New Strategies for the Treatment of Indolent Non-Hodgkin's Lymphomas

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ABSTRACT

Standard therapies for indolent non-Hodgkin's lymphomas (NHLs) have not been curative, and new approaches are needed. New chemotherapy drugs with unique mechanisms of action are in development. Of greatest interest are the effective monoclonal antibodies that have recently revolutionized the management of these patients. The most widely studied of these agents is rituximab, a chimeric anti-CD20 antibody. Rituximab induces responses in almost half of patients with relapsed, follicular/low-grade NHL, and achieves complete remissions in 6%. It is well tolerated in most patients. Combinations of rituximab with chemotherapy regimens such as CHOP (cyclophosphamide, vincristine, doxorubicin, prednisone) may alter the therapeutic paradigm for these patients. Radioimmunoconjugates achieve higher response rates than rituximab, but without a clear prolongation of survival, and are currently recommended for rituximab failures. Other approaches in clinical trials include antisense agents and lymphoma vaccines. These new agents should be used as a foundation on which to develop new strategies to increase the cure rates of patients with indolent NHLs.

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INTRODUCTION

Non-Hodgkin's lymphomas (NHLs) represent the fifth most common type of cancer and the sixth most common cause of cancer death in the United States. More than 56,000 cases are projected for the US in the year 2001.[1] Indolent lymphomas, which represent almost 30% of the NHLs, are a heterogeneous group of primarily B-cell disorders (Table 1).[2]

These diseases are at least initially responsive to therapy, with a relatively long survival, but they are not curable with conventional therapies. Thus, the management of indolent NHLs presents a challenge. Only about 15% of patients are diagnosed with limited disease (stages I and nonbulky II). For those patients, localized irradiation may result in long-term, disease-free survival; however, whether these patients are cured is not known.[3] The remaining 80–90% of patients present with advanced-stage disease. Studies evaluating patients who do not require immediate treatment have failed to demonstrate a survival advantage to early intervention, leading to a "watch and wait" approach.[4-7] Therefore, physicians confront issues of how best to treat these patients, and when to treat them, as well.

TABLE 1. WORLD HEALTH ORGANIZATION CLASSIFICATION OF MATURE MALIGNANT B-CELL NHL

Mature (peripheral) B-cell neoplasms
B-cell chronic lymphocytic leukemia/small B-lymphocytic leukemia
Lymphoplasmacytoid lymphoma
Splenic marginal zone B-cell lymphoma (± villous lymphocytes)
Hairy cell leukemia
Plasma cell leukemia
Plasma cell myeloma/plasmacytoma
Extranodal marginal zone B-cell lymphoma of MALT type
Nodal marginal zone B-cell lymphoma (± monocytoid B cells)
Follicular lymphoma
Mantle cell lymphoma
Diffuse large B-cell lymphoma
Burkitt lymphoma/Burkitt cell leukemia

NHL=non-Hodgkin's lymphoma; MALT=mucosa-associated lymphoid tissue.

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TALKING POINTS Physicians Pharmacy Formulary Cancer Nurses

New therapies are available for indolent non-Hodgkin's lymphoma (NHL). Despite higher response rates and greater expense, there is not yet clear evidence for prolongation of survival.

The paradigm for treatment of indolent NHL is rapidly changing as crucial data become available.

The new radioconjugates involve teamwork among physicians, nuclear medicine, and radiopharmacy, and preparation will differ with the various products.

Rituximab and other antibody therapies may be associated with mild to severe infusional reactions that require premedication, close monitoring, and adjustment of infusion rate.

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Once treatment is indicated by the presence of increasing lymphadenopathy or hepatosplenomegaly, disease-related symptoms, or progressive bone marrow compromise, there is no universally accepted standard treatment, and the preferred options are rapidly changing as new agents are introduced. Traditional chemotherapeutic options have ranged from single agents (eg, chlorambucil or cyclophosphamide), to combinations such as CVP (cyclophosphamide, vincristine, prednisone), to more intensive programs such as CHOP (cyclophosphamide, vincristine, doxorubicin, prednisone) (Table 2). However, none of these has led to a clear prolongation of survival compared with the others.[8,9] Following an initial response, the lymphoma becomes quiescent for a median of 2–4 years but eventually recurs, either with the same histology or as a more aggressive transformed subtype. With each subsequent treatment, response rates are lower and less durable and the median survival is about 6–8 years.[10]

Clearly, new approaches are needed. One area of active clinical research is to identify new chemotherapeutic agents with unique mechanisms of action (Table 3). Perhaps the most active of these are the nucleoside analogs of which fludarabine has been the most widely used. In initial trials conducted in patients with relapsed or refractory disease, responses were induced in 50% of patients, with 10–15% complete remissions (CRs).[11-13] Fludarabine was especially active in small lymphocytic lymphomas. When used as initial treatment, response rates in patients with follicular lymphoma were 65% with 37% CRs.[14] However, the median duration of response was only 18 months, with no evidence for cure. Fludarabine has been associated with CR and overall response rates higher than alkylating agent-based regimens, with more durable responses, but without a demonstrable prolongation of survival. Fludarabine is also associated with more myelosuppression, but with similar rates of infection.[15,16]

Combination regimens have been developed to improve on the activity of single agent fludarabine. One of the most widely used has been FND—fludarabine, mitoxantrone, and dexamethasone.[17,18] In a Phase II study of 51 patients with relapsed or refractory disease, the response rate was 94% (47% CR) with CRs lasting a median of almost 2 years.[18] The Southwest Oncology Group (SWOG) explored the combination of fludarabine and mitoxantrone in previously untreated patients.[19] Despite CR and overall response rates of 43% and 91%, respectively, progression-free survival was not clearly better than in the group's prior experience with other chemotherapy regimens. Whether these somewhat disappointing results were influenced by omission of dexamethasone cannot be determined with certainty.

Combinations of fludarabine and other chemotherapy agents have been evaluated. Flinn et al[20] treated 20 patients with follicular NHL using fludarabine (20 mg/m^2 days 1–5) and cyclophosphamide (600 mg/m^2 day 1) followed by granulocyte colony-stimulating factor. The overall response rate was 92% with 60% CR; however, significant myelosuppression and infections were common. In a Phase I study from the Eastern Cooperative Oncology Group (ECOG) of cyclophosphamide and fludarabine (cyclophosphamide 1,000 mg/m^2 day 1, fludarabine 20 mg /m^2 days 1–5 q28 days) in 27 previously untreated patients, there were 89% CRs with an overall response rate of 100%.[21] This combination was being compared with CVP in an ECOG trial, but it was closed to accrual because of an unexpected rate of life-threatening and fatal toxicity related to an excessive dose of cyclophosphamide.

Cladribine appears to have overall response rates similar to those seen with fludarabine, although with fewer CRs and more toxicity.[22-25] Data with pentostatin are limited and difficult to interpret.[26,27]

Other nucleoside analogs in development include nelarabine[28] and clofarabine (2-chloro-2'-fluoro-

TABLE 2. THERAPEUTIC OPTIONS FOR INDOLENT NON-HODGKIN'S LYMPHOMA

"Watch and wait"
Radiation therapy
Single alkylating agent
Combination alkylating agent-based chemotherapy ± anthracycline
Purine analogs or combinations
New chemotherapy agents
Stem cell transplant
Biologic approaches Monoclonal antibodies Interferon Antisense Vaccine

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TABLE 3. NEW CHEMOTHERAPY DRUGS FOR NON HODGKIN'S LYMPHOMA

Mechanism of Action	Representative Agents
• Cytotoxic	• Oxaliplatin
• Apoptosis induction	• Nucleosides, retinoids, arsenicals
• Protein kinase inhibition	• Bryostatin
• Cyclin inhibition	• Flavopiridol, UCN-01, rapamycin
• Farnesyl transferase inhibition	• R115777, BMS-214662
• Histone deacetylation	• Depsipeptide
• Antiangiogenesis	• Thalidomide, SU5416, SU6668
• Proteosome inhibition	• PS-341
• Antitubulin agents	• Epothilone

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arabinosyladenine),[29] both with activity in patients with chronic lymphocytic leukemia (CLL) and prolymphocytic leukemia; troxacitabine[30]; and bendamustine, a hybrid of a nitrogen mustard group and a purine-like benzimidazole, with demonstrated activity in CLL, NHL, and myeloma.[31]

OTHER NEW AGENTS

Arsenic

Interest in the development of arsenic trioxide as an anticancer agent was stimulated by its efficacy in acute promyelocytic leukemia.[32] Sensitivity to arsenic trioxide in vitro has also been demonstrated against a variety of other tumor types including myeloid leukemias, myeloma, lymphoid leukemia and lymphoma, and various solid tumors. Preclinical studies suggest that arsenic induces apoptosis of malignant lymphocytes at clinically achievable doses. Arsenic reduces survival and viability of both NHL and CLL cells with a significant decrease in bcl-2 expression and apoptosis of CLL cells.[33,34]

DNA Hypomethylation and Histone Deacetylation

The process of neoplastic progression is linked to imbalances in DNA methylation. Hypermethylation of gene-promoter regions is associated with repression of genes that regulate tumor growth and differentiation. Histone acetylation modulates higher order chromatin structure. Acetylation of lysine tails leads to loosening of DNA-histone contacts, which results in increased accessibility of transcription factors and increased gene expression. Coactivator-histone acetylase complexes promote gene expression whereas corepressor histone deacetylase complexes inhibit gene expression. There is potential synergy between DNA methylation inhibition and histone deacetylase inhibition in restoring gene expression silenced by hypermethylation.

