Eur J Cancer, Vol. 27, No. 4, pp. 510–513, 1991.
Printed in Great Britain

0277–5379/91 $3.00 + 0.00

Modern Chemotherapeutic Regimens in the Management of Aggressive Non-Hodgkin Lymphoma: Can They Be Improved?

David J. Inwards and James O. Armitage

INTRODUCTION

THE NON-HODGKIN lymphomas (NHL) are a diverse group of disorders with widely divergent natural histories. The course of low grade NHL is characterised by prolonged survival with little potential for cure by current standard therapeutic interventions. In contrast, intermediate and high grade NHL are often rapidly fatal when untreated, but are among the most curable of all systemic human malignancies. These aggressive tumours are often exquisitely sensitive to chemotherapy. Factors which limit the outcome of chemotherapy for aggressive NHL include the existence of primarily resistant disease, emergence of resistance, relapse of disease, and regimen related toxicity. An understanding of the reasons for treatment failure, and the development of new treatment strategies, should allow for improved outcome in these increasingly common tumours.

CURRENT REGIMENS

In the past 20 years major progress has been made in the development of effective chemotherapy for intermediate and high grade NHL. What was once a nearly uniformly fatal disease is now curable in a considerable fraction of cases [1, 2]. Successive generations of regimens have been developed as treatment theories and philosophies have evolved.

Current regimens include ProMACE-CytaBOM [3], MACOP-B [4], COPBLAM III [5], CHOP [1], CAP-BOP [6], and LNH-84 [7] (Table 1). Experience has demonstrated that cure in aggressive NHL is predicated on the achievement of complete remission (CR) in response to initial chemotherapy [2]. Maintenance therapy appears to be ineffective in this setting [2]. Modern regimens reflect these principles in that they are intense in dosage but brief in duration. Traditional agents with well documented activity in NHL, such as doxorubicin and cyclophosphamide, form the foundation of these regimens. The inclusion of non-myelosuppressive drugs, such as bleomycin and vincristine, allows for administration of active agents during periods of treatment induced cytopenias and facilitates the simultaneous delivery of multiple active agents. Consideration of the Goldie–Coldman somatic mutation hypothesis has led to the administration of alternating non-crossresistant agents in an attempt to overcome the barrier of emerging drug resistance [8]. These regimens call for frequent administration of active agents in order to minimise tumour regrowth within, and between, chemotherapy cycles.

Table 1 compares several popular regimens. MACOP-B is extremely brief, requiring only 12 weeks to complete. It is dose intense for doxorubicin and includes daily administration of high dose prednisone and prophylactic antimicrobials. This regimen has the highest frequency of parenteral drug administration with intravenous chemotherapy scheduled on a weekly basis. Prognostic factors identifying patients unlikely to respond to this regimen were not identified by the original investigators, although few older patients were treated [9]. MACOP-B is poorly tolerated by patients age 65 and older [4] and may not be a wise choice in that subgroup of patients.

Unusual features of COPBLAM III include administration of vincristine and bleomycin by continuous infusion in an attempt to reduce toxicity and overcome resistance due to the cell cycle phase specific nature of these agents [5]. Provision for dose escalation of myelosuppressive agents is incorporated in this regimen. Initial results identified age as a prognostic factor in predicting survival in patients with diffuse large cell lymphoma treated with this regimen with 88% of patients age 60 or younger on the survival plateau versus only 42% of patients over age 60. Toxicity was considerable with 10% toxic deaths and a 39% incidence of pulmonary toxicity.

ProMACE-CytaBOM is an outpatient regimen that contains two alternating non-crossresistant arms [3]. This regimen's efficacy against a variety of histologies of aggressive NHL has recently been reconfirmed [10]. Prognostic factors predicting response to and/or survival following this regimen have not been reported to date. Interstitial pulmonary infiltrates developed in 35% of patients on the initial ProMACE-CytaBOM protocol [11]. Over half of these cases had histologic confirmation of *Pneumocystis carinii* infection. The addition of cotrimoxazole prophylaxis has largely eliminated this complication, leaving myelosuppression as the major toxicity of ProMACE-CytaBOM.

