Drugs 2001; 61 (5): 593-611
0012-6667/01/0005-0593/$27.50/0

Achieving Optimal Outcomes in Chronic Lymphocytic Leukaemia

Terry J. Hamblin

Department of Haematology, Royal Bournemouth Hospital, Bournemouth, England

Contents

Abstract

Chronic lymphocytic leukaemia (CLL) is a disease of late middle age and older. The majority of patients are diagnosed because of a lymphocytosis of at least 5 x 10^9/L on an incidental blood count. It needs to be distinguished from mantle cell lymphoma and splenic marginal zone lymphoma by lymphocyte markers. The immunophenotype of CLL is sparse surface immunoglobulin, CD5+, CD19+, CD23+, CD79b–, and FMC7–. The disease is staged according to the presence of lymphadenopathy and/or splenomegaly and the features of bone marrow suppression. Most patients have an early stage of disease when diagnosed and perhaps 50% will never progress. This group of patients have a normal life expectancy and do not require treatment beyond reassurance.

Progression involves an increasing white cell count, enlarging lymph nodes and spleen, anaemia and thrombocytopenia. Complications of progression include autoimmune haemolytic anaemia and thrombocytopenia, immunodeficiency, and the development of a more aggressive lymphoma. A range of prognostic factors is available to predict progression, but most haematologists rely on close observation of the patient.

Intermittent chlorambucil remains the first choice treatment for the majority of patients. Combination chemotherapy offers no advantage. Intravenous fludarabine is probably more effective than chlorambucil, but no trial has yet shown a survival advantage for using it first rather than as a salvage treatment in patients not responding to chlorambucil. It is at least 40 times as expensive as chlorambucil. Cladribine may be as effective as fludarabine, although it has been used less and is even more expensive.

Patients who relapse after chlorambucil should be offered retreatment with the same agent and if refractory should be switched to fludarabine, which may also be offered for retreatment on relapse. For patients refractory to both drugs, a variety of options are available. High dose corticosteroids, high dose chlorambucil, CHOP (cyclophosphamide, prednisolone, vincristine and doxorubicin), anti-CD52, anti-CD20 and a range of experimental drugs which are being evaluated in clinical trials.

Younger patients should be offered the chance of treatment with curative intent, preferably in the context of a clinical trial. Autologous stem cell transplantation after achieving a remission with fludarabine has relative safety and may produce molecular complete remissions. Only time will tell whether some of these patients are cured but it seems unlikely. Standard allogeneic bone marrow transplant is probably too hazardous for most patients, but non-myeloablative regimens hold out the hope of invoking a graft-versus-leukaemia effect without a high tumour-related mortality.

Trials of immunotherapy are exciting options for a few patients in specialised centres.

Chronic lymphocytic leukaemia (CLL) is the second most common haematological malignancy in the Western world (after myelodysplastic syndrome). In the UK, the incidence is 5.6/100 000.[1] It is a disease of the elderly, with a median age of presentation of 69 years[2] and an incidence in those aged > 75 years of 27/100 000.[1]

1. Definition

Only recently have haematologists agreed on a definition of CLL. Several clinical entities have similar cellular morphology and clinical presentations, with the result that many clinical trials have been contaminated by the inclusion of patients with more or less malignant conditions with a superficial resemblance to CLL. A National Cancer Institute (NCI) sponsored Working Group recommended the diagnostic criteria shown in table I.[3]

These criteria have been to some extent superseded by the Royal Marsden scoring system that makes use of further immunological markers (table I).[4,5] Even within true CLL there is a degree of heterogeneity with respect to immunological markers, but most patients will score 4 or 5 out of 5 for these criteria.[5] The major interlopers into the clinical trials have been mantle cell lymphoma (MCL) and splenic marginal zone lymphoma (SMZL).

1.1 Mantle Cell Lymphoma

MCL[6] was previously known as diffuse centrocytic lymphoma. It is believed to arise from the follicular mantle in the lymph node. The cells are

Table I. Definition of chronic lymphoctic leukaemia

National Cancer Institute Working Group Definition[3]	Royal Marsden Score[4,5]
Blood	
Lymphocytosis of $\geq 5 \times 10^9$/L; morphologically mature; persisting ≥4 wks	
Bone marrow	
≥30 lymphocytosis, normocellular or hypercellular Ig; (not essential)	
Cell markers	Score one for each consistent marker
Monoclonal surface Ig; CD5+; sparce surface Ig	CD5+; CD23+; sparce surface Ig; CD79b neg; FMC7 neg

Ig = immunoglobulin.

thought to be naïve B cells, yet to confront antigen, although this has been disputed.[7] It is generally a very malignant tumour. Although initially sensitive to chemotherapy, it rapidly relapses and the median survival is <2 years. There are no long term survivors even with the most aggressive therapy. Cell markers distinguish it from CLL (table II). There is a characteristic t(11;14) chromosomal translocation which upregulates the expression of nuclear cyclin D1 (also known as BCL-1), which can be stained for. It is still a matter of dispute as to whether there is a type of CLL with the t(11;14) translocation,[8] but certainly some patients with this translocation resemble CLL. Their disease usually behaves like MCL.

1.2 Splenic Marginal Zone Lymphoma

SMZL[9] is the same condition as splenic lymphoma with villous lymphocytes. If only the bipolar villi, characteristic of this condition, were a constant feature there would be less confusion. In general, the cells are larger and with more cytoplasm than in CLL. The tumours arise from the marginal zone surrounding the follicle centre and the cells have been educated following exposure to antigen.[10] Unfortunately, some MCLs can assume marginal zone appearances in splenic histology,[11] which probably accounts for the fact that some marginal zone tumours have been reported as CD5 positive or carrying the t(11;14) translocation. The immunophenotype is shown in table II. Clinically, SMZL is benign in most patients, often requiring no treatment. Splenomegaly is common and lymphadenopathy rare. Two-thirds of patients have a circulating paraprotein.

1.3 Other Lymphomas

More rarely, other lymphoid tumours have been confused with CLL including large granular lymphocytic leukaemia, small-cell Sezary syndrome, T cell prolymphocytic leukaemia, follicular lymphoma, B cell prolymphocytic leukaemia, hairy cell leukaemia (HCL) and hairy cell variant. With up-to-date resources, these mistakes should no longer occur. However, despite these modern resources, we still occasionally see leukaemias that do not fit in well with any category. In general these have a poor prognosis.

2. Staging Systems

There are two staging systems in current use: the American Rai system[12] and the European Binet system[13] (table III). Both systems have prognos-

Table II. Differential diagnosis - cell markers

	Surface Ig	CD5+	CD19+	CD23+	Cyclin D1	Cd79b	FMC7
CLL	Sparse	Pos	Pos	Pos	Neg[a]	Neg	Neg
MCL	Dense	Pos	Pos	Neg	Pos	Pos	Pos
SMZL	Dense	Neg[b]	Pos	Neg	Neg[b]	Pos	Pos

a Some CLLs have been described with the t(11;14) translocation and consequent cyclin D1 upregulation. The matter is contentious, but the author's contention is that these are cases of MCL masquerading as CLL.

b Some MCLs adopt the morphology of SMZL in the spleen and are consequently misdiagnosed.

CLL = chronic lymphocytic leukaemia; **Ig** = immunoglobulin; **MCL** = mantle cell lymphoma; **Neg** = negative; **Pos** = positive; **SMZL** = splenic marginal zone lymphoma.

Table III. Staging systems for chronic lymphocytic leukaemia

Stage	Characteristics	Survival (y)
Rai		
0	Lymphocytosis in blood and bone marrow only	12
I	Lymphocytosis plus lymphadenopathy	7
II	Lymphocytosis plus splenomegaly or hepatomegaly	7
III	Lymphocytosis plus anaemia (Hb <110 g/L)	<1
IV	Lymphocytosis plus thrombocytopenia (platelets $<100 \times 10^9$/L)	<1
Binet[a]		
A	<3 sites involved, Hb >100 g/L, platelets $>100 \times 10^9$/L	9
B	≥3 sites involved, Hb >100 g/L, platelets $>100 \times 10^9$/L	5
C	Hb <100 g/L, platelets $>100 \times 10^9$/L	2

a Involved sites are liver, spleen and lymph nodes in inguinal, axilliary and cervical regions.

Hb = haemoglobin levels.

tic value. Neither system decrees that the anaemia and thrombocytopenia should be the result of marrow failure, but clearly when they have another cause they do not necessarily carry the same grave prognosis. It is bizarre that the same levels of haemoglobin are required for both genders, but it is nevertheless true. Several groups have noted that a falling haemoglobin is a poor prognostic feature even if it is not so severe as to advance the stage of the disease.

3. Other Prognostic Factors

Evidence will be presented later in this review to show that early stage CLL does not benefit from early treatment, and that late stage disease is incurable. All late stage disease was once at an early stage. Because of this, it would be useful to be able to distinguish at an early stage those patients who will progress from those who will never have any trouble. A number of adverse prognostic factors have been identified. Absolute lymphocyte count is not of itself a prognostic factor, although patients with counts $<30 \times 10^9$/L are best left untreated.[14] A lymphocyte doubling time of <12 months carries an adverse prognosis.[15] Diffuse bone marrow histology carries a worse prognosis than either nodular or interstitial patterns.[16] Typical rather atypical morphology,[17] low serum CD23+[18] and β_2-microglobulin[19] levels, normal karyotype or 13q deletions rather than trisomy 12 or 11q deletions,[20] and CD38 negativity[21] all denote the more benign variant of the disease.

The French group described an especially benign A′ group of patients with haemoglobin (Hb) >120 g/L and lymphocyte counts $<30 \times 10^9$/L with a survival curve identical to age-matched controls.[22] Similarly, a Spanish group[23] described smouldering CLL with the characteristics in table IV.