Agents that inhibit histone deacetylase in vitro include sodium phenylbutyrate, depsipeptide, and hybrid polar compounds.[35,36] These agents induce terminal differentiation in vitro as well as cell-cycle arrest and reversion of the malignant phenotype of a variety of malignancies.

Depsipeptide (NSC 630176) is a bicyclic peptide originally isolated from *Chromobacterium violaceum*, strain 968, by Fujisawa Pharmaceutical Co., Ltd., that has been shown to be a histone deacetylase inhibitor.[37] Incubation of CLL cells with depsipeptide resulted in an alteration in apoptosis-associated proteins: an increase in BAX, no change in bcl-2, and a decrease in p27 expression.[38] Impressive activity has been noted in patients with T-cell lymphomas, particularly cutaneous T-cell NHL.

Two currently available hypomethylating agents include 5-azacytidine and 5-aza-2-deoxycytidine (decitabine). The combined administration of a demethylating agent and a histone deacetylase inhibitor synergize in reactivating genes that were silenced in cancer cells.[39] Such combinations represent a unique strategy that is currently being studied.

Antitubulin Agents

Antitubulin agents include dolastatin-10, a naturally occurring pentapeptide isolated from the marine mollusk *Dolabella auricularia*. Dolastatin-10 is one of the most potent in vitro cytotoxic anticancer compounds. The compound has antitumor activity against human leukemia and lymphoma cell lines and solid tumors.[40] Its cytotoxic effects may be related to modulation of apoptosis-associated proteins, such as bcl-2. Reversible myelosuppression was the dose-limiting toxicity in Phase I trials. Epothilone B analogs appear to be effective even in taxane-resistant cells and anecdotal responses have been reported in lymphomas.[41]

Cell-Cycle Inhibitors

Flavopiridol is a semisynthetic flavone derivative. It is derived from the plant alkaloid rohitukine, which is isolated from the leaves and stems of *Dysoxylum binectariferer um* used in India as an herbal medicine. Flavopiridol has in vitro activity against cycling as well as noncycling cells. Its antitumor activity may reflect the specific inhibition of cyclin-dependent kinases (CDKs), which regulate progression through the cell cycle. These include cyclin D1, which has been implicated in the pathogenesis of mantle cell lymphoma, as well as CDK1, CDK23, and CDK4. It induces growth arrest, cytotoxic cell death, and apoptotic changes in a variety of tumor types, including leukemias and lymphomas. It has also demonstrated sequence-specific synergy with cell cycle-active agents including fludarabine and cytarabine. Flavopiridol induces apoptosis of B-CLL and lymphoma cells. Little activity has been observed using a 72-hour infusion schedule, whereas responses have been reported with a 1-hour schedule in CLL and mantle cell lymphoma (MCL).[42-46] The Cancer and Leukemia Group B (CALGB) is carrying out a trial in refractory CLL patients with the shorter infusion schedule.

UCN-01 is an analog of staurosporine isolated from a *Streptomyces* species originally identified as a selective inhibitor of PKC.[47,48] Subsequently, it has been found to inhibit a number of serine/threonine kinases with arrest of cells in G1 and abrogation of the G2/M checkpoint. UCN-01 demonstrated cytotoxic effects in vitro and in vivo against a variety of murine and human malignant cell lines. UCN-01 can induce apoptosis in leukemia cells lacking functional p53 and resistant to apoptosis induced by DNA-damaging agents. Dose-limiting effects include hypoxia, self-limited hyperglycemia, lactic acidosis with hyperglycemia, nausea and vomiting, and transient elevation of liver transaminases. UCN-01 appears to potentiate the activity of fludarabine[49,50] and a combination of these two drugs in CLL is now being studied at the National Cancer Institute.

Antiangiogenesis Agents

An increasing body of evidence implicates angiogenesis in hematologic malignancies such as multiple myeloma, lymphoma, and CLL.[51,52] Angiogenesis factors such as basic fibroblast growth factor (bFGF) upregulate bcl-2, thereby delaying programmed cell death. New antiangiogenesis

agents available for clinical trials include thalidomide, SU5416, and SU6668.[53,54] After its approval for the treatment of leprosy and Kaposi's sarcoma and its recent efficacy in patients with refractorymultiple myeloma, thalidomide is being investigated in indolent lymphomas as a single agent and in combination with other chemotherapeutic agents.

SU5416 specifically inhibits vascular endothelial cell growth factor (VEGF) signaling through the Flk-1 receptor in endothelial cells. SU6668 inhibits the VEGF receptor Flk 1/KDR, the platelet-derived growth factor receptor-b, and the fibroblast growth factor receptor-1 tyrosine kinase. Both intravenous and oral formulations of SU6668 are entering Phase I trials.

Proteasome Inhibition

The proteasome is a large, multicentric protease complex with a pivotal role in cellular protein regulation. The proteasome degrades proteins that have been conjugated to ubiquitin, resulting in what is referred to as the ubiquitin-proteasome pathway. The ubiquitin-proteasome pathway plays a critical role in the degradation of intracellular proteins involved in cell-cycle control and tumor growth. Many tumor cells depend on rapid cell cycling, which requires expression and degradation of numerous regulatory proteins. Cells accumulate in the G2-M phase of the cell cycle with a decrease of cells in G1. The proteasome is also required for activation of NFkB, which plays a role in maintaining cell viability through the transcription of inhibitors of apoptosis. Since NFkB can induce drug resistance, this agent may make cells more chemosensitive.

PS-341, a dipeptidyl boronic acid, is a specific and selective inhibitor of the 26S proteasome.[55,56] It helps eliminate damaged or misfolded proteins and plays a regulatory role in multiple cellular pathways involving cell cycle, transcription factor activation, and cell trafficking. PS-341 may also induce apoptosis, and it has shown activity in cell types characterized by overexpression of bcl-2.

BIOLOGIC AGENTS

The introduction of new and active biologic agents has revolutionized our approach to the indolent NHLs. The first to be widely tested was interferon-α (IFN). IFN had modest single-agent activity in the treatment of patients with low-grade NHL,[57-59] leading to a series of Phase III trials. In a study from the Groupe d'Etude Lymphomes Folliculaire,[60] 242 evaluable patients with follicular NHL and a high tumor burden were treated with doxorubicin, cyclophosphamide, teniposide, and prednisone for 1 year ± concurrent IFN, which was continued out to 18 months. The advantage in response rate (85% versus 69%), event-free survival (34 months versus 19 months), and overall survival at 3 years (86% versus 69%) all favored the group that received IFN, and the results remained significant with additional follow-up.[61]

However, at least nine other randomized trials looked at IFN during induction,[62] as maintenance,[63-65] or both,[66-69] with inconsistent results. To resolve the controversy, Rohatiner et al[70] conducted a meta-analysis using the 1,671 newly diagnosed patients from the randomized IFN trials. In five studies, the chemotherapy was considered to be less intensive, defined as not including an anthracycline or anthracene agent.[65-69] In these studies, there was no evidence of any benefit from IFN. In contrast were the results of the analysis of the four trials of "more intensive" treatment.[61-64] Although there was still no improvement in the response rate when IFN was added to the chemotherapy agents, time to disease progression was prolonged, with a significant survival advantage in favor of the IFN arms; 14% at 5 years and 22% at 8 years, although this benefit was limited to responding patients.