Table 1. Commonly used chemotherapy regimens for aggressive NHL

Ref.	Regimen	Histology	Cr rate (%)	Survival/FU (%)	Toxic deaths (%)
9	MACOP-B	DLC	84	69/18 mo. med.	5
5	COPBLAM III	DLC	84	65/24 mo. min	10
12	ProMACE-CytaBOM	Aggressive	79	70/24 mo. med.	4
26,10	CHOP	DLC	53	30/3 yr. min.	3
7	LNH-84	Aggressive	75	67/23 mo. med.	8
6	CAP-BOP	Aggressive	65	42/36 mo. med.	7
25,1	m-BACOD	Aggressive	72	54/48 mo. med.	6–13

CR = complete remission; FU = follow-up period; DLC = diffuse large cell.

Correspondence to D. J. Inwards.
D. J. Inwards is at the Department of Hematology and Oncology and J. O. Armitage is at the Department of Internal Medicine, University of Nebraska Medical Center, 600 South 42nd Street, Omaha, NE 68198–3330, U.S.A.
Received 10 Jan. 1991; accepted 22 Jan. 1991.

CHOP was one of the first doxorubicin containing regimens for NHL. Extensive clinical trials with long term follow-up have established this regimen as a standard of comparison of treatment programs for aggressive NHL [1]. CHOP is a simple regimen that consists of repeated outpatient administrations of a four drug combination. Age has been identified as a prognostic factor in predicting survival of patients treated with this regimen. Reported toxic death rates for CHOP are low despite the inclusion of many elderly patients in treatment trials (Table 1).

LNH-84 is a novel regimen which incorporates twelve drugs in three treatment phases [7]. Omission of the final, or intensification, phase does not alter relapse rates. This regimen has proven activity against a variety of aggressive lymphomas. Prognostic factors predicting lower CR rates and higher relapse rates include classic indicators of extensive disease, such as stage, B symptoms, diameter of tumour and bone marrow involvement. The major toxicity reported in LNH-84 treatment trials has been myelosuppression. The induction phase has been associated with a 7% frequency of septic death.

CAP-BOP is an outpatient regimen composed of two alternating non-crossresistant arms. Myelosuppressive drugs are dose reduced by one third when patients over age 70 are treated with this regimen. Patients over age 60 with aggressive NHL treated with CAP-BOP have been shown to have CR rates, CR durability and toxicity similar to patients under age 60 [6]. Differences in survival are apparently explained by causes unrelated to lymphoma or chemotherapy. Myelosuppression and bleomycin induced pulmonary toxicity have been the primary toxicities of CAP-BOP. Pulmonary function monitoring reduces the incidence of clinically relevant interstitial pneumonitis.

Comparison of regimens for aggressive NHL has been limited by a paucity of multiple regimen prospective randomised trials. Factors including uneven distribution of prognostic factors among patient populations and variable follow-up periods make comparisons between studies unreliable [2]. At this time no single regimen can be identified that is clearly superior to the others. It is also unclear whether newer regimens are superior to earlier regimens, such as CHOP. Recently initiated trials are designed to address these issues [1].

REASONS FOR TREATMENT FAILURE

Even in the most promising reports, at least one third of patients with aggressive NHL treated with chemotherapy fail to achieve prolonged survival [9,12]. Treatment failures may be a consequence of failure to achieve an initial remission, relapse of tumour, or drug induced toxicity. Properties of the tumour and/or features of the patient may predict treatment failure.

