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.[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

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Distinguishing Features of Mantle Cell Lymphoma

The lymphomas represent one of the most heterogenous 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.

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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[1] and Palutke[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.[3] Believing this represented the follicular counterpart of diffuse intermediate lymphocytic lymphoma, they coined the term *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[4] and Banks[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 ($CD20^+$, $CD22^+$, though typically CD23 negative), with monotypic immunoglobulin (IgM^+D^+) 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 patten commonly seen in other B

Table 5. Distinguishing features of mantle cell lymphoma (MCL) compared to three non-Hodgkin's lymphoma (NHL) subtypes.

Pathology		Immunophenotype
Follicular center cell lymphoma	FCCL	$CD5^-$, $CD23^\pm$, $CD10^+$, $CD43^-$
Small cleaved cell	Grade 1	
Mixed small cleaved and large cell	Grade 2	
Large cell	Grade 3	
Small lymphocytic lymphoma with plasmacytoid differentiation	SLL-pl	$CD5^-$, $CD23^-$, $CD10^-$, $CD43^\pm$
Small lymphocytic lymphoma (CLL type)	SLL	$CD5^+$, $CD23^+$, $CD43^+$, $CD11a^+$, $FMC7^-$
Mantle cell lymphoma	MCL	$CD5^+$, $CD23^-$, $CD10^\pm$, $CD43^+$, $CyclinD1^+$, $FMC7^+$
Marginal zone lymphoma	MZL	$CD5^-$, $CD23^-$, $CD10^-$, $CD43^\pm$
Mucosa-associated lymphoid tissue	MALT	
Monocytoid B-cell lymphoma	MBCL	
Splenic	SMZL	
Splenic lymphoma with villous lymphocytes	SLVL	$CD5^-$, $CD11c^\pm$, $FMC7^+$, $CD22^+$, $CD24^+$
Hairy cell leukemia	HCL	$CD5^-$, $CD10^-$, $CD25^+$, $CD11c^+$, $CD103^+$, $B\text{-}ly\text{-}7^+$

cell lymphomas.[6] These 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.[1,7] 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.[3]

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,[8] 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

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

IWF Diagnosis	Total (N)	MCL
A	70	6
B	171	9
C	40	0
D	29	0
E	66	21
Total Reviewed	376	36 (10%)

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 al[11] 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 al[14] conducted a small randomized study through the European Organisation for Reseach 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).[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; $P = 0.01$), 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 ($P = 0.0139$) while in the MCL patients a significantly longer OS was noted ($P = 0.0042$), 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.[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; $P = 0.51$). 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.[20-22] In one of the earlier reports, Romaguera et al[22] reported on the use of the regimen in 25 patients older than 65 who received HyperCVAD followed by methotrexate/cytarabine, 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[23] with the addition of rituximab (375 mg/m^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[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.[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.[24] Another potentially important mechanism revolves around the potential direct induction of apoptosis through the modulation of anti- and proapoptotic proteins, namely *bax* and *bik*.[25,26] To date, the most extensively studied mechanism revolves around the inhibition of NF-κB.[27] Many investigators have demonstrated that inhibitors of the proteasome can block the activation of the transactivating transcription factor 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.[28] 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/m^2 on the typical day 1, 4, 8 and 11 schedule. Based on an update of the original data set,[29] 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,[30] 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.[31] 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/m^2). 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 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.[32] 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 pleotrophic effects on cells, ranging from anti-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[33] 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[34] conducted a Phase II study of rituximab (R; 375 mg/m^2 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 are 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. Flavoperidol is a large multicyclic compound originally derived from a plant indigenous to India known as *Dysoxylum binectariferum*. 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.[33] It is a potent inhibitor of the Cyclin D1, D2, D3-cdk4/6 complex, the Cyclin E/cdk2 complex, the Cylin B/cdk1 complex, and the Cyclin A/cdk 1complex. 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 al[38] 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.[44-46] 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.[47] 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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