However, following publication of this meta-analysis, Fisher et al[71] reported their study of 571 previously untreated patients with stage III or IV low-grade NHL who received a minimum of six courses of ProMACE-MOPP chemotherapy (procarbazine, methotrexate, doxorubicin, cyclophosphamide, etoposide-mechlorethamine, vincristine, procarbazine, prednisone) with involved-field radiation to convert partial responses with a negative bone marrow to CRs. Responding patients were randomized either to receive IFN for 2 years or to observation. There was no difference in progression-free survival or overall survival. It will be interesting to see the revised meta-analysis once the SWOG data are included.

A problem in comparing the various trials is that they differ in eligible histologies, patient risk factors, interferon type, dose, schedule, whether the agent was administered to all patients or only responders, and the percent of patients actually able to complete the full course of IFN therapy. Unfortunately, these studies were all initiated before publication of the International Prognostic Index,[72] which permits comparison of prognostic groups among studies.

The role of IFN in NHL remains controversial and its future will depend on whether it favorably interacts with the new chemotherapeutic, antiangiogenic and biologic therapies currently in development.

TABLE 4. MONOCLONAL ANTIBODIES AND RADIOIMMUNOCONJUGATES FOR NON-HODGKIN'S LYMPHOMA

Antibody	Antigen	Conjugate
Rituximab	CD20	None
CAMPATH-1H	CD52	None
Epratuzumab	CD22	None, I-131, Y-90
Hu-1D10 (apoltuzimab)	HLA-DR	None
Bevacizumab	VEG-F	None
Tositumomab	CD20	I-131
Ibritumomab	CD20	Y-90
Lym1	HLA-DR	I-131

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MONOCLONAL ANTIBODIES

Rituximab is one of a growing number of monoclonal antibodies, immunotoxins, and radioimmunoconjugates being evaluated for the treatment of B-cell malignancies (Table 4). Rituximab is a chimeric IgG1 anti-CD20 monoclonal antibody that was genetically engineered in 1990 by IDEC Pharmaceuticals. This antibody demonstrated excellent B-cell–depleting properties with no toxicity in preclinical animal models. In 1997, rituximab became the first monoclonal antibody approved by the US Food and Drug Administration.

Use in Treating Relapsed/Refractory Follicular/Low-Grade NHL

Rituximab binds with high affinity to CD20 present on the surface of most normal and malignant B cells (Tables 4 and 5). A number of mechanisms of action have been proposed including antibody-dependent cellular cytotoxicity, complement-mediated cytotoxicity, induction of apoptosis, recruitment of effector cells, and elaboration of cytokines.[81] Rituximab down-regulates bcl-2 expression in some NHL cell lines through an interleukin-10–dependent autocrine loop, which may reflect a decrease in STAT 3 (signal transducer and activator of transcription 3) activation.[82]

Initial Phase I studies of rituximab explored doses from 10–500 mg/m² as a single dose, then 125–375 mg/m² weekly for 4 weeks, but no maximum tolerated dose was identified even at the highest doses.[73,83] Treatment-related adverse events included fever, chills, nausea, vomiting, urticaria, orthostatic hypotension, and bronchospasm, and occurred mostly during infusion. Peripheral blood B cells were rapidly depleted but recovered within 6 months. Furthermore, 40% of the patients with low-grade histologies responded. The schedule of 375 mg/m² weekly for 4 weeks was selected for subsequent testing based on drug availability and because that schedule could be delivered on an outpatient basis.

In the first Phase II multicenter trial,[74] 37 patients who had relapsed after a median of two prior regimens were treated with 375 mg/m² for 4 consecutive weeks. There were three CRs and 14 partial remissions (response rate, 46%). Responses started as early as 1 month following therapy and reached a maximum at 4 months. The median time to progression for responders was 10.2 months, with a median duration of response of 8.2 months. The subsequent pivotal Phase II multicenter trial was conducted in 166 patients with relapsed and refractory follicular/low-grade NHL, mostly follicular grades I and II, but also 10 patients with follicular grade III NHL, and 33 with small lymphocytic lymphoma.[75] There were 45 patients resistant to their most recent chemotherapy, and 22 who were resistant to all prior treatments. The median number of prior regimens was three (range 1–10). The overall response rate was 48%, including 6% CRs; however, 76% of patients had at least a 20% reduction in tumor size, although in many of insufficient magnitude to qualify for a partial response. Responses lasted a median of about a year. Most adverse events occurred during infusion and included transient nausea, fever, bronchospasm, rash, hypotension, and pruritus.[75]

Unfortunately, all patients eventually relapse following rituximab and require additional therapy. The ability to re-treat would be desirable in order to avoid the myelosuppression associated with repeated courses of chemotherapy. In a report by Davis et al,[84] 60 patients who had previously responded to rituximab and relapsed at least 6 months later were re-treated with the antibody using the same dose and schedule. The overall response rate was 38%, including 10% CRs. Responses were still ongoing in 6 of 23 patients. No patient developed a human antichimeric antibody. The time to treatment failure of the initial response and the current response was 9.8 months versus 15.0+ months, and the time to progression was 12.4 months versus 16.7+ months, respectively.

TABLE 5. RESPONSE OF FOLLICULAR/ LOW-GRADE NON-HODGKIN'S LYMPHOMA TO RITUXIMAB

Study	Patients	Prior Therapy	CR (%)	RR (%)	Median TTP (months)
Maloney[73]*	15	Yes	0	33	6.4
Maloney[74]	37	Yes	9	41	10.2
McLaughlin[75]	166	Yes	6	48	13.0
Davis[76†]	31	Yes	3	39	8.1
Foran[77]	70	Yes	3	46	11.0
Piro[78‡]	37	Yes	14	43	7.7+
Colombat[79]	50	No	20	73	12
Hainsworth[80]	41	No	5	54	NR

*Phase I.
† Bulky disease.
‡ 8 weekly infusions.
CR=complete remission; RR=response rate; TTP=time to progression; NR=not reached.

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Increasing the Activity of Rituximab

A variety of approaches are currently being tried to increase rituximab's activity: use earlier in the course of the disease, increasing the dose or dose intensity, up-regulating CD20 expression on tumor cells,[85] enhancing effector cell function,[81] and developing combinations with chemotherapy drugs or other biologic agents.

A logical step was to determine if there was a higher level of activity for rituximab in previously untreated patients. Colombat et al[79] published the results of the French experience with 50 patients who had low-risk follicular NHL. The overall response rate was 73%, including 20% CRs and 6% unconfirmed CRs (CRu).[86] Of those patients whose tumors were PCR positive for the bcl-2 rearrangement prior to therapy, 57% became negative after treatment in the peripheral blood and 31% in the bone marrow. During the first year, 8% of the CR patients, 39% of the partial responders, and 50% of those with stable disease had progressed. The investigators found a close correlation between molecular and clinical responses: Bcl-2 negative patients had a 59% likelihood of a CR with 1/17 progressing by 12 months versus no CRs in patients persistently PCR+, of whom 62% relapsed by 12 months. Longer follow-up will be required to determine if a molecular response is associated with more durable responses and a potential for prolongation of survival.

Hainsworth et al studied 62 previously untreated patients[80]: 39% small lymphocytic lymphoma, 39% grade I, 27% grade II, 21% stage II, 32% stage III, and 44% stage IV (Table 2). Patients who responded to the initial course of therapy received repeat 4-week courses at 6-month intervals. After 6 weeks, the response rate was 54% with 5% CRs; this increased to 64% and 15% at 6 months prior to maintenance therapy. Following a second course, response status improved in two patients from stable disease to partial response and in one patient from partial to CR. Response rates were similar in the small lymphocytic lymphoma and follicular histologies. With a median follow-up of only 8 months, the actuarial 1-year progression-free survival was 77%. Although encouraging, the data with rituximab as initial treatment are too limited to support its routine use in this setting.

Increasing Dose Intensity

Several approaches to increasing dose or dose intensity have been explored including eight weekly infusions instead of four, thrice weekly administration, and intensified dosing.[76,87,88] None of these has clearly improved outcome. Consolidation courses should be studied further.[80]

Rituximab and Chemotherapy Combinations

Czuczman et al[89] published the first study of 40 patients with indolent NHL, 31 of whom were previously untreated, who received CHOP + rituximab. The response rate was 100% with 58% CRs. The median duration of response and time to progression had still not yet been reached at 45.8+ and 47.2+ months, respectively. Almost half the patients were still in remission from 36+ to 54.5+ months. Of interest was that seven of the eight tested patients became PCR-negative in both blood and bone marrow, and 6 of them remained in CR. To confirm these promising results, SWOG and CALGB are comparing CHOP with CHOP + rituximab, with CHOP followed by tositumomab, in patients with previously untreated follicular and low-grade NHL.