Primary and acquired drug resistance are commonly encountered in aggressive NHL. Although in most cases the mechanism remains unknown, recent publications have identified several potential mechanisms. Deuchars and Ling [13] described a membrane glycoprotein, known as P-glycoprotein, which appears to serve as a pump mediating ATP dependent efflux of a number of naturally occurring drugs including vincristine, doxorubicin, actinomycin D, vinblastine and daunorubicin. Overexpression of P-glycoprotein confers the MDR (multidrug resistance) phenotype, in which resistance to these and other related drugs is evident. The gene coding for P-glycoprotein has been named *mdr*1. The homologous gene *mdr*3 has been identified in several haematological malignancies [14]. Salmon *et al.* [15] studied a variety of tumour types and found that P-glycoprotein expression correlated with *in vitro* doxorubicin resistance. Included in this study were three P-glycoprotein positive doxorubicin resistant lymphomas and three P-glycoprotein negative lymphomas, one of which was doxorubicin resistant. Further studies are needed to define the frequency and significance of P-glycoprotein overexpression in untreated, resistant and relapsed NHL.

Diddens and Niethammer [16, 17] studied resistance to methotrexate in a number of *in vitro* systems including a Burkitt's lymphoma subline (RAJI). These authors were able to produce a 550 × normal level of cellular dihydrofolate reductase in RAJI cells by means of progressive increases in ambient methotrexate concentration. This adaptation conferred methotrexate resistance, and was mediated by amplification of the dihydrofolate reductase gene. Other cell lines demonstrated decreased transport of methotrexate as a mechanism of drug resistance.

Several studies have demonstrated that high proliferative activity as measured by flow cytometric S-phase determination correlates with poor response to therapy and survival [18, 19]. In at least one study, S-phase analysis was found to be a stronger predictor of survival than histopathology, stage or age [20]. Increasing proliferative activity on serial biopsies in relapsed disease has been shown to predict poor survival as well [21]. Other methods of measuring prliferative activity have yielded similar results. Silvestrini *et al.* [22] found that tritiated thymidine labeling index gives additional information beyond age, stage and histopathology in predicting 6 year survival in non-Hodgkin lymphoma. Again high proliferative activity, as reflected by a high labelling index, correlates with higher grade lymphomas and predicts a poor prognosis. Nuclear proliferation antigen staining by Ki-67 demonstrates the same relationship between proliferative activity and prognosis [23,24]. The mechanism of therapy resistance conferred by high proliferative activity is unclear. One possible explanation is enhanced emergence of drug resistant clones through increased frequency of somatic mutations. Another is impaired cytoreduction secondary to rapid tumour regrowth between drug administrations.

A number of prognostic factors have been identified which appear to reflect differences in tumour burden. These include tumour diameter >10 cm [25] and stage [2]. More advanced tumours may be more likely to contain drug resistant clones than smaller, lower stage tumours simply due to an increased number of mutation generating cell divisions. Drug delivery to the inner regions of bulky tumours may be impaired by limited vascular access. Finally, patients with advanced disease may be unable to tolerate full doses of chemotherapy, or may metabolise chemotherapeutic agents differently than patients with more limited disease.

Elevated LDH levels have been associated with low CR rates and an increased risk of lymphoma relapse [7]. High lactate dehydrogenase (LDH) levels likely reflect rapid tumour proliferation and/or extensive tumour. Potential mechanisms of therapy resistance are as discussed in association with high proliferative activity and large tumour burden.

In the past, two years of continuous complete remission has been accepted as evidence of probable cure of aggressive NHL. Prolonged follow-up of Southwest Oncology Group patients treated with CHOP and CHOP variants has demonstrated that as many as 50% of complete remissions may relapse, and that relapses as late as 7 years following chemotherapy occur [26]. Subclinical persistence of disease capable of producing delayed relapse remains a significant barrier to successful treatment outcome.

Elimination of tumour by a chemotherapeutic regimen may still result in an unsatisfactory outcome if excessive toxicity is

produced. Unacceptable toxicity may be a consequence of the composition of a particular regimen, or to inappropriate use of an otherwise acceptable regimen in patients who are unusually susceptible to the toxicity of that regimen. The toxicity may be more severe when the patient is elderly [9] or has a significant medical condition involving major organs. Severely immunocompromised patients with NHL have proven to be particularly sensitive to the toxic effects of chemotherapy [27]. Increased utilisation of organ transplantation technology and the epidemic of AIDS have combined to greatly increase the prevalence of this clinical situation.

