymphoma. Constitutive activation of the Akt pathway has been described in many cell lines, including multiple myeloma. The importance of this pathway may be an extremely important determinant in explaining the sensitivity of to the rapamycin analogs against T and B cells.

Recently, Witzig et al reported on the results of a single-agent Phase II study of the rapamycin analog CCI-779 in previously treated patients with MCL. They treated the patients with 250 mg IV on days 1, 8, 15 and 22 every 4 weeks. Based on an interim evaluation of the results reported at ASH 2003, 18 eligible patients had been registered, with an overall response rate of 44%, including 1 CR (7 PR). Though the study is still ongoing, the scientific rationale coupled with the preclinical and now clinical data suggest that this target warrants further investigation.

Future Directions
Progress in medical oncology is characterized by a constant process of building upon the (sometimes small) successes of the past. The panoply of new targets, and now new agents to affect these targets, has begun to lay a platform of novel drug development that offers the potential to get to the heart of the underlying pathology that defines discrete lymphoproliferative malignancies. The major objective at hand is to begin using our existing preclinical models to explore the merits of combining these novel agents with each other, hopefully defining new ways to sensitize malignant cells to the effects of conventional chemotherapy agents. Integral to this will be the need to understand the importance of scheduling these agents as we expand the pool of drugs with which they will be given. Because it is still unclear what one strategy will emerge as the standard of upfront and second-line care of our patients with MCL, enrollment in clinical trials remains for now the new standard of care for MCL.

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