Fludarabine has also been combined with rituximab in indolent NHL[90] with an overall response rate of 93%, which included 80% CRs. Initially, toxicity was greater than anticipated, requiring a 40% dose reduction of fludarabine and discontinuation of trimethoprim sulfa. Bcl-2 was no longer detectable in the blood of all nine patients studied and in six of seven bone marrow samples.[91] The clinical significance of molecular remissions following rituximab therapy is unclear. Patients may become PCR-negative yet have persistent lymphadenopathy.[75,92]

Rituximab is being evaluated in combination with a variety of other combination regimens.[92] Davis et al[93] combined IFN with rituximab in 38 patients with relapsed/refractory follicular/low-grade NHL. The overall response rate was 45%, with 11% CRs, which is comparable to single-agent rituximab. The median time to progression was 25.2 months.

Combination trials with granulocyte colony-stimulating factor, granulocyte-macrophage colony-stimulating factor, interleukin-2, interleukin-12, and other cytokines are being conducted in an attempt to augment effector cell function.[94,95]

OTHER MONOCLONAL ANTIBODIES

Other monoclonal antibodies in development for indolent NHL include CAMPATH, a human IgG1 monoclonal antibody directed against the CD52 antigen present on most normal and malignant B- and T-cells. This antibody is effective in the treatment of cutaneous T-cell lymphoma,[96] T-prolymphocytic leukemia,[97] and CLL.[98] Unfortunately, its activity in indolent NHL has been limited, with 14% partial responses,[96] which may reflect its diminished activity in the setting of bulky lymphadenopathy.

Epratuzumab, a humanized monoclonal antibody directed against CD22 on B cells, has activity in indolent and aggressive NHL.[99,100] Hu1D10 is a humanized antibody directed against an epitope of HLA-DR and shows activity with excellent tolerability in early studies.[101]

RADIOIMMUNOCONJUGATES

The two most widely studied of the promising radioimmunoconjugates are iodine-131 tositumomab and yttrium 90-ibritumomab tiuxetan.

I-131 Tositumomab

I-131 tositumomab consists of an anti-CD20 monoclonal antibody (B1) bound to I-131. The isotope is primarily a γ-emitter, and dosimetry provides the correct infused dose (mCi) to deliver the desired absorbed therapeutic total body dose (cGy). The pilot study included 53 patients who had received a median of three prior regimens,[102,103] with an

overall response rate of 71% including 38% CRs, and a median progression-free survival of 12 months, with 20.3 months for the CRs. In a follow-up Phase II study,[104] 47 patients with relapsed or refractory low-grade or transformed lymphomas were treated with tositumomab with an overall response of 57% and CR of 32%. The multicenter pivotal trial[105] included 60 patients who had a median of four prior regimens. Responses were induced in 65% (81% in those with a low-grade [primarily follicular] histology), including 20% CRs. Response correlated with histology, tumor burden, prior radiotherapy, and number of prior therapies. Importantly, the response duration in 53% of patients was longer than with the last chemotherapy, which is unusual with chemotherapy for follicular NHL.[10] The median duration of response (6.5 months) was also significantly longer than to the last therapy (3.4 months), and the median duration of response for the CRs was not reached at a median follow-up of 47+ months versus 6.1 months for the patients' last therapy.

Data are limited on using a radioimmunoconjugate as initial therapy. Kaminski et al reported on 76 previously untreated patients with follicular lymphoma,[106] of whom 97% achieved a response with a CR in 76%. Further follow-up is needed to better assess the duration of response. These patients had less hematologic toxicity but a higher incidence of flulike symptoms and human antimurine antibody (65%).

Several approaches are under investigation to improve the activity of I-131 tositumomab, including repeated courses,[103] dose escalation requiring stem cell support,[107] and sequencing with chemotherapy.

Y-90 Ibritumomab Tiuxetan

Y-90-ibritumomab tiuxetan consists of the murine rituximab covalently bound to tiuxetan, which stably chelates Y-90. Y-90 differs from I-131 by being a β-emitter with a longer path length. Unlike I-131, Y-90 cannot be used for imaging; therefore, I-111 is used instead. In the Phase I/II studies, patients with a low-grade histology had an overall response of 82% with 26% CR and 56% partial response.[108]

To better clarify its role, a subsequent Phase III trial was conducted with 143 patients with relapsed CD20-positive NHLs (small lymphocytic, follicular, and transformed) who were randomized to receive either rituximab or ibritumomab tiuxetan.[109] The two treatment groups were similar with respect to known prognostic factors. Ibritumomab tiuxetan resulted in grade IV neutropenia in 32% of patients, although only 7% required hospitalization for infection. Grade IV thrombocytopenia occurred in 5%. Ibritumomab tiuxetan was more active with an overall response rate of 80% versus 56% for rituximab (P=.002); with 34% CR/CRu with the radioimmunoconjugate and 20% with the unconjugated antibody. Surprisingly, there was no difference in the median response duration (13.8+ and 15.5+ months).

Both ibritumomab tiuxetan and tositumomab appear to be highly effective in patients who have failed rituximab. Witzig et al[110] reported on 54 patients refractory to rituximab. The overall response rate was 54% with 15% CRs. The median duration of response of 7.7+ months compared favorably with the 6.5 months following the prior chemotherapy. The median time to progression had not been reached. Toxicities included a median absolute neutrophil count nadir of 700/mm^3 and platelets of 50,000/mm^3. Horning et al[111] treated 21 patients with I-131 tositumomab, most with a follicular grade I or II NHL. The overall response rate was 57% with 14% CRs, lasting a median of 16 months. The median time to progression for all patients and for responding patients was 6 months and 19 months, respectively. The median neutrophil nadir was 1,200/mm^3, and platelets were 90,000/mm^3. The high salvage rate of rituximab failures with a radioimmunoconjugate and their greater toxicity supports the use of a radioimmunoconjugate after rituximab therapy.

Further studies are need to better define the role of tositumomab and ibritumomab tiuxetan. Whether one or the other would better serve a particular subpopulation has yet to be determined. Longer follow-up is also needed to assess impact on overall survival and long-term effects including secondary malignancies and myelodysplastic syndromes. It is also not yet known whether chemotherapy can be delivered safely and effectively following radioimmunoconjugate therapy.

ANTISENSE AGENTS

Antisense bcl-2 Oligonucleotides

The bcl-2 protein results from the t(14;18) translocation commonly found in NHL and associated with inhibition of apoptosis. Attempts to down-regulate bcl-2 have led to development of antisense bcl-2 oligonucleotides. Waters et al[112] published their results of a Phase I trial of a bcl-2 antisense agent in 17 patients with relapsed and refractory indolent NHL. There was one CR and two minor responses. Reduction in bcl-2 protein correlated with response. The drug was relatively well tolerated with dose-limiting toxicities including thrombocytopenia, hypotension, fever, and asthenia. This agent will likely have its greatest effect by sensitizing tumor cells to other therapies.

VACCINES

Several vaccine strategies are being evaluated for lymphomas.[113,114] The most common are anti-idiotype vaccines, DNA vaccines, and those that use dendritic cells to augment the host antitumor response. Randomized studies are ongoing.

STEM CELL TRANSPLANTATION

The experience with stem cell transplantation (SCT) for low grade NHL is limited because of the older age of the population, the relatively long natural history of the disease, and the tendency for peripheral blood and bone marrow involvement; also, most patients have already received extensive prior therapy for their disease. The experience with autologous SCT (ASCT) for low-grade NHL has

generally been disappointing with little indication for a major impact on survival.[115-118] The most favorable results have been reported in highly selected cases. Serious complications of ASCT for NHL include up to a 20% actuarial risk of myelodysplastic syndrome and acute myelogenous leukemia.[119-122] The actuarial incidence at 10 years is almost 20%, and the outcome in these patients is extremely poor. This therapy should only be given in a clinical research trial. Ongoing studies are exploring the role of rituximab as a potential in vivo purging agent.