Attempts to minimise toxicity by attenuating therapy may compromise therapeutic efficacy. Dixon *et al.* [28] reported decreased complete remission rates in patients over age 65 given initial 50% dose reductions of CHOP on the basis of age alone in comparison to patients over age 65 given full dose therapy. This result is particularly unsatisfying given that the group receiving reduced doses did not have appreciably diminished toxicity.

IMPROVING THE RESULTS OF THERAPY

Strategies to improve chemotherapeutic outcome in aggressive non-Hodgkin lymphoma should involve optimal application of currently available agents and regimens as well as investigation of new therapies. As mechanisms of drug resistance are elucidated, specific strategies to overcome resistance will be investigated. In a preliminary report, Dalton *et al.* [29] demonstrated that some P-glycoprotein positive chemotherapy refractory haematologic malignancies respond to a combination of verapamil and conventional vincristine, doxorubicin and dexamethasone chemotherapy. *In vitro* studies had previously shown that several substances, including verapamil, are capable of blocking P-glycoprotein mediated drug efflux. Other potential techniques to circumvent P-glycoprotein mediated drug resistance include avoidance of cross-resistant drugs, P-glycoprotein directed immunotherapy and P-glycoprotein blockade [13].

Dose escalation of drugs with favorable dose response relationships may overcome drug resistance. Certain drugs, such as methotrexate and cytarabine, may be dose escalated without stem cell rescue. Other drugs with dose limiting myelosuppression may be dose escalated in bone marrow transplant conditioning regimens. The primary role of transplantation is in the treatment of patients who have failed first-line therapy. Optimal application of this technique depends upon the identification of patients whose tumours are still responsive to conventional doses of chemotherapeutic agents [30].

The role of colony stimulating factors in future NHL chemotherapy regimens remains to be defined. By abrogating drug induced myelosuppression, these factors may improve outcome via reduced treatment related toxicity. Alternatively, the use of these factors may allow for dose escalation of myelosuppressive agents.

Strategies incorporated in newer chemotherapy regimens address the problem of kinetic resistance. High frequency drug administration should reduce tumour regrowth between treatments. Alternating non-crossresistant therapy is designed to prevent the emergence of drug resistant clones. Stratification of regimen comparison trial subjects on the basis of proliferative activity will allow assessment of the efficacy of these approaches.

Ideally, early detection of aggressive NHL with prompt initiation of therapy while tumour burden is small would be expected to improve outcome. Efforts to enhance early detection may be impractical for the general population given the lack of an appropriate screening test. However, some high risk populations, such as the severely immunocompromised, may benefit from surveillance for early stage aggressive NHL. Outcome in selected patients with locally bulky disease may be improved by systemic chemotherapy followed by involved field radiotherapy. However, the value of this approach needs to be documented before it is widely applieed since it may compromise the use of future marrow transplantation.

The barrier of late relapse deserves further attention. Long term follow-up will be required in order to determine which regimens provide the most durable remissions, and what factors predict late relapse.

Knowledge of regimen related toxicity profiles for a variety of alternative regimens, and consideration of an individual patient's susceptibility to specific toxicities based on pretreatment factors such as age and major organ system disease should allow for optimal regimen selection for individual cases. This approach may improve treatment outcome by reducing toxicity. In order to maintain efficacy it is important to attempt to administer the chosen regimen as it is was administered in treatment trials. Certain regimens may need to be utilised in specific situations such as in the treatment of aggressive NHL in the immunocompromised host [27].

Ideal therapy for aggresssive NHL would eliminate tumour without damaging host tissue. Systemic administration of chemotherapeutic agents may achieve the former, but is nonspecific in its effect on normal tissue. Mechanisms to deliver active agents exclusively to tumour cells could theoretically decrease toxicity while providing a concentrated assault on the tumour in question. This has been an area of active research [31]. In general, antibodies or antibody fragments are used to identify targetted cells by means of antigen antibody interactions. A variety of effectors may be employed including biotoxins, radioisotopes and cytotoxic drugs. Alternatively, a two stage procedure has been described in which the initial step is tumour localisation of an enzyme capable of converting an inactive prodrug into a cytotoxic drug, followed by administration of prodrug. This process is known as antibody directed enzyme prodrug therapy (ADEPT). These approaches are far from ready for general application, but are theoretically appealing and merit further research.