Two recent studies suggest that CLL is really two diseases.[24,25] One, deriving from memory B cells, detected by the presence of somatically mutated immunoglobulin (Ig) variable region (V) genes, has a median survival of 25 years, while the other, deriving from naïve B cells with unmutated V genes, has a median survival of only 8 years. Sequencing Ig V genes is a costly and time-consuming technique unavailable to most laboratories. One group has suggested that CD38 positivity might be a surrogate assay,[25] but the other has disputed this.[26] The importance of this study is that it distinguishes two separate diseases, one of which requires no treatment. There is no suggestion that one could transform into the other.

4. Complications of Chronic Lymphocytic Leukaemia

Patients with CLL seldom die of leucostasis. Lymphadenopathy may be unsightly, but it is not invasive. In the abdomen, enlarged nodes seldom cause ureteric obstruction. As with most leukaemias, bone marrow failure is the principle hazard.

Table IV. Criteria for smouldering chronic lymphocytic leukaemia

Stage A
Non-diffuse bone marrow histopathology
Haemoglobin ≥130 g/L
Lymphocyte count $<30 \times 10^9$/L
Lymphocyte doubling time >12 months

The drugs used to treat the disease themselves damage bone marrow stem cells. In addition, patients with CLL, including those with the more benign subtype, develop immunodeficiency. There is a profound hypogammaglobulinaemia, but T cells are also abnormal. Between 10% and 20% of patients develop autoimmune complications of which autoimmune haemolytic anaemia (AIHA) is the most common,[27] but immune thrombocytopenia,[28] pure red cell aplasia,[29] paraneoplastic pemphigus,[30] acquired angio-oedema[31] and glomerulonephritis[32] also occur.

Three types of transformation of CLL are reported, all to some extent misunderstood. Prolymphocytic (sometimes prolymphocytoid) transformation refers to the presence in the blood of more than 10% prolymphocytes in a patient who otherwise clearly has CLL.[33] Although the number of prolymphocytes may increase with time, in many cases it remains constant. Others have referred to this as one of the forms of atypical CLL.[34] Prolymphocytic leukaemia is a quite separate disease, and one does not transform into the other.

Transformation to acute lymphoblastic leukaemia is even more controversial. Most incidences belong to a time when the diseases were more poorly defined[35] and most CLL experts are skeptical that such a phenomenon exists.

Richter's syndrome, the development of an aggressive lymphoma on a background of CLL, certainly occurs,[36] but there are few patients in whom the aggressive lymphoma has been shown to be a sub-clone of the CLL.[37,38] An interesting recent suggestion implicates the proliferation of Epstein-Barr virus (EBV)-stimulated clones in an immunodeficient patient.[39]

Many workers believe that other cancers, both solid tumours and haematological neoplasms are more common in patients with CLL, but because these are all quite common diseases of the elderly it is difficult to be sure that this is so.

5. Aims of Treatment

The primary aim of all medical treatment is 'first do no harm'. Since there is a group of patients in which CLL causes no symptoms and does not shorten life, it is important to identify these and refrain from treating them. For those with CLL that causes mortality or morbidity, the aim, were it possible, should be cure, and if not, the greatest prolongation of life for the least toxicity. For acute leukaemias and aggressive lymphomas, complete remission (CR) is a prerequisite for the optimal outcome. It may not necessarily be the case for CLL, which even if it is progressive, is often quite slowly so. In an older person, the disease may outlive the patient. Nevertheless, the NCI Working Group has recommended criteria of response which include CR.[40] A modification of these recommendations introduced the concept of a nodular partial response (PRn).[3] These patients fulfilled the criteria for a CR except for the presence of lymphoid nodules on trephine biopsy (table V).

In all these responses it is usual to find that the disease remains easily detectable by more sensitive methods. A population of CD5/CD19 positive cells can be detected by dual colour immunofluorescence, whereas molecular techniques will detect a single Ig heavy-chain-gene rearrangement.

6. The Option of No Treatment

Large numbers of patients (76% in one series[2]) present incidentally without symptoms or signs, simply because a blood test has been requested for an unrelated reason. In many of these patients CLL never progresses. Clearly, no treatment is the best option for these patients, but those who do need treatment often present in the same way. At the moment there is no sure way of identifying those who do not need treatment.

Several trials have addressed the question as to whether in stage A (Binet) or stage I and II (Rai) patients it would be safe to avoid treatment until progression occurs. A meta-analysis of 2048 such patients in 7 trials randomised between immediate or deferred treatment with chlorambucil (with or without prednisolone) suggested no benefit for either arm.[41] The 10-year survival was slightly worse (but not statistically significantly so) for those treated early (44 *vs* 47%). There is, therefore,

Table V. National Cancer Institute criteria for response (improvements must persist for at least 2 months) in patients with chronic lymphocytic leukaemia

Criterion	CR	PRn	PR
Hb g/L	≥110	≥110	≥110 or 50% reduction in transfusion requirement[a]
$N \times 10^9$/L	≥1.5	≥1.5	≥1.5 or 50% improvement[a]
$Pl \times 10^9$/L	≥100	≥100	≥100 or 50% improvement[a]
$L \times 10^9$/L	≤4	≤4	50% reduction
BM lymphocytes	≤30% no nodules in trephine biopsy	≤30% with nodules in trephine biopsy	No requirement
LN	0	0	50% reduction
L/S	0	0	50% reduction
Other	Absence of constitutional symptoms	Absence of constitutional symptoms	

a Only one of these required for a partial response.

BM = bone marrow; **CR** = complete response; **Hb** = haemoglobin; **L** = lymphocytes; **LN** = lymph nodes; **L/S** = liver/spleen; **N** = neutrophils; **Pl** = platelets; **PR** = partial response; **PRn** = nodular partial response.

no need to treat patients with stage A disease with chlorambucil unless there is evidence of a falling haemoglobin or platelet count, progression to a later stage of disease, unsightly or uncomfortable lymphadenopathy, a lymphocyte doubling time of <12 months or transformation of the disease.

In a French study,[42] 51% of patients allocated to the deferred treatment arm eventually required treatment, and 27% eventually died of a cause related to CLL. The outcome of patients who progressed to stage B was the same as patients who presented *de novo* with stage B.

Of course, were a superior treatment to chlorambucil to become available the question of early versus delayed treatment in stage A disease might have to be revisited.

7. Corticosteroids

Corticosteroids are effective agents in many cases of CLL. They are generally reserved as first-line agents in patients presenting with autoimmune haemolytic anaemia or thrombocytopenia, or as salvage therapy in patients with refractory disease. The dose for autoimmune disease is prednisolone 40 mg/m^2 daily for 14 days, tapering over another 2 weeks. Prednisolone alone will control CLL in about 10% of patients, but because of steroid-induced adverse effects it should not be used as a treatment of choice. Characteristically, patients on corticosteroids have an initial rise in lymphocyte count. This is due to redistribution of lymphocytes among compartments and does not carry a sinister significance.

High dose methylprednisolone has a place in end-stage, refractory CLL. Dosages of 250 mg/m^2 daily for 5 days often restore normal Hb and platelet counts, although the risk of infection is increased.[43]

8. Alkylating Agents

Chlorambucil is well tolerated in CLL as it avoids the usual alkylating agent adverse effects of alopecia and gastrointestinal intolerance. Dosages from 0.1 mg/kg daily given indefinitely to 0.7 mg/kg daily for 4 days every month have been used. No randomised study has demonstrated that one regimen is preferable. Many doctors suspect that continuous chlorambucil might induce secondary myelodysplastic syndrome, but there is no firm evidence that his is so.

A Croatian trial[44] compared a continuous high dosage of chlorambucil (15 mg/day) with a single dose (75mg) given intermittently every 4 weeks together with prednisolone. The prednisolone dosage was 50 mg/day tapered to 15 mg/day over 6 weeks followed by 30 mg/day for 7 days with each subsequent course of chlorambucil. The high daily dose was associated with greater toxicity and di-

minished compliance but gave a higher CR rate (70 *vs* 30%) and a significantly longer survival (median survival 6 *vs* 3 years; $p < 0.01$). It should be noted, however, that the requirements for a CR were less stringent than in NCI guidelines in that it was not necessary to perform a post-treatment bone marrow biopsy. This study is of great interest since it suggests that chlorambucil is regularly given in less than the best schedule. As shown in the following sections, this more intense schedule performs well against newer agents and is considerably cheaper.

Three trials involving 424 patients have addressed the question of whether adding prednisolone to chlorambucil improves response or survival.[45-47] None of them demonstrated any advantage in adding the corticosteroid, but the trials are too small to know for certain that there is none. Given that better antibacterials and antifungals are now available to us, both of which would tend to diminish steroid-induced toxicity, the question of whether or not to add corticosteroids should be regarded as moot. Current practice in the UK is to use prednisolone if there is evidence of autoimmune disease, and during the first course of treatment if the patient has significant thrombocytopenia.

Cyclophosphamide 50 to 100 mg/day is an acceptable alternative in the few patients who are chlorambucil intolerant. Cyclophosphamide causes more alopecia but less marrow toxicity than chlorambucil.

Within clinical trials, chlorambucil is usually continued until maximal response, but this is seldom CR, and it virtually never eliminates all trace of the disease. In clinical practice outside trials, treatment is often discontinued when the blood count becomes relatively normal and the patient is symptom free. There have been no comparative trials of these practices.

9. Combination Chemotherapy

A study of patients with Stage C disease in France[48] suggested a benefit for patients treated with low dose CHOP (cyclophosphamide 300 mg/m^2, prednisolone 40 mg/m^2 orally for 5 days, vincristine 1 mg/m^2 and doxorubicin 25 mg/m^2 intravenously on day 1). This was a small trial with only 70 patients randomised. However, a number of trials have now randomised patients between treatment with chlorambucil or combination chemotherapy (generally cyclophosphamide, vincristine and prednisolone, with or without an anthracycline). Although responses to regimens containing an anthracycline may be quicker, a meta-analysis of 10 trials involving 2035 patients with stage B and C disease[41] shows that there is no difference in overall survival between the 2 types of treatment. The presence of an anthracycline in the combination does not affect the outcome. Of particular interest among these trials was the International Society for Chemo-Immunotherapy trial comparing the high dose Croatian chlorambucil regimen with the low dose French CHOP regimen.[49] Among 228 randomised patients both the response rate (89.5 *vs* 75%) and the median overall survival (68 *vs* 47 months) was better for the chlorambucil arm.