There are limited data available on the use of allogeneic bone marrow transplant in indolent NHL. In an analysis of 81 patients from the International Bone Marrow Transplant Registry,[123] transplanted at a median age of 41 years, 56% never achieved a CR; the projected survival at 3 years was 46%, with 43% disease-free survival. The median follow-up was only 23 months. However, transplant-related mortality was 44%. Chemosensitivity prior to transplant was the strongest predictor of outcome. Studies are ongoing to determine if a radioimmunoconjugate such as tositumomab or ibritumomab tiuxetan can replace total body irradiation in the bone marrow transplantation setting.[107]

When bone marrow transplant and ASCT are compared, the long-term survival figures are relatively comparable; ASCT is accompanied by a greater likelihood of dying from disease recurrence, whereas bone marrow transplant results in a high frequency of death from graft versus host disease, infection, and veno-occlusive disease. On the other hand, there may be some benefit from a moderate amount of graft versus host disease in the form of graft versus lymphoma effect.

Preliminary data are promising for submyeloablative SCT.[124,125] The goal is to achieve immunosuppression with myelosuppression, but without myeloablation. Patients with residual disease or mixed chimerism are then treated with donor leukocyte infusions. This approach appears to be most promising in patients with chemotherapy-responsive disease and low tumor burden CLL and indolent NHL.

CONCLUSIONS

We are clearly in a transition period in treating patients with indolent NHL. Even within histologic types there is marked clinical heterogeneity. New technologies such as DNA microarray analysis, initially used for diffuse large B-cell lymphomas,[126] are currently being evaluated for low-grade lymphomas. The result might be the ability to more accurately classify patients into prognostic subgroups and to direct more targeted therapies.

We are fortunate to now have several new, unique chemotherapy agents and, more importantly, active monoclonal antibodies and radioimmunoconjugates. Nevertheless, we are merely at the beginning of a new era, with numerous important issues to be addressed. The role of rituximab in relation to chemotherapy and the radioimmunoconjugates remains to be defined. A new paradigm—based on data rather than intuition—will have to be developed for the treatment of lymphomas. Physicians should be encouraged to enter their patients into clinical trials so that we can continue to make progress toward increasing the likelihood of cure in patients with indolent NHL. **OS**

REFERENCES

1. Greenlee RT, Hill-Harmon MB, Murray T, Thun M. Cancer statistics, 2001. *CA*. 2001;51:15-36.
2. Harris NL, Jaffe ES, Diebold J, et al. World Health Organization classification of neoplastic diseases of the hematopoietic and lymphoid tissues: report of the clinical advisory committee meeting—Airlie House, Virginia. *J Clin Oncol*. 1999;17:3835-3849.
3. Mac Manus M, Hoppe RT. Is radiotherapy curative for stage I and II low-grade follicular lymphoma? Results of a long-term follow-up study of patients treated at Stanford University. *J Clin Oncol*. 1996;14:1282-1290.
4. Portlock CS, Rosenberg SA, Glatstein E, Kaplan HS. Treatment of advanced non-Hodgkin's lymphomas with favorable histologies: preliminary results of a prospective trial. *Blood*. 1976;47:747-756.
5. Portlock CS, Rosenberg SA. No initial therapy for stages III and IV non-Hodgkin's lymphomas of favorable histologic types. *Ann Intern Med*. 1979;90:10-13.
6. Young RC, Longo DL, Glatstein E, Ihde DC, Jaffe ES, De Vita VT Jr. The treatment of indolent lymphomas: watchful waiting v aggressive combined modality treatment. *Semin Hematol*. 1988;25(suppl 2):11-16.
7. Brice P, Bastion Y, Lepage E, et al. Comparison of low-tumor burden follicular lymphomas between an initial no-treatment policy, prednimustine, or interferon alfa: a randomized study from the Group D'Etude des Lymphomes Folliculares. *J Clin Oncol*. 1997;15:1110-1117.
8. Ezdinli EZ, Anderson JR, Melvin F, Glick JH, Davis TE, O'Connell MJ. Moderate versus aggressive chemotherapy of nodular lymphocytic poorly differentiated lymphoma. *J Clin Oncol*. 1985;3:769-775.
9. Dana BW, Dahlberg S, Nathwani BN, et al. Long-term follow-up of patients with low-grade malignant lymphomas treated with doxorubicin-based chemotherapy or chemoimmunotherapy. *J Clin Oncol*. 1993;11:644-651.
10. Gallagher CJ, Gregory WM, Jones AE, et al. Follicular lymphoma: prognostic factors for response and survival. *J Clin Oncol*. 1986;4:1470-1480.
11. Redman JR, Cabanillas F, Velasquez WS, et al. Phase II trial of fludarabine phosphate in lymphoma: an effective new agent in low-grade lymphoma. *J Clin Oncol*. 1992;10:790-794.
12. Hochster HS, Kim K, Green MD, et al. Activity of fludarabine in previously treated non-Hodgkin's low-grade lymphoma: results of an Eastern Cooperative Oncology Group Study. *J Clin Oncol*. 1992;10:28-32.
13. Pigaditou A, Rohatiner AZS, Whelan JS, et al. Fludarabine in low-grade lymphoma. *Semin Oncol*. 1993;20(suppl 7):24-27.
14. Solal-Céligny P, Brice P, Brousse N, et al. Phase II trial of fludarabine monophosphate as first-line treatment in patients with advanced follicular lymphoma: a multicenter study by the Groupe d'Etude des Lymphomes de l'Adulte. *J Clin Oncol*. 1996;14:514-519.
15. Hagenbeek A, Eghbali H, Monfardini S, et al. Fludarabine versus conventional CVP chemotherapy in newly diagnosed patients with stages III and IV low grade malignant non-Hodgkin's lymphoma. Preliminary results from a prospective randomized phase III clinical trial in 381 patients. *Blood*. 1998;92(suppl 1):315a. Abstract 1294.
16. Klasa R, Meyer R, Shustik C, et al. Fludarabine versus CVP in previously treated patients with progressive low grade non-Hodgkin's lymphomas (lg-NHL). *Proc ASCO*. 1999;18:9a. Abstract 28.
17. McLaughlin P, Hagemeister FB, Swan F Jr, et al. Phase I study of the combination of fludarabine, mitoxantrone, and dexamethasone in low-grade lymphoma. *J Clin Oncol*. 1994;12:575-579.
18. McLaughlin P, Hagemeister FB, Romaguera JE, et al. Fludarabine, mitoxantrone, and dexamethasone: an effective new regimen for indolent lymphoma trial. *J Clin Oncol*. 1996;14:1262-1268.