New chemotherapeutic agents, combinations and schedules will continue to be investigated in the search for more efficacious and less toxic therapy. In designing and evaluating these trials it will be necessary to consider the current barriers to improved outcome discussed previously.

SUMMARY

Major advances in the treatment of aggressive NHL have occurred in the last 20 years. Modern combination chemotherapy provides a complete remission in a majority of patients and cures a significant number of these patients. Treatment failures are a consequence of the existence or development of resistant disease and regimen related toxicity. Identification of mechanisms of tumour resistance and prognostic factors, together with consideration of regimen related toxicity, should aid in the optimal application of currently available agents and regimens and guide the development of future regimens. Novel delivery systems may allow more tumour specific therapy in the future.

1. Fisher RI, Miller TP, Dana BW, *et al.* Southwest Oncology Group clinical trials for intermediate- and high-grade non-Hodgkin's lymphomas. *Semin Hematol* 1987, **24** (suppl. 1), 21–25.

2. Armitage JO, Cheson BD. Interpretation of clinical trials in diffuse large-Cell lymphoma. *J Clin Oncol* 1988, **6**, 1335–1347.
3. Fisher RI, DeVita VT, Hubbard SM, *et al.* Randomized trial of ProMACE-MOPP vs. ProMACE-CytaBOM in previously untreated advanced stage, diffuse aggressive lymphomas. *Proc Am Soc Clin Oncol* 1984, **3**, 242.
4. Klimo P, Connors JM. MACOP-B chemotherapy for the treatment of diffuse large-cell lymphoma. *Ann Intern Med* 1985, **102**, 596–602.
5. Boyd DB, Coleman M, Papish SW, *et al.* COPBLAM III: Infusional combination chemotherapy for diffuse large-cell lymphoma. *J Clin Oncol* 1988, **6**, 425–433.
6. Vose JM, Armitage JO, Weisenburger DD, *et al.* The importance of age in survival of patients treated with chemotherapy for aggressive non-Hodgkin's lymphoma. *J Clin Oncol* 1988, **6**, 1838–1844.
7. Coiffier B, Gisselbrecht C, Herbrecht R, *et al.* LNH-84 regimen: A multicenter study of intensive chemotherapy in 737 patients with aggressive malignant lymphoma. *J Clin Oncol* 1989, **7**, 1018–1026.
8. Goldie JH, Coldman AJ, Gudauskas GA. Rationale for the use of alternating non-cross-resistant chemotherapy. *Cancer Treat Rep* 1982, **66**, 439–449.
9. Klimo P, Connors JM. Updated clinical experience with MACOP-B. *Semin Hematol* 1987 **24** (suppl. 1), 26–34.
10. Miller TP, Dahlberg S, Weick JK, *et al.* Unfavorable histologies of non-Hodgkin's lymphoma treated with ProMACE-CytaBOM: a groupwide Southwest Oncology Group study. *J Clin Oncol* 1990, **8**, 1951–1958.
11. Browne MJ, Hubbard SM, Longo DL, *et al.* Excess prevalance of *Pneumocystis carinii* pneumonia in patients treated for lymphoma with combination chemotherapy. *Ann Intern Med* 1986, **104**, 338–344.
12. Longo D, DeVita V, Duffey P, *et al.* Randomized trial of ProMACE-MOPP (Day (D) 1, D 8) (PM) vs ProMACE-CytaBOM (PC) in stage II–IV aggressive non-Hodgkin's lymphoma. *Proc Am Soc Clin Oncol* 1987, **6**, 206.
13. Deuchars KL, Ling V. P-Glycoprotein and multidrug resistance in cancer chemotherapy. *Semin Oncol* 1989, **16**, 156–165.
14. Herweijer H, Sonneveld P, Baas F, *et al.* Expression of *mdr*1 and *mdr*3 multidrug-resistance genes in human acute and chronic leukemias and association with stimulation of drug accumulation by cyclosporine. *J Natl Cancer Inst* 1990, **82**, 1133–1140.