10. Purine Analogues

Fludarabine, cladribine and pentostatin all have therapeutic effect in CLL. The molecules are similar in structure. Fludarabine is the most frequently used, cladribine less so and pentostatin hardly at all.

10.1 Fludarabine

At dosages of 25 mg/m^2/day given by an intravenous infusion lasting 30 minutes for 5 days every 28 days, fludarabine is the most effective single agent known in CLL. In 12 phase I and phase II studies[50] among 1298 patients with relapsed or refractory disease, a response rate (CR + PR) of 41% was seen and among previously untreated patients the response rate was almost twice as high (79%), with the majority being complete or nodular responses. In the study of 703 patients receiving fludarabine via the Group C mechanism of the NCI,[51] the response rate was only 32% with 3% CRs, but these included many elderly patients with

poor performance scores and extensive prior therapy. Interestingly, in the Medical Research Council (MRC) of the UK CLL3NR trial, patients given salvage fludarabine after failure to respond to either chlorambucil or chlorambucil plus epirubicin, had a response rate of over 80%.[52]

There have been 5 phase III studies comparing fludarabine with other treatments. All demonstrate that it is probably the most effective drug available to treat CLL, but most patients failing other treatments will eventually receive it and as yet there is no survival advantage for those patients receiving it as first-line therapy.

Fludarabine has been compared with CAP (cyclophosphamide 750 mg/m^2, doxorubicin 50 mg/m^2 both intravenously day 1, and prednisolone 40 mg/m^2 orally on days 1 to 5) in a multicentre European study.[53] In previously treated patients the response rates were significantly greater in the fludarabine arm (48 *vs* 27%). However, median remission duration at 324 days for the fludarabine group and 179 days for the CAP group was not significantly different, while the median survivals of the 2 arms of the study were very similar (at 728 and 731 days, respectively). For previously untreated patients, the response rates were not significantly different (71 *vs* 60%). However, fludarabine induced significantly longer remissions in this group. The overall survival of the fludarabine group in this ongoing study also looks better but this has not yet reached statistical significance and, because patients were allowed to cross over, may never do so. Alopecia, nausea and vomiting were significantly more frequent in the CAP arm, but infections were more common in the fludarabine arm.

In a similar French study,[54] fludarabine was compared with either CAP or the French low dose CHOP regimen. Randomisation to the CAP arm was stopped early because of a poorer outcome. Between 1990 and 1998 651 and 287 patients with stage B and stage C disease, respectively, were randomised. Clinical and haematological remission was significantly higher in the fludarabine arm (40 *vs* 30% for CHOP *vs* 15% for CAP), although failure to respond was higher with CAP (41 *vs* 27% with CHOP). Grade 3 or 4 toxicities were similar in each group. Median overall survival was similar in all 3 groups (70 months for CHOP, 73 months for CAP and 74 months for fludarabine).

An American study[55,56] of previously untreated patients randomised more than 500 patients between fludarabine 25 mg/m^2/day for 5 days (n = 179), chlorambucil 40 mg/m^2 on day 1 (n = 193) and the combination of the 2, fludarabine 20 mg/m^2/day for 5 days and chlorambucil 20 mg/m^2 on day 1 (n = 137). Randomisation to the combination arm was stopped before the end of the trial because it was more toxic and had no chance of proving better than the fludarabine arm. The incidence of CR and overall responses was significantly greater for patients treated with fludarabine than those treated with chlorambucil (20 and 64% *vs* 5 and 39%). Median duration of response was 28 months for fludarabine and 19 months for chlorambucil, but overall survival was not significantly different for any of the 3 groups (66 months for fludarabine, 56 months for chlorambucil and 55 months for the combination). Non-responding patients were allowed to cross over to another arm.

An Italian trial[57] randomised 150 previously untreated patients with 'active' CLL between the standard fludarabine regimen and chlorambucil 30 mg/m^2 on days 1 and 15 plus prednisone 40 mg/m^2 intramuscularly on days 1 to 5 and 15 to 19. The overall response rate was 71% in both arms with more CRs with fludarabine (46 *vs* 37%). Toxicity was similar in both arms but response duration was longer for fludarabine (28 *vs* 21 months).

An European Organisation for Research and Treatment of Cancer (EORTC) trial[58] is comparing the Croatian high dose chlorambucil regimen with fludarabine 25 mg/m^2/day every 3 weeks. Overall response rate is slightly higher for chlorambucil (86 *vs* 75%) which also produced more CRs (39 *vs* 36%). It should be noted that non-standard response criteria are used in this trial. The differences are not statistically significant and the trial is too immature for survival data. There has been more haematological toxicity in the chlorambucil group and more infections in the fludarabine group.

Complete remissions obtained with fludarabine are long lasting. Mean time to progression was 21 months for previously treated patients and 33 months for those not previously treated.[59] Second remissions are usually obtainable in those patients who received fludarabine as first-line therapy.[59] Occasionally, molecular remissions can be obtained.[60]

Fludarabine has also been used with other agents, but only the fludarabine and cyclophosphamide combination seems promising with 100% response rates in previously untreated patients and 38% response rates in patients refractory to fludarabine have been reported.[61] The addition of prednisolone to fludarabine does not improve the response rate but does increase the risk of opportunistic infections.[62]

An oral formulation of fludarabine is available with a bioavailability of 55% of the intravenous dose.[63] Early studies suggest that it is equally as effective as the intravenous preparation.[64]

10.2 Cladribine

Cladribine (2-chlorodeoxyadenosine) is converted to its active triphosphate in cells with high levels of deoxycytidine kinase and low 5′-nucleotidase activity.[65] It may be given as a 7-day continuous infusion as a dosage of 0.1 mg/kg/day or as a daily 2-hour infusion of 0.12 mg/kg/day for 5 days. In each case, the dose is repeated every 28 days.[66-68] In 8 phase I/II studies[66-73] involving 458 patients with CLL, the overall response rate was 48% with CRs of 14%. In 4 trials of 103 untreated patients, the response rate was 83% with 32% CR. Remissions in patients achieving CR lasted a median of 42 months and those achieving PR for a median 18 months. It is usual for retreated patients to respond. On the other hand it is very unusual that patients who fail to respond to one purine analogue will respond to another, despite earlier claims to the contrary.[74,75]

There have been no head-to-head comparisons with fludarabine. It is difficult to judge which drug is preferable. Some of the trials of cladribine use non-standard response criteria. There is quite a strong feeling in the US that cladribine is an inferior drug and quite a strong feeling in Europe that it is not. An *in vitro* comparison of the 2 drugs showed them to be similarly effective against CLL cells.[76]

10.3 Pentostatin

Pentostatin (2-deoxycoformycin) is a purine analogue harvested from fermentation cultures of the soil organism *Streptomyces antibioticus*.[77] It is a potent inhibitor of the enzyme adenosine deaminase. This leads to the accumulation of 2-deoxyadenosine and its nucleotides within the cell. 2-Deoxyadenosine is phosphorylated to dATP (adenosine triphosphate), which accumulates in cells and down-regulates ribonuleoside reductase so that DNA replication and repair is impaired.[50]

Although originally introduced for the treatment of HCL, pentostatin is also effective in the treatment of CLL, although apparently less so than fludarabine or cladrabine. In 5 separate studies involving 136 previously treated patients, there were 4 CRs and 32 PRs (overall response rate 26%).[78-82] The usual dosage is 4 mg/m^2 weekly intravenously. It is well tolerated.

10.4 Adverse Effects

Neutropenia occurs in between 30 and 80% of patients, being worse in those who have been previously treated or are unresponsive. Neutrophil counts seldom fall below 0.5×10^9/L. Thrombocytopenia also occurs with all 3 drugs but is seldom severe. Myelotoxicity usually recovers within 3 weeks of treatment, but in a minority of patients is long lasting and severe.[83]

Immunosuppression is a more important adverse effect and it is cumulative. Lymphocytopenia develops rapidly, mainly affecting the CD4+ T cells. Median CD4+ counts declined from 1.015 $\times 10^9$/L to 0.169×10^9/L after 3 treatments.[63] The CD4+-cytopenia is prolonged and may barely have recovered after 2 years. The immunosuppression has important consequences. Opportunistic infections, including *Pneumocystis carinii*,[84] *Listeria monocytogenes*,[85] disseminated candidiasis,[84]

Aspergillus niger,[84] atypical mycobacteria,[84] cytomegalovirus,[86] cryptococcoses,[87] *Oochroconis gallopava*,[88] and astrovirus[89] have all been reported. In particular, *P. carinii* pneumonia is a serious hazard to be guarded against. Low dose cotrimoxazole, 480mg twice a day 3 days a week or 480mg daily 7 days a week, is recommended. These doses have been shown to be adequate in patients with AIDS.[90] Prophylaxis is essential while the CD4+ count remains depressed.

Transfusion related graft-versus-host disease (GVHD) has been observed and it is recommended that all patients treated with purine analogues should receive irradiated blood products.[91] Tumour lysis syndrome is a rare complication of treatment with purine analogues.[92,93]

Although AIHA is a relatively common complication of CLL, and indeed may be triggered by treatment,[94] haemolysis after treatment with the purine analogues is much more common than after other forms of treatment.[95] The first report appeared as a letter in 1992.[96] However, workers from the MD Anderson Cancer Center, Houston, Texas, USA, who had most experience of the new drug, argued that the cases they had seen (9 of 112) represented the natural prevalence of AIHA in CLL.[97] In 4 of 8 patients with pre-existing AIHA, fludarabine was given safely.