19. Velasquez W, Lew D, Miller T, Fisher R. SWOG 95-01: a phase II trial of a combination of fludarabine and mitoxantrone (FN) in untreated advanced low grade lymphoma. An effective, well tolerated therapy. *Proc ASCO*. 1999;18:9a. Abstract 27.
20. Flinn IW, Byrd JC, Morrison C, et al. Fludarabine and cyclophosphamide with filgrastim support in patients with previously untreated indolent lymphoid malignancies. *Blood*. 2000;96:71-75.
21. Hochster HS, Oken MM, Winter JN, et al. Phase I study of fludarabine plus cyclophosphamide in patients with previously untreated low-grade lymphoma: results and long-term follow-up—a report from the Eastern Cooperative Oncology Group. *J Clin Oncol*. 2000;18:987-994.
22. Kay AC, Saven A, Carrera CJ, Piro LD. 2-Chlorodeoxyadenosine treatment of low-grade lymphomas. *J Clin Oncol*. 1992;10:371-377.
23. Hickish T, Serafinowski P, Cunningham D, et al. 2'-Chlorodeoxyadenosine: evaluation of a novel predominately lymphocyte selective agent in lymphoid malignancies. *Br J Cancer*. 1993;67:139-143.
24. Betticher DC, Fey MF, von Rohr A, et al. High incidence of infections after 2-chlorodeoxyadenosine (2-CDA) therapy in patients with malignant lymphomas and chronic and acute leukaemias. *Ann Oncol*. 1994;5:57-64.
25. Hoffman M, Tallman MS, Hakimian D, et al. 2-Chlorodeoxyadenosine is an active salvage therapy in advanced indolent non-Hodgkin's lymphoma. *J Clin Oncol*. 1994;12:788-792.
26. Cummings FJ, Kim K, Neiman RS, et al. Phase II trial of pentostatin in refractory lymphomas and cutaneous T-cell disease. *J Clin Oncol*. 1991;9:565-571.
27. Duggan DB, Anderson JR, Dillman R, Case D, Gottlieb AJ. 2'Deoxycoformycin (pentostatin) for refractory non-Hodgkin's lymphoma: a CALGB phase II study. *Med Ped Oncol*. 1990;18:203-206.
28. Gandhi V, Keating M, O'Brien S, Plunkett W. Compound GW506U78 in refractory hematologic malignancies: relationship between cellular pharmacokinetics and clinical response. *J Clin Oncol*. 1998;16:3607-3615.
29. Kozuch P, Ibrahim N, Khuri F, et al. Phase I clinical and pharmacological study of clofarabine. *Blood*. 1999;94(suppl 1):127a. Abstract 558.
30. Giles FJ, Cortes JE, Baker SD, et al. Troxacitabine, a novel dioxalone nucleoside analog, has activity in patients with advanced leukemia. *J Clin Oncol*. 2001;19:762-771.
31. Kath R, Blumenstengel K, Fricke HJ, Höffken K. Bendamustine monotherapy in advanced and refractory chronic lymphocytic leukemia. *J Cancer Res Clin Oncol*. 2001;127:48-54.
32. Soignet SL, Maslak P, Wang ZG, et al. Complete remission after treatment of acute promyelocytic leukemia with arsenic trioxide. *N Engl J Med*. 1998;339(19):1341-1348.
33. König A, Wrazel L, Warrell RP Jr, et al. Comparative activity of melarsoprol and arsenic trioxide in chronic B-cell leukemia lines. *Blood*. 1997;90:562-570.
34. Zhang W, Ohnishi K, Shigeno K, et al. The induction of apoptosis and cell cycle arrest by arsenic trioxide in lymphoid neoplasms. *Leukemia*. 1998;12(9):1383-1391.
35. Richon VM, Emiliani S, Verdin E, et al. A class of hybrid polar inducers of transformed cell differentiation inhibits histone deacetylases. *Proc Natl Acad Sci U S A*. 1998;95(6):3003-3007.
36. Saito A, Yamashita T, Mariko Y, et al. A synthetic inhibitor of histone deacetylase, MS-27-275, with marked in vivo antitumor activity against human tumors. *Proc Natl Acad Sci U S A*. 1999;96(8):4592-4597.
37. Nakajima H, Kim YB, Terano H, Yoshida M, Horinouchi S. FR901228, a potent antitumor antibiotic, is a novel histone deacetylase inhibitor. *Exp Cell Res*. 1998;241(1):126-133.
38. Byrd JC, Shinn C, Ravi R, et al. Depsipeptide (FR901228): a novel therapeutic agent with selective, in vitro activity against human B-cell chronic lymphocytic leukemia cells. *Blood*. 1999;94:1401-1408.
39. Cameron EE, Bachman KE, Myohanen S, Herman JG, Baylin SB. Synergy of demethylation and histone deacetylase inhibition in the re-expression of genes silenced in cancer. *Nat Genet*. 1999;21(1):103-107.
40. Beckwith M, Urba WJ, Longo DL. Growth inhibition of human lymphoma cell lines by the marine products, dolastatins 10 and 15. *J Natl Cancer Inst*. 1993;85:483-488.
41. Lee FY, Borzilleri R, Fairchild CR, et al. BMS-247550: a novel epothilone analog with a mode of action similar to paclitaxel but possessing superior antitumor activity. *Clin Cancer Res*. 2001;7:1429-1437.
42. Achenbach TV, Müller R, Slater EP. BCL-2 independence of flavopiridol-induced apoptosis. *J Biol Chem*. 2000;275:32089-32097.
43. Arguello F, Alexander M, Sterry JA, et al. Flavopiridol induces apoptosis of normal lymphoid cells, causes immunosuppression, and has potent antitumor activity in vivo against human leukemia and lymphoma xenografts. *Blood*. 1998;91:2482-2490.
44. Bible KC, Kaufmann SH. Cytotoxic synergy between flavopiridol (NSC 649890, L86-8275) and various antineoplastic agents: the importance of sequence of administration. *Cancer Res*. 1997;57:3375-3380.
45. Byrd JC, Shinn C, Waselenko JK, et al. Flavopiridol induces apoptosis in chronic lymphocytic leukemia cells via activation of caspase-3 without evidence of bcl-2 modulation or dependence on functional p53. *Blood*. 1998;92:3804-3816.
46. König A, Schwartz G, Mohammed RM, Al-Katib A, Gabrilove J. The novel cyclin-dependent kinase inhibitor flavopiridol downregulates bcl-2 and induces growth arrest and apoptosis in chronic B-cell leukemia lines. *Blood*. 1997;90:4307-4312.
47. Wang Q, Fan S, Eastman A, Worland PJ, Sausville EA, O'Connor PM. UCN-01: a potent abrogator of G2 checkpoint function in cancer cells with disrupted p53. *J Natl Cancer Inst*. 1996;17:956-965.
48. Waselenko JK, Przygodzki R, Grever MR, Shinn CA, Sausville E, Byrd JC. Flavopiridol, UCN-01, FR901228 and gemcitabine have significant in vitro activity in de novo B-cell prolymphocytic leukemia with aberrant p53 function. *Blood*. 1997;90(suppl 1):532a. Abstract 2369.
49. Monks A, Harris E, Connelly J, et al. Enhancement of fludarabine and gemcitabine toxicity by UCN-01 in a variety of tumor cell lines. *Proc Am Assoc Cancer Res*. 1999;40. Abstract 45.
50. Harvey S, Decker R, Dai Y, et al. Interactions between 2-fluoroadenine 9-β-D-arabinofuranoside and the kinase inhibitor UCN-01 in human leukemia and lymphoma cells. *Clin Cancer Res*. 2001;7:320-330.
51. Salven P, Teerenhovi L, Joensuu H. A high pretreatment serum vascular endothelial growth factor concentration is associated with poor outcome in non-Hodgkin's lymphoma. *Blood*. 1997;90:3167-3172.
52. Molica S, Vitelli G, Levato D, Gandolfo GM, Liso V. Increased serum levels of vascular endothelial growth factor predict risk of progression in early B-cell chronic lymphocytic leukaemia. *Br J Haematol*. 1999;107:605-610.
53. Singhal S, Mehta J, Desikan R, et al. Antitumor activity of thalidomide in refractory multiple myeloma. *N Engl J Med*. 1999;341:1565-1571.
54. Smolich BD, Yuen HA, West KA, Giles FJ, Albitar M, Cherrington JM. The anti-angiogenic protein kinase inhibitors SU5416 and SU6668 inhibit the SCF receptor (c-kit) in a human myeloid leukemia cell line and in human myeloid leukemia blasts. *Blood*. 2001;97:1413-1421.
55. Adams J, Stein R. Novel inhibitors of the proteasome and their therapeutic use in inflammation. *Ann Rep Med Chem*. 1996;31:279-288.
56. Adams J, Palombella VJ, Sausville EA, et al. Proteasome inhibitors: a novel class of potent and effective antitumor agents. *Cancer Res*. 1999;59:2615-2622.
57. Merigan TC, Sikora K, Breeden JH, Levy R, Rosenberg SA. Preliminary observations on the effect of human leukocyte interferon in non-Hodgkin's lymphoma. *N Engl J Med*. 1978;299:1449-1553.
58. Foon KA, Sherwin SA, Abrams PG, et al. Treatment of advanced non-Hodgkin's lymphoma with recombinant leukocyte A interferon. *N Engl J Med*. 1984;311:1148-1152.
59. O'Connell MJ, Colgan JP, Oken MM, Ritts RE Jr, Kay NE, Itri LM. Clinical trial of recombinant leukocyte A interferon as initial therapy for favorable histology non-Hodgkin's lymphomas and chronic lymphocytic leukemia. An Eastern Cooperative Oncology Group pilot study. *J Clin Oncol*. 1986;4:128-136.