15. Salmon SE, Grogan TM, Miller T, *et al.* Prediction of doxorubicin resistance in vitro in myeloma, lymphoma, and breast cancer by P-glycoprotein staining. *J Natl Cancer Inst* 1989, **81**, 696–701.
16. Diddens H, Niethammer D, Jackson RC. Patterns of cross-resistance to the anti-folate drugs trimetrexate, metoprine, homofolate, and CB3717 in human lymphoma and osteosarcoma cells resistant to methotrexate. *Cancer Res* 1983, **43**, 5286–5292.
17. Niethammer D, Diddens H, Gekeler V, *et al.* Resistance to methotrexate and multidrug resistance in childhood malignancies. *Adv Enzyme Regul* 1989, **29**, 231–245.
18. O'Brien CJ, Holgate C, Quirke P, *et al.* Correlation of morphology, immunophenotype, and flow cytometry with remission induction and survival in high grade non-Hodgkin's lymphoma. *J Pathol* 1989, **158**, 31–39.
19. Wooldridge TN, Grierson HL, Weisenburger DD, *et al.* Association of DNA content and proliferative activity with clinical outcome in patients with diffuse mixed cell and large cell non-Hodgkin's lymphoma. *Cancer Res* 1988, **48**, 6608–6613.
20. Christensson B, Lindemalm C, Johansson B, *et al.* Flow cytometric DNA analysis: A prognostic tool in non-Hodgkin's lymphoma. *Leuk Res* 1989, **13**, 307–314.
21. Joensuu H, Klemi PJ, Jalkanen S. Biologic progression in non-Hodgkin's lymphoma. *Cancer* 1990, **65**, 2564–2571.
22. Silvestrini R, Costa A, Giardini R, *et al.* Prognostic implications of cell kinetics, histopathology and pathologic stage in non-Hodgkin's lymphomas. *Hematol Oncol* 1989, **7**, 411–422.
23. Weiss LM, Strickler JG, Medeiros LF, *et al.* Proliferative rates of non-Hodgkin's lymphomas as assessed by Ki-67 antibody. *Hum Pathol* 1987, **18**, 1155–1159.
24. Grogan TM, Lippman SM, Spier CM, *et al.* Independent prognostic significance of a nuclear proliferation antigen in diffuse large cell lymphomas as determined by the monoclonal antibody Ki-67. *Blood* 1988, **71**, 1157–1160.
25. Shipp MA, Harrington DP, Klatt MM, *et al.* Identification of major prognostic subgroups of patients with large-cell lymphoma treated with m-BACOD or M-BACOD. *Ann Intern Med* 1986, **104**, 757–765.
26. Coltman CA, Dahlberg S, Jones SE, *et al.* CHOP is curative in thirty percent of patients with diffuse large cell lymphoma: A twelve year Southwest Oncology Group follow up. *Proc Am Soc Clin Oncol* 1986, **5**, 197.
27. Levine AM. Lymphoma in acquired immunodeficiency syndrome. *Semin Oncol* 1990, **17**, 104–112.
28. Dixon DO, Neilan B, Jones SE, *et al.* Effect of age on therapeutic outcome in advanced diffuse histiocytic lymphoma: the Southwest Oncology Group experience. *J Clin Oncol* 1986, **4**, 295–305.
29. Dalton WS, Grogan TM, Meltzer PS, *et al.* Drug-resistance in multiple myeloma and non-Hodgkin's lymphoma: Detection of P-glycoprotein and potential circumvention by addition of verapamil to chemotherapy. *J Clin Oncol* 1989, **7**, 415–424.
30. Philip T, Armitage JO, Spitzer G, *et al.* High-dose therapy and autologous bone marrow transplantation after failure of conventional chemotherapy in adults with intermediate-grade or high-grade NHL. *N Engl J Med* 1987, **316**, 1493–1498.
31. Bagshawe KD. Towards generating cytotoxic agents at cancer sites. *Br J Cancer* 1989, **60**, 275–281.