Byrd et al.[98] reported a further incidence and stated that the association had been reported to the US Food and Drug Administration on 30 occasions. In 1995, Myint et al.[99] reported that of 52 heavily pretreated patients 12 developed AIHA after between 2 and 6 courses of fludarabine. Since then many reports have confirmed the association and there are now reports of over 100 cases in the literature.[95] Only about 2% of patients treated for the first time develop AIHA, compared with about 5% of patients who have received some previous treatment and over 20% of heavily pretreated patients.[64,97,99-109]

Autoimmune thrombocytopenia may also be triggered by fludarabine. Montillo et al.[110] first reported relapse of CLL-associated immune thrombocytopenia purpura (ITP) after exposure to fludarabine. A total of 25 cases of fludarabine-related ITP have now been reported.[95] Only one possible case of immune neutropenia has been reported[111] and 3 cases of pure red cell aplasia.[97,112,113] Paraneoplastic pemphigus has been reported in 5 patients after fludarabine.[114-116] There have been 2 cases of post-fludarabine glomerulonephritis.[117,118] The other purine analogues, cladribine and pentostatin are also capable of triggering autoimmune complications.[100,103,119-121]

In summary, almost all cases of autoimmunity following treatment with a purine analogue have occurred in patients who have been previously treated with an alkylating agent. The autoimmune disease is often very severe and resistant to treatment. Immunosuppressive treatment to control the autoimmune complication is hazardous because of the risk of infection. After gaining control of the autoimmune disease, re-exposure to any of the purine analogues is likely to lead to recurrence of the autoimmunity in an even more virulent form.[122,123] Recurrence may also be triggered by re-exposure to alkylating agents.[102] Although most cases of autoimmunity following treatment with a purine analogue have been in patients with CLL, it has also been seen in patients with other low grade lymphomas, and especially in Waldenström's macroglobulinaemia.

The autoimmune complications of the purine analogues are probably caused by loss of T cell regulatory control of autoreactive T cells.[99] Autoreactive T cells can readily be identified in the peripheral lymphocyte pool of both humans and mice.[124] In the mouse they can be identified as CD4+, CD25+.[125] Elimination of this population leads to the spontaneous development of autoimmune disease,[124] whereas these cells can also prevent the transfer of disease by cloned autoreactive T cells.[126] They suppress autoreactive T cells by specifically inhibiting the production of interleukin (IL)-2, an action remarkably like that of cyclosporin.[127] This subset of T cells is very susceptible to killing with chemotherapeutic agents compared with the CD4+/CD25 negative subset. In the human, the CD4+/CD45RA+ subset, which is be-

lieved to fulfil the same function, has been shown to be selectively lost in patients with more advanced stages of CLL, especially in those with AIHA.[128]

Autoimmune complications have proved difficult to treat but in most patients cyclosporin 2.5 to 5 mg/kg/day has been effective. It should be continued as long as the risk persists. We aim to keep the blood level at about 100 μg/L. It is not clear whether patients with a history of AIHA or a positive antiglobulin test should be treated with purine analogues. Some authors have reported effective control of both the CLL and the autoimmune complication.[104,129] the author would counsel that such patients should only be treated with these drugs under cover with corticosteroids or preferably cyclosporin.

There is a possibility that second malignancies are commoner in patients treated with purine analogues. In a retrospective study at the NCI there were 111 second malignancies among 2014 patients who had received 1 of the 3 drugs. Although this was slightly (but significantly) higher than the rate of malignant disease in age-matched controls, it was within the excess that might have been expected for patients with CLL or HCL.[130] There have been several reports of myelodysplasia following therapy with purine analogues,[131-134] but even in those patients where the bone marrow changed from non-dysplastic to dysplastic after therapy, it is difficult to apportion blame between the purine analogue and the previous alkylating agent treatment.

11. Monoclonal Antibodies

Despite many attempts to treat CLL with monoclonal antibodies,[135-137] only 2 specificities have shown activity. CD20 is present on the surface of CLL cells at slightly lower densities than in other B cell lymphomas. Responses to anti-CD20 antibody have been disappointing, with <20% of patients benefiting from the standard regimen of 375mg/m^2 intravenously once a week for 4 weeks.[138] Trials with larger doses are continuing, as are studies with radiolabelled antibody.

Campath-1H (anti-CD52) has shown great promise in clearing the tumour from blood and bone marrow. In a study of 29 previously treated patients, the blood was cleared in 97%, with spleen and marrow clearance in about one-third.[139] However, only 7% showed a lymph node response. Among 9 previously untreated patients, 100% showed clearance of tumour cells from the blood, and two-thirds showed clearance from the spleen and bone marrow. Only 37% had a lymph node response. Response duration was between 6 months and over 2 years. The antibody is administered either intravenously or subcutaneously 3 times a week and the major adverse effects are fevers, rigors and rashes related to the infusion. Profound immunosuppression is likely to occur and cases of autoimmunity have been reported. The use of anti-CD52 therapy to clear residual disease from bone marrow after a fludarabine-induced partial remission and before a stem-cell harvest is becoming increasingly popular.

12. Transplantation

Although it is only applicable to a small minority of young patients, allogeneic bone marrow transplantation offers the best chance for cure of CLL. The combined registries of the European Group for Blood and Marrow Transplant (EBMT) and International Bone Marrow Registry (IBMR) reported 70 allogeneic bone marrow transplants in CLL between 1984 and 1995.[140] Conventional transplant regimens were used. Only 3% were in CR before transplantation. Engraftment occurred in all except 2 patients and CR was achieved in 75%. Actuarial survival was 48% at 3 years.

Data from the EBMT have now been extended.[141] 188 patients have now received allografts, 63% under the age of 45. 25% were in CR before transplant and 85% after. 39% developed grade 2-4 acute GVHD and 20% chronic GVHD. Treatment-related mortality was 49%. The probability of survival at 3 years was 46%, and the probability of relapse 14%. The same group has reported its experience with autografts.[141] Of 225 patients three-quarters received stem cells from pe-

ripheral blood rather than marrow. 51% were in CR before treatment and 85% after treatment. Treatment-related mortality was 14%. The probability of survival at 3 years was 78% and the probability of relapse 45%.

A group in Boston demonstrated the importance of eliminating minimal residual disease.[142] They treated 41 patients with CLL, apparently in CR, with high dose chemoradiotherapy followed by either a T-cell–depleted matched sibling allogeneic bone marrow transplant (13 patients) or a monoclonal antibody purged marrow autograft (28 patients). Four patients died of infection-related GVHD and 2 died of autologous transplant-related complications. 31 patients had molecularly detectable disease at the time of transplant and all except 5 eventually became negative. Only these 5 relapsed. The actuarial disease-free survival for the whole group was 77% at 19 months.

The experience of the Houston group stresses the hazards of further immunosuppressive therapy in patients who have already been treated with fludarabine. They reported 22 patients with advanced CLL receiving transplants.[143] 11 received syngeneic or allogeneic grafts and 11 autografts. Of the first group, 7 achieved CR, 2 a nodular CR and 1 a PR. Ten were alive 2 to 36 months after the transplant. Greater toxicity occurred in those receiving autologous marrow. Although 6 patients achieved CR, 4 a nodular CR and 1 a PR, 2 patients relapsed with CLL and 3 with a Richter's transformation. Two developed immune cytopenias while in morphological remission and 1 of these died of cytomegalovirus pneumonia. Six of 11 remained in CR 2 to 29 months after transplantation. Notably, the patients with allografts received cyclosporin while those with autografts did not.

An MRC pilot study has examined the feasibility of a stem-cell autograft programme in patients treated with 3 to 6 cycles of fludarabine.[144] Fludarabine may inhibit the generation of stem cells. 99 patients <60 years of age and early in their disease were entered; 2 were withdrawn after being reclassified as having MCL. 78 completed fludarabine therapy; 42 achieved a CR or PRn. One died of transfusion-related GVHD. Some of the patients failing to achieve sufficient response had further treatment, and eventually attempts were made to harvest stem cells in 50 of them. 45 have been autografted, but only 14 had sufficient cells for the planned CD34+ cell selection. There has been 1 transplant-related death (due to transverse myelitis). Of those tested, 17 of 22 achieved a molecular remission. 3 of these 17 have had a molecular relapse, and 1 has died from secondary myelodysplastic syndrome. Although the trial is immature, the outcome so far suggests that the adage 'many are called but few are chosen' might apply.

Because of the high treatment-related mortality of bone marrow allograft in CLL, attempts have been made to evaluate 'mini-allografts'. These avoid myelo-ablation but rely on conditioning with either fludarabine or Campath-1H to ensure engraftment and on a graft-versus-leukaemia effect to achieve remissions. Donor lymphocyte infusions may also be used. Early results from the MD Anderson Cancer Center[145] suggest that this is as yet a hazardous procedure, with only 3 patients alive and in remission of the first 8 treated. Many other groups are experimenting with this technique which offers the chance of extending bone marrow transplantation into the age range in which CLL usually occurs.

13. Experimental Agents

A range of new agents currently under investigation in CLL has been reviewed by Byrd et al.[146] UCN-01 is a hydroxylated derivative of staurosporine, believed to abrogate the checkpoints of cell division. It is able to induce apoptosis independently of p53. Preliminary data suggest that it might sensitise CLL cells to the effect of fludarabine, and clinical trials are continuing.[147]

Several topoisomerase-I inhibitors are under investigation. Although topotecan seems to have no useful activity in CLL,[148] a second study using IDEC-132 (9-aminocamptothecin), an agent less susceptible to multiple drug resistance, is under way.

Bryostatin, a protein kinase C activator, alters the morphology of CLL cells to resemble HCL. On this basis, it has been used as a preliminary to treatment with agents effective in HCL.[146]

Flavopiridol is a synthetic flavone that antagonises several cell cycle proteins and is toxic to CLL cells *in vitro*. Again, it induces apoptosis independently of p53.[146] Phase II trials in CLL are currently taking place.