60. Solal-Céligny P, Lepage E, Brousse N, et al. Recombinant interferon alfa-2b combined with a regimen containing doxorubicin in patients with advanced follicular lymphoma. *N Engl J Med.* 1993;329:1608-1614.
61. Solal-Céligny P, Lepage E, Brousse N, et al. A doxorubicin-containing regimen with or without interferon alpha-2b for advanced follicular lymphomas: final analysis of survival and toxicity in the GELF 86 trial. *J Clin Oncol.* 1998;16:2332-2338.
62. Smalley RV, Andersen JW, Hawkins MJ, et al. Interferon alfa combined with cytotoxic chemotherapy for patients with non-Hodgkin's lymphoma. *N Engl J Med.* 1992;327:1336-1341.
63. Avilés A, Duque G, Talavera A, Guzmán R. Interferon alpha 2b as maintenance therapy in low grade malignant lymphoma improves duration of remission and survival. *Leuk Lymphoma.* 1996;20:495-499.
64. Unterhalt M, Hermann R, Koch P, et al. Long term interferon alpha maintenance prolongs remission duration in advanced low grade lymphomas and is related to the efficacy of initial cytoreductive therapy. *Blood.* 1996;88(suppl 1):453a. Abstract 1801.
65. Hagenbeek A, Carde P, Meerwaldt JH, et al. Maintenance of remission with human recombinant interferon alfa-2a in patients with stages III and IV low-grade malignant non-Hodgkin's lymphoma. *J Clin Oncol.* 1998;16:41-47.
66. Chisesi T, Congiu M, Contu A, et al. Randomized study of chlorambucil (CB) compared to interferon (alfa-2b) in low-grade non-Hodgkin's lymphoma: an interim report of a randomized study. *Eur J Cancer.* 1991;27(suppl 4):S31-33.
67. Price CGA, Rohatiner AZS, Steward W, et al. Interferon alfa-2b in addition to chlorambucil in the treatment of follicular lymphoma: preliminary results of a randomized trial in progress. *Eur J Cancer.* 1991;27(suppl 4):S34-S36.
68. Peterson BA, Petroni G, Oken MM, Ozer H. Cyclophosphamide versus cyclophosphamide plus interferon alfa-2b in follicular low grade lymphomas: a preliminary report of an intergroup trial (CALGB 8691 and EST 7486). *Proc ASCO.* 1993;12:366. Abstract 1240.
69. Arranz R, García-Alfonso P, Sobrino P, et al. Role of interferon alfa-2b in the induction and maintenance treatment of low-grade non-Hodgkin's lymphoma: results from a prospective, multicenter trial with double randomization. *J Clin Oncol.* 1998;16:1538-1546.
70. Rohatiner AZS, Gregory W, Peterson B, et al. A meta-analysis (MA) of randomised trials evaluating the role of interferon (IFN) as treatment for follicular lymphoma (FL). *Proc ASCO.* 1998;17:4a. Abstract 11.
71. Fisher RI, Dana BW, LeBlanc M, et al. Alpha-interferon consolidation following intensive chemotherapy does not prolong the failure-free survival of patients with low grade non-Hodgkin's lymphoma: results of SWOG-8809, a randomized phase III study. *J Clin Oncol.* 2000;18:2101-2016.
72. Shipp MA, Harrington DP, Anderson JR, et al. Development of a predictive model for aggressive lymphoma: the International Non-Hodgkin's Lymphoma Prognostic Factors Project. *N Engl J Med.* 1993;329:987-994.
73. Maloney DG, Grillo-López AJ, Bodkin DJ, et al. IDEC-C2B8: results of a phase I multiple-dose trial in patients with relapsed non-Hodgkin's lymphoma. *J Clin Oncol.* 1997;15:3266-3274.
74. Maloney DG, Grillo-López AJ, White CA, et al. IDEC-C2B8 (rituximab) anti-CD20 monoclonal antibody therapy in patients with relapsed low-grade non-Hodgkin's lymphoma. *Blood.* 1997;90:2188-2195.
75. McLaughlin P, Grillo-López AJ, Link BK, et al. Rituximab chimeric anti-CD20 monoclonal antibody therapy of relapsed indolent lymphoma: half of patients respond to a four-dose treatment program. *J Clin Oncol.* 1998;16:2825-2833.
76. Piro LD, White CA, Grillo-Lopez AJ, et al. Extended rituximab (anti-CD20 monoclonal antibody) therapy for relapsed or refractory low-grade or follicular non-Hodgkin's lymphoma. *Ann Oncol.* 1999;10:655-661.
77. Foran JM, Gupta RK, Cunningham D, et al. A UK multicentre phase II study of rituximab (chimaeric anti-CD20 monoclonal antibody) in patients with follicular lymphoma, with PCR monitoring of molecular response. *Br J Haematol.* 2000;109:81-88.
78. Davis TA, White CA, Grillo-Lopez AJ, et al. Single-agent monoclonal antibody efficacy in bulky non-Hodgkin's lymphoma: results of a phase II trial of rituximab. *J Clin Oncol.* 1999;17:1851-1857.
79. Colombat P, Salles G, Brousse N, et al. Rituximab (anti-CD20 monoclonal antibody) as single first-line therapy for patents with follicular lymphoma with a low tumor burden: clinical and molecular evaluation. *Blood.* 2001;97:101-106.
80. Hainsworth JD, Burris HA III, Morrissey LH, Litchy S, Scullin DC Jr, Bearden JD III. Rituximab monoclonal antibody as initial systemic therapy for patients with low-grade non-Hodgkin's lymphoma. *Blood.* 2000;95:3052-3056.
81. Ansell SM, Tschumper RC, Witzig TE, Kurtin PJ, Jelinek DF. Chemokine upregulation in response to rituximab in B-cell non-Hodgkin's lymphoma. *Blood.* 1999;94(suppl 1):86a. Abstract 377.
82. Alas S, Bonavida B. Rituximab inactivates signal transducer and activator of transcription 3 (STAT3) activity in B-non-Hodgkin's lymphoma through inhibition of the interleukin 10 autocrine/paracrine loop and results in down-regulation of bcl-2 and sensitization to cytotoxic drugs. *Cancer Res.* 2001;61:5137-5144.
83. Maloney DG, Liles TM, Czerwinski DK, et al. Phase I clinical trial using escalating single-dose infusion of chimeric anti-CD20 monoclonal antibody (IDEC-C2B8) in patients with recurrent B-cell lymphoma. *Blood.* 1994;84:2457-2466.
84. Davis TA, Grillo-López AJ, White CA, et al. Final report on the safety and efficacy of retreatment with rituximab for patients with non-Hodgkin's lymphoma. *Blood.* 1999;94(suppl 1):88a. Abstract 385.
85. Venugopal P, Sivaraman S, Huang X, et al. Upregulation of CD20 expression in chronic lymphocytic leukemia (CLL) cells by in vitro exposure to cytokines. *Blood.* 1998;92(suppl 1):247a. Abstract 1009.
86. Cheson BD, Horning SJ, Coiffier B, et al. Report of an International Workshop to standardize response criteria for non-Hodgkin's lymphomas. *J Clin Oncol.* 1999;17:1244-1253.
87. Byrd JC, Murphy T, Howard RS, et al. Rituximab using a thrice weekly dosing schedule in B-cell chronic lymphocytic leukemia and small lymphocytic lymphoma demonstrates clinical activity and acceptable toxicity. *J Clin Oncol.* 2001;19:2153-2164.
88. O'Brien SM, Kantarjian H, Thomas DA, et al. Rituximab dose-escalation trial in chronic lymphocytic leukemia. *J Clin Oncol.* 2001;19:2165-2170.
89. Czuczman MS, Grillo-López AJ, White CA, et al. Treatment of patients with low-grade B-cell lymphoma with the combination of chimeric anti-CD20 monoclonal antibody and CHOP chemotherapy. *J Clin Oncol.* 1999;17:268-276.
90. Czuczman MS, Jonas C, Stephan M, et al. Pilot study of Rituxan in combination with fludarabine chemotherapy in patients with low-grade or follicular non-Hodgkin's lymphoma. *Proc ASCO.* 1999;18:17a. Abstract 61.
91. Czuczman MS, Fallon A, Scarpace A, et al. Phase II study of rituximab in combination with fludarabine in patients (pts) with low-grade or follicular B-cell lymphoma. *Blood.* 2000;96(suppl 1):729a. Abstract 3154.
92. Cabanillas F, McLaughlin P, Hagemeister F, et al. Molecular responses with fludarabine/mitoxantrone/dexamethasone plus rituximab chemoimmunotherapy for stage IV indolent follicular non-Hodgkin's lymphoma. *Blood.* 2000;96(suppl 1). Abstract 1429.
93. Davis TA, Maloney DG, Grillo-López AJ, et al. Combination immunotherapy of relapsed or refractory low-grade or follicular non-Hodgkin's lymphoma with rituximab and interferon-alpha-2a. *Clin Cancer Res.* 2000;6:2644-2652.
94. van der Kolk LE, Grillo-López AJ, Baars JW, van Oers MHJ. Treatment of relapsed B-cell non Hodgkin's lymphoma with a combination of chimeric anti-CD20 monoclonal antibodies (rituximab) and G-CSF: final report on safety and efficacy. *Blood.* 2000;96(suppl 1):732a. Abstract 3167.
95. Ansell SM, Witzig TE, Kurtin PJ, et al. Phase I study of interleukin-12 in combination with rituximab in patients with B-cell non-Hodgkin's lymphoma (NHL). *Blood.* 2000;96(suppl 1):577a. Abstract 2478.
96. Lundin J, Österborg A, Brittinger G, et al. CAMPATH-1H monoclonal antibody therapy for previously treated low-grade non-Hodgkin's lymphomas: a phase II multicenter study. *J Clin Oncol.* 1998;16:3257-3263.