Nelarabine or compound 506-U78, a methoxy derivative of guanine arabinoside, has undergone phase I testing at the MD Anderson Cancer Center.[149] Responses of 31% were seen in 16 elderly patients with advanced refractory CLL.

14. Immunotherapy

Kipps et al.[150] transduced CLL cells with an adenovirus vector encoding CD40 ligand. Such cells became highly effective stimulators of autologous T cells that could generate CLL specific cytotoxic T cells *in vitro*. In a phase I trial patients were infused with increasing amounts of transduced autologous CLL cells. Toxic effects were minor, but bystander leukaemia cells were induced to upregulate CD80 and CD86, there were 2- to 3-fold increases in circulating T cells and 20 to 70% reductions in absolute lymphocyte counts with 33 to 90% reductions in lymph node size.

A phase II trial of DNA vaccination for CLL is taking place in the UK.[151] Idiotypic determinants encoded by Ig heavy and light chains are assembled as single chain Fv fused to the gene for fragment C of tetanus toxin in a plasmid vector. The vaccine is protective against both B cell lymphoma and myeloma in mice.

15. Radiotherapy

There is a limited role for radiotherapy in CLL. Some groups have used total body irradiation for transplant conditioning. In the rare situation of patients with retro-orbital or cerebral deposits, localised radiotherapy is useful, and sometimes large and unsightly nodal enlargement is better treated by local radiotherapy to spare the use of systemic chemotherapy.

16. Splenectomy

Removal of the spleen is indicated in patients with hypersplenism and often in those with immune thrombocytopenia. Even in elderly and frail patients, it may be possible to remove the spleen with little trauma laparoscopically. Splenic irradiation should be considered in patients too frail even for this procedure.

17. Supportive Care

All patients surviving long enough will eventually develop hypogammaglobulinaemia.[2] Nevertheless, prophylactic immunoglobulin is neither beneficial nor cost effective except in patients with recurrent bacterial infections.[152-154] Prophylactic antibacterials are not indicated except for cotrimoxazole prophylaxis against *P. carinii* in patients who have received a purine analogue.[155]

18. Conclusion: A Strategy for Treatment

At present there are no indications to treat patients with stage A disease unless it is progressive (fig. 1). Such patients should be observed at a frequency determined by the presence or absence of adverse prognostic factors. If any of the prognostic indicators were shown to unerringly select stage A patients destined to progress then there would be a case for treating such patients early, and if they were younger, to treat with curative intent.

For patients who require treatment there is no certain recommendation. For many years chlorambucil has been the standard treatment and it has certainly not been supplanted by anthracycline-containing combinations. In many respects fludarabine is an attractive alternative. It is probably more active than chlorambucil and produces more complete remissions with longer disease-free survivals. On the other hand it does not cure CLL, it has not been shown to increase overall survival and it has to be given intravenously, although an oral form is in development. There is a high response rate in relapsed and refractory CLL so that it is a

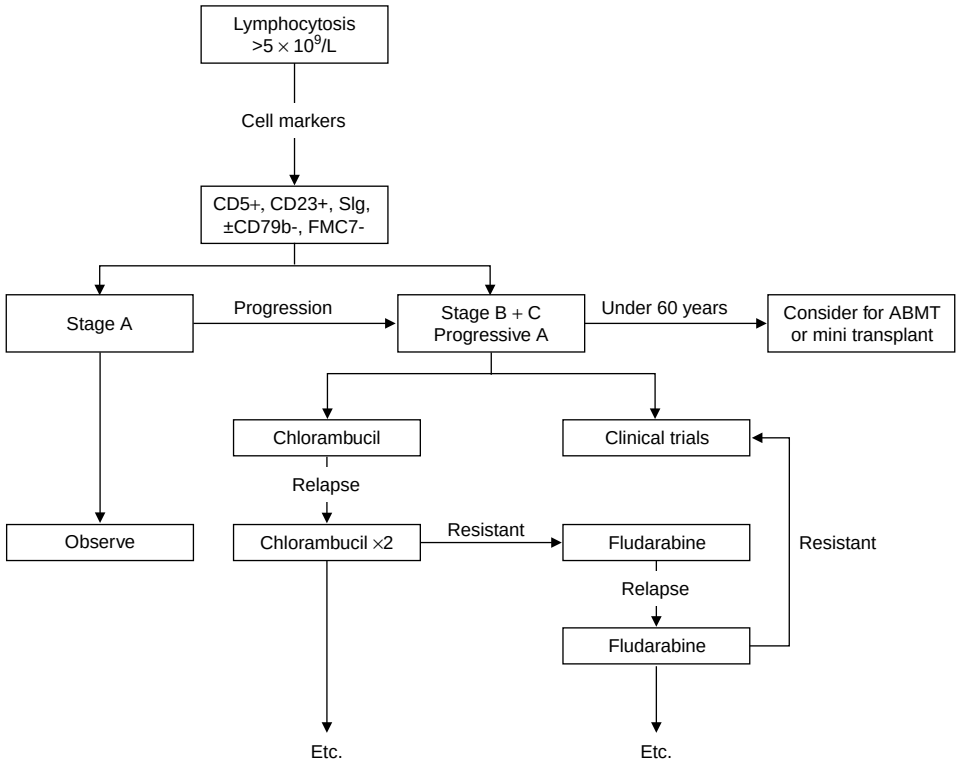

Fig. 1. Flow chart for treatment recommendations in patients with chronic lymphocytic leukaemia. **ABMT** = autologous bone marrow transplant; **SIg** = surface immunoglobulin.

useful second-line treatment. The argument is over whether it should be used as first-line.

Cheson[156] has advocated using our best drug first. He stresses that it is much more effective when given first-line. He concedes that there is a financial disincentive but argues that since the patient will eventually receive fludarabine, it does not matter whether the bill comes earlier or later. It might also be argued that fludarabine given late in the disease is far more likely to trigger autoimmune and infectious complications, either of which might have daunting financial consequences.

The cost difference between the 2 drugs is substantial. In the UK, a month's fludarabine for a patient of 1.7m^2 costs £552.50 compared with £12.45 for chlorambucil (British National Formulary, September 2000). When the cost of intravenous administration and co-trimoxazole prophylaxis is added the difference is greater. If every patient who received chlorambucil were destined eventually to receive fludarabine then the cost might be justified, but there is no evidence in real life of what proportion of chlorambucil-treated patients would opt for subsequent fludarabine treatment.

Until a clear survival advantage can be demonstrated for first-line fludarabine, patients requiring treatment should preferably be entered into clinical trials. The high dose Croatian chlorambucil regimen still requires further evaluation against fludarabine. All such trials should evaluate quality of life and cost-benefit. For patients not entered into trials, intermittent chlorambucil remains the treatment of choice with fludarabine or cladribine reserved for unresponsive disease.

When a patient relapses, re-treatment with chlorambucil should be offered. Most patients will respond again and this process may be repeated on several occasions. Chlorambucil treatment will probably eventually cause bone marrow damage and a judgement must be made when to switch to fludarabine. There is no clinical trial evidence to help this decision. Fludarabine may also be offered as many times as the patient tolerates it and their disease responds to it.

A number of options are available for refractory disease. The combination of fludarabine and cyclophosphamide, CHOP, high dose 'Croatian' chlorambucil, anti-CD52, anti-CD20 and a range of experimental drugs are available; they should best be used in clinical trials. One alternative is to be guided by *in vitro* chemosensitivity testing.[43] There is a dearth of comparative clinical trials for end-stage, drug-resistant CLL, although the MRC CLL4 trial is comparing treatment determined by the DiSC assay with treatment determined by physicians' best guess.

Younger patients should be given the opportunity of entering trials of treatment with curative intent. Stem-cell autograft has not been demonstrated to give a survival advantage and should be compared with conventional treatment in phase III trials. Allogeneic stem-cell transplantation is a hazardous procedure with a high treatment-related mortality. Transplants with non-ablative conditioning regimens appear promising but need further evaluation in clinical trials. Some patients will be entered into trials of experimental immunotherapy.

Acknowledgements

Professor Hamblin is supported by Tenovus.

References

1. Cartwright RA, Alexander FE, McKinney PA, et al. Leukaemia and lymphoma. An atlas of distribution within areas of England and Wales 1984-8. London: Leukaemia Research Fund, 1990
2. Hamblin TJ. Chronic lymphocytic leukaemia. Balliere's Clin Haematol 1987; 1: 449-91
3. Cheson BD, Bennett JM, Grever M, et al. National Cancer Institute Sponsored Working Group guidelines for chronic lymphocytic leukemia: revised guidelines for diagnosis and treatment. Blood 1996; 87: 4990-7
4. Moreau EJ, Matutes E, A'Hern RP, et al. Improvement of the chronic lymphocytic leukemia scoring system with the monoclonal antibody SN8 (CD79b). Am J Clin Pathol 1997; 108: 378-82
5. Matutes E, Owusu-Ankomah K, Morilla R, et al. The immunological profile of B-cell disorders and proposal of a scoring system for the diagnosis of CLL. Leukemia 1994; 8: 1640-5
6. Campo E, Raffeld M, Jaffe ES. Mantle-cell lymphoma. Semin Hematol 1999; 36: 115-27
7. Laszlo T, Nagy M, Kelenyi G, et al. Immunoglobulin V(H) gene mutational analysis suggests that blastic variant of mantle cell lymphoma derives from different stages of B-cell maturation. Leuk Res 2000; 4: 27-31
8. Cuneo A, Bigoni R, Negrini M, et al. Cytogenetic and interphase cytogenetic characterization of atypical chronic lymphocytic leukemia carrying BCL1 translocation. Cancer Res 1997; 57: 1144-50
9. Catovsky D, Matutes E. Splenic lymphoma with circulating villous lymphocytes/splenic marginal-zone lymphoma. Semin Hematol 1999; 36: 148-54
10. Stevenson F, Sahota S, Zhu D, et al. Insight into the origin and clonal history of B-cell tumors as revealed by analysis of immunoglobulin variable region genes. Immunol Rev 1998; 162: 247-59
11. Mollejo M, Lloret E, Solares J, et al. Splenic involvement by blastic mantle cell lymphoma (large cell/anaplastic variant) mimicking splenic marginal zone lymphoma. Am J Hematol 1999; 62: 242-6
12. Rai KR, Sawitsky A, Cronkite ER, et al. Clinical staging of chronic lymphocytic leukemia. Blood 1977; 46: 219-34
13. Binet J-L, Leporier M, Dighiero G, et al. A clinical staging system for chronic lymphocytic leukemia. Cancer 1977; 40: 855-64
14. Montserrat E, Vinolas E, Reverter JC, et al. Natural history of chronic lymphocytic leukemia; on the progression and prognosis of early clinical stages. Nouv Rev Fr Hematol 1988; 30: 359-61
15. Montserrat E, Sanchez-Bisono J, Vinolus N, et al. Lymphocyte doubling time in chronic lymphocytic leukaemia: analysis of prognostic significance. Br J Haematol 1986; 62: 567-75
16. Rozman C, Hernandez-Nieto L, Montserrat E, et al. Prognostic significance of bone marrow patterns in chronic lymphocytic leukaemia. Br J Haematol 1981; 47: 529-37
17. Oscier DG, Matutes E, Copplestone A, et al. Prognostic factors in stage A chronic lymphocytic leukaemia; the importance of atypical lymphocyte morphology and abnormal karyotype for disease progression in stage A CLL. Brit J Haem 1997; 98: 934-9
18. Safati M, Chevret S, Chastang C, et al. Prognostic importance of serum soluble CD23 level in chronic lymphocytic leukemia. Blood 1996; 88: 4259-64
19. Molica S, Levato D, Cascavilla N, et al. Clinico-prognostic implications of simultaneous increased serum levels of soluble CD23 and beta2-microglobulin in B-cell chronic lymphocytic leukemia. Eur J Haematol 1999; 62: 117-22
20. Juliusson G, Oscier DG, Fitchett M, et al. Prognostic subgroups in B-cell chronic lymphocytic leukemia defined by specific chromosomal abnormalities. N Engl J Med 1990; 323: 720-4
21. Zupo S, Isnardi L, Megna M, et al. CD38 expression distinguishes two groups of B-cell chronic lymphocytic leukemias with different responses to anti-IgM antibodies and propensity to apoptosis. Blood 1996; 88: 1365-74

22. International Workshop on CLL. Prognostic factors in early chronic lymphocytic leukaemia. Lancet 1989; II: 968-9
23. Montserrat E, Rozman C. Chronic lymphocytic leukaemia: prognostic factors and natural history. Balliere's Clin Haematol 1993; 4: 849-66
24. Hamblin TJ, Davis Z, Gardiner A, et al. Unmutated Ig V_H genes are associated with a more aggressive form of chronic lymphocytic leukemia. Blood 1999; 94: 1848-54
25. Damle RN, Wasil T, Fais F, et al. Ig V gene mutation status and CD38 expression as novel prognostic indicators in chronic lymphocytic leukemia. Blood 1999; 94: 1840-7
26. Hamblin TJ, Orchard JA, Davis Z, et al. Immunoglobulin V genes and CD38 expression in CLL. Blood 2000; 95: 2455-7
27. Hamblin TJ, Oscier DG, Young BJ. Autoimmunity in chronic lymphocytic leukaemia. J Clin Pathol 1986; 39: 713-6
28. Diehl LF, Ketchum LH. Autoimmune disease and chronic lymphocytic leukemia: autoimmune hemolytic anemia, pure red cell aplasia and autoimmune thrombocytopenia. Sem Hematol 1998; 25: 80-97
29. Chikkappa G, Zarrabi MH, Tsan MF. Pure red cell aplasia in patients with chronic lymphocytic leukemia. Medicine (Baltimore) 1986; 65: 339-51
30. Anhalt GJ, Kim SC, Stanley JR, et al. Paraneoplastic pemphigus. An autoimmune mucocutaneous disease associated with neoplasia. N Engl J Med 1990; 323: 1729-35
31. Chevailler A, Arlaud G, Ponard D, et al. C-1-inhibitor binding monoclonal immunoglobulins in three patients with acquired angioneurotic edema. J Allergy Clin Immunol 1996; 97: 998-1008
32. Dathan JRE, Heyworth MF, MacIver AG. Nephtotic syndrome in chronic lymphocytic leukaemia. BMJ 1974; 3: 655-7
33. Melo JV, Catovsky D, Galton DAG. The relationship between chronic lymphocytic leukaemia and prolymphocytic leukaemia. I. Clinical and laboratory features of 300 patients and characterisation of an intermediate group. Br J Haematol 1986; 63: 377-87
34. Matutes E, Oscier D, Garcia-Marco J, et al. Trisomy 12 defines a group of CLL with atypical morphology: correlation between cytogenetic, clinical and laboratory features in 544 patients. Br J Haematol 1996; 92: 382-8
35. Brouet JC, Preud'homme JL, Seligmann M, et al. Blast cells with surface immunoglobulin in two cases of acute blast crisis supervening on chronic lymphocytic leukaemia. BMJ 1973; 4: 23-4
36. Richter N. Generalised reticular sarcoma of lymph nodes associated with lymphatic leukemia. Amer J Pathol 1928; 4: 285-99
37. Miyamura K, Osada H, Yamaguchi T, et al. Single clonal origin of neoplastic B cells with different immunoglobulin light chains in a patient with Richter's syndrome. Cancer 1990; 86: 140-4
38. Tohda S, Morio T, Suzuki T, et al. Richter's syndrome with two B cell clones possessing different surface immunoglobulins and immunoglobulin gene rearrangements. Am J Hematol 1990; 35: 32-6
39. Ansell SM, Li CY, Lloyd RV, et al. Epstein-Barr virus infection in Richter's transformation. Am J Hematol 1999; 60: 99-104
40. Cheson BD, Bennett JM, Rai KR, et al. Guidelines for clinical protocols for chronic lymphocytic leukemia: recommendations of the National Cancer Institute – sponsored working group. Am J Hematol 1988; 29: 152-63
41. CLL Trialists' Collaborative Group. Chemotherapeutic options in chronic lymphocytic leukaemia: a meta-analysis of the randomised trials. J Natl Cancer Inst 1999; 91: 861-8
42. Dighiero G, Maloum K, Desablens B, et al. Chlorambucil in indolent chronic lymphocytic leukaemia. N Engl J Med 1998; 338: 1506-14
43. Bosanquet AG, McCann SR, Crotty GM, et al. Methylprednisolone in advanced chronic lymphocytic leukaemia: rationale for, and effectiveness of treatment suggested by DiSC essay. Acta Haematol 1995; 93: 73-9
44. Jacksic B, Brugiatelli H. High dose continuous chlorambucil vs intermittent chlorambucil plus prednisolone for treatment of B CLL: IGCI CLL-01 trial. Nouv Rev Fr Hematol 1988; 30: 437-42
45. Catovsky D, Richards S, Fooks J, et al. CLL trials in the United Kingdom. MRC CLL trials 1,2 and 3. Leuk Lymphoma 1991; 5 Suppl. 4: 105-12
46. Spanish Cooperative Group Pethema. Treatment of chronic lymphocytic leukaemia: a preliminary report of Spanish (Pethema) trials. Leuk Lymphoma 1991; 5 Suppl. 4: 89-91
47. Han T, Ezdinli EZ, Shimaoka K, et al. Chlorambucil vs. Combined chlorambucil-corticosteroid therapy in chronic lymphocytic leukemia. Cancer 1973; 31: 502-8
48. French Co-operative Group on Chronic Lymphocytic Leukemia. Benefit of the CHOP regimen in advanced untreated chronic lymphocytic leukemia. Results of a randomized clinical trial. Lancet 1986; I: 346-9
49. Jaksic B, Brugiatelli M, Krc I, et al. High dose chlorambucil versus Bonet's modified cyclophosphamide, doxorubicin, vincristine, and prednisone regimen in the treatment of patients with advanced B-cell chronic lymphocytic leukemia. Results of an international multicenter randomized trial. International Society for Chemo-Immunotherapy, Vienna. Cancer 1997; 79: 2107-14
50. Pott-Hoeck J, Hidderman W. Purine analogues in the treatment of low grade lymphomas and chronic lymphocytic leukemia. Ann Oncol 1995; 6: 421-33
51. Sorensen JM, Vena D, Fallavollitta A, et al. Treatment of refractory chronic lymphocytic leukemia with fludarabine phosphate via the Group C mechanism of the National Cancer Institute. J Clin Oncol 1997; 15: 458-65
52. Catovsky D, Hamblin TJ, Richards S. UK Medical Research Council - CLL3: a randomised trial of the addition of epirumbicin to standard chlorambucil treatment [abstract]. Blood 2000; 96 Suppl. 1: 3263
53. French Co-operative Group on Chronic Lymphocytic Leukemia, Johnson S, Smith AG, et al. Multicentre prospective randomised trial of fludarabine versus cyclophosphamide, doxorubicin, and prednisone (CAP) for treatment of advanced stage chronic lymphocytic leukaemia. Lancet 1996; 347: 1432-38
54. Leporrier M, Chevret S, Cazin B, et al. Randomized clinical trial comparing two anthracycline-containing regimens (ChOP and CAP) with fludarabine in advanced CLL [abstract P101]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
55. Rai KR, Peterson B, Elias L, et al. A randomized comparison of fludarabine and chlorambucil for patients with previously untreated chronic lymphocytic leukemia: a CALGB, SWOG, CTC/NCI-C and ECOG Intergroup Study [abstract]. Blood 1996; 88: 141a
56. Rai KR, Gupta N. International randomized trials of chemotherapy for CLL [abstract S12]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
57. Spriano M, Chiurazzi F, Liso V, et al. Multicentre prospective randomized trial of fludarabine versus chlorambucil and prednisone in previously untreated patients with active B-CLL.