97. Pawson R, Dyer MJS, Barge R, et al. Treatment of T-cell prolymphocytic leukemia with human CD52 antibody. *J Clin Oncol.* 1997;15:2667-2672.
98. Keating MJ, Byrd J, Rai K, et al. Multicenter study of CAMPATH-1H in patients with chronic lymphocytic leukemia refractory to fludarabine. *Blood.* 1999;94(suppl 1):705a. Abstract 3118.
99. Leonard JP, Coleman M, Schuster MW, et al. Epratuzumab, a new anti-CD22, humanized, monoclonal antibody for the therapy of non-Hodgkin's lymphoma: phase I/II trial results. *Blood.* 1999;94(suppl 1):92a. Abstract 404.
100. Leonard JP, Coleman M, Chadburn A, et al. Epratuzumab (anti-CD22 humanized monoclonal antibody) is an active and well-tolerated therapy for refractory/relapsed diffuse large B-cell non-Hodgkin's lymphoma. *Blood.* 2000;96(suppl 1). Abstract 2482.
101. Link BK, Wang H, Byrd JC, et al. Phase I trial of humanized 1D10 (Hu1D10) monoclonal antibody targeting class II molecules in patients with relapsed lymphoma. *Proc ASCO.* 2000;19:24a. Abstract 86.
102. Kaminski MS, Zasadny KR, Francis IR, et al. Iodine-131-anti-B1 radioimmunotherapy for B-cell lymphoma. *J Clin Oncol.* 1996;14:1974-1981.
103. Kaminski MS, Estes J, Zasadry KR, et al. Radioimmunotherapy with iodine 131 tositumomab for relapsed or refractory B-cell non-Hodgkin lymphoma: updated results and long-term follow-up of the University of Michigan experience. *Blood.* 2000;96:1259-1266.
104. Vose JM, Wahl RL, Saleh M, et al. Multicenter phase II study of iodine-131 tositumomab for chemotherapy-relapsed/refractory low-grade and transformed low-grade B-cell non-Hodgkin's lymphomas. *J Clin Oncol.* 2000;18:1316-1323.
105. Kaminski MS, Zelenetz AD, Press OW, et al. Pivotal study of Bexxar (Iodine I131 tositumomab) for chemotherapy-refractory low-grade or transformed low-grade B-cell non-Hodgkin's lymphomas. *J Clin Oncol.* 2001. In press.
106. Kaminski MS, Estes J, Tuck M, et al. Iodine I 131 tositumomab for previously untreated follicular lymphoma (FL). *Proc ASCO.* 2000;19:5a. Abstract 11.
107. Press OW, Eary JF, Gooley T, et al. A phase I/II trial of iodine-131-tositumomab (anti-CD20), etoposide, cyclophosphamide, and autologous stem cell transplantation for relapsed B-cell lymphomas. *Blood.* 2000;96:2934-2942.
108. Witzig TE, White CA, Wiseman GA, et al. Phase I/II trial of IDEC-Y2B8 radioimmunotherapy for treatment of relapsed or refractory CD20(+) B-cell non-Hodgkin's lymphoma. *J Clin Oncol.* 1999;17:3793-3803.
109. Witzig TE, White CA, Gordon LI, et al. Response to Zevalin is superior to response to rituximab regardless of age and extent of disease. *Proc ASCO.* 2001;20:279a. Abstract 1115.
110. Witzig TE, White CA, Flinn IW, et al. Zevalin radioimmunotherapy of rituximab-refractory follicular non-Hodgkin's lymphoma. *Blood.* 2000;96(suppl 1). Abstract 2183.
111. Horning SJ, Lucas JB, Younes A, et al. Iodine-131 tositumomab for non-Hodgkin's lymphoma (NHL) patients who progressed after treatment with rituximab: results of a multicenter phase II study. *Blood.* 2000;96(suppl 1). Abstract 2184.
112. Waters JS, Webb A, Cunningham D, et al. Phase I and pharmacokinetic study of bcl-2 antisense oligonucleotide therapy in patients with non-Hodgkin's lymphoma. *J Clin Oncol.* 2000;18:1809-1811.
113. Hsu FJ, Caspar C, Czerwinski D, et al. Tumor-specific idiotype vaccines in the treatment of patients with B-cell lymphoma—long term results of a clinical trial. *Blood.* 1997;89:3129-3135.
114. Bendandi M, Gocke CD, Kobrin CB, et al. Complete molecular remissions induced by patient-specific vaccination plus granulocyte-monocyte colony stimulating factor against lymphoma. *Nature Med.* 1999;10:1171-1177.
115. Freedman AS, Nadler LM. Which patients with relapsed non-Hodgkin's lymphoma benefit from high-dose therapy and hematopoietic stem-cell transplantation? *J Clin Oncol.* 1993;11:1841-1843.
116. Freedman AS, Gribben JG, Neuberg D, et al. High-dose therapy and autologous bone marrow transplantation in patients with follicular lymphoma. *Blood.* 1996;88:2780-2786.
117. Rohatiner AZS, Johnson PWM, Price CGA, et al. Myeloablative therapy with autologous bone marrow transplantation as consolidation therapy for recurrent follicular lymphoma. *J Clin Oncol.* 1994;12:1177-1184.
118. Freedman AS, Neuberg D, Mauch P, et al. Long-term follow-up of autologous bone marrow transplantation in patients with relapsed follicular lymphoma. *Blood.* 1999;94:3325-3333.
119. Darrington DL, Vose JM, Anderson JR, et al. Incidence and characterization of secondary myelodysplastic syndrome and acute myelogenous leukemia following high-dose chemoradiotherapy and autologous stem-cell transplantation for lymphoid malignancies. *J Clin Oncol.* 1994;12:2527-2534.
120. Miller JS, Arthur DC, Litz CE, Neglia JP, Miller WJ, Weisdorf DJ. Myelodysplastic syndrome after autologous bone marrow transplantation: an additional late complication of curative cancer therapy. *Blood.* 1994;83:3780-3788.
121. Stone RM, Neuberg D, Soiffer R, et al. Myelodysplastic syndrome as a late complication following autologous bone marrow transplantation for non-Hodgkin's lymphoma. *J Clin Oncol.* 1994;12:2535-2542.
122. Friedberg JW, Neuberg D, Stone RM, et al. Outcome in patients with myelodysplastic syndrome after autologous bone marrow transplantation for non-Hodgkin's lymphoma. J Clin Oncol. 1999;17:3128-2135.
123. van Besien KW, Khouri IF, Giralt S, et al. Allogeneic bone marrow transplantation for refractory and recurrent low-grade lymphoma: the case for aggressive management. *J Clin Oncol.* 1995;13:1096-1102.
124. Slavin S, Nagler A, Naparstek E, et al. Nonmyeloablative stem cell transplantation and cell therapy as an alternative to conventional bone marrow transplantation with lethal cytoreduction for the treatment of malignant and nonmalignant hematologic diseases. *Blood.* 1998;91:756-763.
125. Khouri IF, Keating M, Körbling M, et al. Transplant-lite: induction of graft-versus malignancy using fludarabine-based nonablative chemotherapy and allogeneic blood progenitor-cell transplantation as treatment for lymphoid malignancies. *J Clin Oncol.* 1998;16:2817-2824.
126. Alizadeh AA, Eisen MB, Davis RE, et al. Distinct types of diffuse large B-cell lymphoma identified by gene expression profiling. *Nature.* 2000;403:503-511.