[abstract P086]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
58. Jaksic B, Brugiatelli M, Suciu S, et al. Fludarabine versus high dose continuous chlorambucil in untreated patients with B-CLL: results of CLL1 EORTC randomized trial [abstract P096]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
59. Keating MR, O'Brien S, Kantarjian H, et al. Long term follow up of patients with chronic lymphocytic leukemia treated with fludarabine as a single agent. Blood 1993; 81: 2878-84
60. Cull G, Richardson DS, Howe DJ, et al. Molecular complete response in a patient with chronic lymphocytic leukaemia treated with 2-chlorodeoxyadenosine. Acta Oncol 1995; 34: 536-7
61. O'Brien S, Kantarjian H, Beran M, et al. Fludarabine and cyclophosphamide therapy in chronic lymphocytic leukemia. Blood 1996; 88: 480a
62. O'Brien S, Kantarjian H, Beran M, et al. Results of fludarabine and prednisolone therapy in 264 patients with chronic lymphocytic leukemia with multi-variate analysis-derived prognostic model for response to treatment. Blood 1993; 82: 1695-700
63. Foran JM, Oscier DG, Orchard J, et al. Pharmacokinetic study of single doses of oral fludarabine phosphate. J Clin Oncol 1999; 17: 1574-9
64. Boogaerts MA, Van Hoof A, Catovsky D, et al. Treatment of alkylator resistant CLL with oral fludarabine phosphate [abstract P113]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
65. Kawasaki H, Camera CJ, Piro ID, et al. Relationship of deoxycytidine kinase and cytoplasmic 5' nucleotidase to the chemotherapeutic efficacy of 2-chlorodeoxyadenosine. Blood 1993; 81: 597-601
66. Saven A, Carrera CJ, Carson DA, et al. Chlorodeoxyadenosine treatment of refractory chronic lymphocytic leukemia. Leuk Lymphoma 1991; 5: 133-8
67. Juliusson G, Liliemark J. High complete remission rate from 2-chloro-2'-deoxyadenosine in previously treated patients with B-cell chronic lymphocytic leukemia: response predicted by rapid decrease in blood lymphocyte count. J Clin Oncol 1993; 11: 679-89
68. Robak T, Blasinka-Morawiec M, Krykowski E, et al. Intermittent 2-hour intravenous infusion of 2-chlorodeoxyadenosine in treatment of 110 patients with refractory or previously untreated B-ce; chronic lymphocytic leukemia. Leuk Lymphoma 1996; 22: 509-14
69. Tallman MS, Hakiman D, Zanzig C, et al. Cladribine in the treatment of relapsing or refractory chronic lymphocytic leukemia. J Clin Oncol 1995; 13: 983-8
70. Delannoy A, Martiat P, Gala SL, et al. 2-chlorodeoxyadenosine for patients with previously untreated chronic lymphocytic leukemia (CLL). Leukemia 1995; 9: 1130-5
71. Rondelli D, Lauria F, Zinzani PL, et al. 2-Chlorodeoxyadenosine in the treatemnt of relapsed/refractory chronic lymphoproliferative disorders. Eur J Haematol 1997; 58: 46-50
72. Robak T, Blonski JZ, Urbamska-Rys H, et al. 2-chlorodeoxyadenosine (cladribine) in the treatment of patients with chronic lymphocytic leukemia 55 years old and younger. Leukemia 1999; 13: 518-23
73. Robak T, Blasinska-Morawiec M, Blonski JZ, et al. 2-chlorodeoxyadenosine (cladribine) in the treatment of elderly patients with B-cell chronic lymphocytic leukemia. Leuk Lymphoma 1999; 34: 151-7
74. Juliusson G, Elmhorn-Rosenberg A, Liliemark J. Response to 2-chlorodeoxyadenosine in patients with chronic lymphocytic leukemia resistant to fludarabine. N Engl J Med 1992; 327: 1056-61
75. O'Brien S, Kantarjian H, Estey E, et al. Lack of effect of 2-chlorodeoxyadenosine in patients with chronic lymphocytic leukemia refractory to fludarabine. N Engl J Med 1994; 330: 319-22
76. Koski T, Vilpo L, Vilpo J. Chemosensitivity in vitro to 2-chlorodeoxyadenosine and 9-beta-D-arabinofuranosyl-2-fluoroadenine in previously unexposed cases of chronic lymphocytic leukemia. Leuk Res 1999; 23: 277-9
77. Kane BJ, Kuhn JG, Roush MK. Pentostatin: an adenosine deaminase inhibitor for the treatment of hairy cell leukemia. Ann Pharmacother 1992; 26: 939-47
78. Grever MR, Leiby JM, Kraut EH. Low dose deoxycoformycin in lymphoid malignancy. J Clin Oncol 1985; 3: 1196-201
79. O'Dwyer P, Wagner B, Leyland-Jones B, et al. 2-Deoxycoformycin (Pentostatin) for lymphoid malignancies. Ann Int Med 1988; 108: 733-43
80. Dillman RO, Mick R, McIntyre OR. Pentostatin in chronic lymphocytic leukemia. A phase II trial of cancer and leukemia group B. J Clin Oncol 1989; 7: 433-8
81. Ho AD, Thaler J, Strykmans P, et al. Pentostatin in refractory CLL: a phase II trial of the EORTC. J Natl Cancer Inst 1990; 82: 1416-20
82. Dearden C, Catovsky D. Deoxycoformycin in the treatment of mature B-cell malignancies. Br J Cancer 1990; 62: 4-5
83. Cheson B. Infectious and immunosuppresive complications of purine analogue therapy. J Clin Oncol 1995; 13: 2431-48
84. Byrd JC, Hargis JB, Kester KE, et al. Opportunistic infections with fludaribine in previously treated patients with low grade lymphoid malignancies: a role for pneumocystis carinii pneumonia prophylaxis. Amer J Hematol 1995; 49: 135-42
85. Spielberger RT, Stock W, Larson RA. Listeriosis after 2-chlorodeoxyadenosine treatment. N Engl J Med 1993; 328: 813-4
86. Schilling PJ, Vadham-Raj S. Concurrent cytomegalovirus and pneumocystis pneumonia after fludarabine therapy for chronic lymphocytic leukemia. N Engl J Med 1990; 323: 833-4
87. Leenders A, Sonnenveld P, de Marie S. Cryptococcal meningitis following fludarabine treatment for chronic lymphocytic leukemia. Eur J Microbiol Infest Dis 1995; 14: 826-8
88. Bowyer JD, Johnson EM, Horn EH, et al. Oochroconis gallopava endophthalmitis in fludarabine treated chronic lymphocytic leukaemia. Br J Ophthalmol 2000; 84: 117
89. Coppo P, Scieux C, Ferchal F, et al. Astrovirus enteritis in a chronic lymphocytic leukemia patient treated with fludarabine monophosphate. Ann Hematol 2000; 79: 43-5
90. Ioannidis JP, Cappelleri JC, Skolnik PR, et al. A meta-analysis of the relative efficacy and toxicity of *Pneumocystis carinii* prophylactic regimens. Archives of Internal Medicine 1996; 156: 177-88
91. Maung ZT, Wood AC, Jackson GH, et al. Transfusion associated graft versus host disease in fludarabine treated B chronic lymphocytic leukemia. Br J Haematol 1994; 88: 649-52
92. Cheson BD, Frame JN, Vena D, et al. Tumor lysis syndrome: an uncomon complication of fludarabine therapy of chronic lymphocytic leukemia. J Clin Oncol 1998; 16: 2313-20
93. List AF, Kummet TD, Adams JD, et al. Tumor lysis syndrome complicating treatment of chronic lymphocytic leukemia with fludarabine phosphate. Am J Med 1990; 89: 388-90
94. Lewis FB, Schwarz RS, Damashek W. X-irradiation and alkylating agents as possible trigger mechanisms in autoimmune complications of malignant lymphoproliferative diseases. Clin Exper Immunol 1966; 1: 3-11

95. Hamblin TJ. Autoimmune disease and its management in CLL. In: Cheson BD, editor. Chronic lymphocytic leukemia: scientific advances and clinical developments. New York: Marcel Dekker. 2nd edition. In press
96. Bastion Y, Coiffier B, Dumontet C, et al. Severe autoimmune hemolytic anemia in two patients treated with fludarabine for chronic lymphocytic leukemia. Ann Oncol 1992; 3: 171-2
97. Di Raimondo F, Guistolisi R, Caccioa E, et al. Autoimmune hemolytic anemia in chronic lymphocytic leukemia patients treated with fludarabine. Leuk Lymphoma 1993; 1: 63-8
98. Byrd JC, Weiss RB, Kweeder SL, et al. Fludarabine therapy with lymphoid malignancies is associated autoimmune hemolytic anemia. [abstract no. 304a]. American Society of Clinical Oncology 30th Annual Meeting; 1994 May 14-17; Dallas
99. Myint H, Copplestone JA, Orchard J, et al. Fludarabine-related autoimmune haemolytic anaemia in patients with chronic lymphocytic leukaemia. Br J Haematol 1995; 91: 341-4
100. Montillo M, Tedeschi A, Delfini C, et al. Effectiveness of fludarabine in advanced β-cell chronic lymphocytic leukaemia. Tumori 1995; 81: 419-23
101. Gonzalez H, Leblond V, Azar N, et al. Severe autoimmune hemolytic anemia in eight patients treated with fludarabine. Hematol Cell Ther 1998; 40: 113-8
102. Orchard J. Bolam S, Myint H, et al. In patients with lymphoid tumours recovering from the autoimmune complications of fludarabine, relapse may be triggered by conventional chemotherapy. Br J Haematol 1998; 102: 1112-3
103. Hamblin TJ, Orchard JA, Myint H, et al. Fludarabine and hemolytic anemia in chronic lymphocytic leukemia. J Clin Oncol 1998; 16: 3209
104. Montillo M, Tedeschi A, O'Brien S, et al. Autoimmune phenomena against hematopoietic cells and myelosuppression in CLL treated with fludarabine-including regimens as frontline therapy [abstract P091]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
105. Spriano M, Clavio M, Ballerini F, et al. Fludarabine plus cyclophosphamide in the treatment of previously treated CLL [abstract p114]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
106. Aguirre R, Anselmo A, Basso A, et al. A multicentre trial in CLL with fludarabine: third interim analysis [abstract P135]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
107. Fenaux P, Binet JL, Leporrier M, et al. Multicentre, prospective, randomised trial of fludarabine versus cyclophosphamide, dovorubium, and prednisolone (CAP) for treatment of advanced stage chronic lymphocytic leukaemia. Lancet 1996; 347: 1432-8
108. Leporrier M, Chevret S, Feugier P, et al. Randomized comparison of fludarabine. Early stopping of CAP accrual [abstract 2357]. Blood 1997; 90 Suppl. 1: 529
109. Mauro FR, Mandelli F, Foa R, et al. Autoimmune hemolytic anemia in chronic lymphocytic leukemia (CLL): a retrospective study of 55 cases [abstract 4137]. Blood 1997; 90 Suppl. 1: 308
110. Montillo M, Tedeschi A, Leoni P. Recurrence of autoimmune thrombocytopenia after treatment with fludarabine in a patient with chronic lymphocytic leukemia. Leuk Lymphoma 1994; 15: 187-8
111. Stern SC, Shah S, Costello C. Probable autoimmune neutropenia induced by fludarabine treatment for chronic lymphocytic leukaemia. Br J Haematol 1999; 106: 836-7
112. Antich Rojas J, Balaguer H, Cladera A. Selective aplasia of the red-cell series after fludarabine administration in a patient with chronic B-cell lymphatic leukemia. Sangre (Barc) 1997; 42: 254-6
113. Leporier M, Reman O, Troussard X. Pure red cell aplasia with fludarabine for chronic lymphocytic leukemia. Lancet 1993; 342: 555
114. Bazarbachi A, Bachelez H, Dehen L, et al. Lethal paraneoplastic pemphigus following treatment of chronic lymphocytic leukemia with fludarabine. Ann Oncol 1995; 6: 730-1
115. Braess J, Reich K, Willert S, et al. Mucocutaneous autoimmune syndrome following fludarabine therapy for low-grade non-Hodgkin's lymphoma of B-cell type (B-NHL). Ann Hematol 1997; 75: 227-30
116. Littlewood TJ, Gooplu C, Lyon CC, et al. Paraneoplastic pemhigus – an association with fludarabine [abstract 4207]. Blood 1998; 92: Suppl. 1: 280
117. Macheta MP, Parapia LA, Gouldesbrough DR Renal failure in a patient with chronic lymphocytic leukaemia treated with fludarabine. J Clin Pathol 1995; 48: 181-2
118. Tisler A, Pierratos A, Lipton JH. Crescentic glomerulonephritis associated with p-ANCA positivity in fludarabine-treated chronic lymphocytic leukaemia. Nephrol Dial Transplant 1996; 11: 2306-8
119. Fleischman RA, Croy D. Acute onset of severe autoimmune hemolytic anemia after treatment with 2-chlorodeoxyadenosine for chronic lymphocytic leukemia. Am J Hematol 1995; 48: 293
120. Chasty RC, Myint H, Oscier DG, et al. Autoimmune haemolysis in patients with B-CLL treated with chlorodeoxyadenosine (CDA). Leuk Lymphoma 1998; 29: 391-8
121. Robak T, Blasinska-Morawiec M, Krykowski E, et al. Autoimmune haemolytic anaemia in patients with chronic lymphocytic leukaemia treated with 2-chlorodeoxyadenosine (cladribine). Eur J Haematol 1997; 58: 109-13
122. Weiss RB, Freiman J, Kweder SL, et al. Hemolytic anemia after fludarabine therapy for chronic lymphocytic leukemia. J Clin Oncol 1998; 16: 1885-9
123. Byrd JC, Hertler AA, Weiss RB, et al. Fatal recurrence of autoimmune hemolytic anemia following pentostatin therapy in a patient with a history of fludarabine-associated hemolytic anemia. Ann Oncol 1995; 6: 300-1
124. Rosenkrantz K, Dupont B, Flomenberg N. Relevance of autocytotoxic and autoregulatory lymphocytes in the maintenance of self tolerance. Concepts Immunopathol 1987; 4: 22-41
125. Shevach EM, Thornton A, Suri-Payer E. T lymphocyte-mediated control of autoimmunity. Novartis Found Symp 1998; 215: 200-11
126. Takahashi T, Kuniyasu Y, Toda M, et al. Immunologic self-tolerance maintained by CD25+ CD4+ naturally anergic and suppressive T cells: induction of autoimmune disease by breaking their anergic/suppressive state. Int Immunol 1998; 19: 1969-80
127. Thornton AM, Shevach EM. CD4+ CD25+ immunoregulatory T cells suppress polyclonal T cell activation by inhibiting interleukin 2 production. J Exp Med 1998; 188: 287-96
128. Peller S, Kaufman S. Decreased CD45RA T cells in B-cell chronic lymphatic leukemia patients: correlation with disease stage. Blood 1991; 78: 1569-73
129. Tosti S, Caruso R, D'Adamo F, et al. Severe autoimmune hemolytic anemia in a patient with chronic lymphocytic leukemia responsive to fludarabine responsive treatment. Ann Hematol 1992; 65: 238-9
130. Cheson BD, Vena DA, Barrett J, et al. Second malignancies as a consequence of nucleoside analog therapy for chronic lymphoid leukemias. J Clin Oncol 1999; 17: 2454-60

131. Kroft SH, Tallman MS, Shaw JM, et al. Myelodysplasia following treatment of chronic lymphocytic leukemia (CLL) with 2-chlorodeoxyadenosine (2-CdA). Leukemia 1997; 11: 170
132. Frewin RJ, Provan D, Smith AG. Myelodysplasia occurring after fludarabine treatment for chronic lymphocytic leukaemia. Clin Lab Haematol 1997; 19: 151-2
133. Orchard JA, Bolam S, Oscier DG. Association of myelodysplastic changes with purine analogues. Br J Haematol 1998; 100: 677-9
134. Coso D, Costello R, Cohen-Valensi R, et al. Acute myeloid leukemia and myelodysplasia in patients with chronic lymphocytic leukemia receiving fludarabine as initial therapy. Ann Oncol 1999; 10: 362-3
135. Gordon J, Abdul-Ahad A, Hamblin TJ, et al. Barriers to successful immunotherapy with anti-idiotype antibody. Br J Cancer 1984; 49: 547-57
136. Dillman RO, Shawler DL, Dillman JB, et al. Therapy of chronic lymphocytic leukemia and cutaneous T0 cell lymphoma with T101 monoclonal antibody. J Clin Oncol 1984; 2: 881-91
137. DeNardo GJ, Lewis JP, DeNardo SJ, et al. Effect of lym-1 radioimmunoconjugate on refractory chronic lymphocytic leukemia. Cancer 1993; 73: 1425-32
138. Maloney DG, Grillo-Lopez 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: 2188-95
139. Osterborg A, Dyer MJ, Bunjes D, et al. Phase II multicentre study of human CD52 antibody in previously treated chronic lymphocytic leukaemia. J Clin Oncol 1997; 15: 1567-74
140. Michallet M, Archimbaud E, Bandini G, et al. HLA identical sibling bone marrow transplantation in younger patients with chronic lymphocytic leukemia. Ann Int Med 1996; 124: 311-5
141. Michallet M, Van Biezen A, Bandini G, et al. Allotransplants and autotransplants in CLL in Europe [abstract P085]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
142. Gribben JG, Barlett-Pandite L, Provan A, et al. Disease free status and absence of PCR detectable minimal residual disease suggests eradication of B-CLL following autologous and allogeneic BMT. Blood 1995; 86 Suppl. 1: 457
143. Khouri IF, Keating MJ, Vriesendorp HM, et al. Autologous and allogeneic bone marrow transplantation for chronic lymphocytic leukemia: preliminary results. J Clin Oncol 1994; 12: 748-58
144. Milligan D. MRC Pilot Study for younger patients with chronic lymphocytic leukaemia [abstract]. Report of MRC Adult Leukaemia Working Party and Childhood Leukaemia Working Party Annual Review; 2000 Mar 29; Bournemouth
145. Khouri IF, Keating M, Korbling M, et al. Transplant-lite: induction of graft-v-malignancy using fludarabine-based non-ablative chemotherapy and allogeneic blood progenitor cell transplantation as treatment for lymphoid malignancies. J Clin Oncol 1998; 16: 2817-24
146. Byrd JC, Rai KR, Sausvile EA, et al. Old and new therapies in chronic lymphocytic leukemias. Semin Oncol 1998; 25: 65-74
147. Willis CR, Byrd JC, Shinn CA, et al. UCN-01: a promising new agent for B-CLL that induces apoptosis independent of p53 status and chemosensitizes cells to the effect of fludarabine [abstract P122]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
148. O'Brien S, Kantarjian H, Ellis A, et al. Topotecan in chronic lymphocytic leukemia. Cancer 1996; 75: 1104-8
149. Keating MJ. Compound 506U78: a new guanosine analogue in the treatment of refractory CLL [abstract S18]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
150. Kipps TJ, Wierda WG, Woods SJ, et al. Gene therapy for CLL. [abstract S21]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
151. Stevenson FK, Zhu D, Spellerberg MB, et al. Vaccine development in CLL: lessons from lymphoma [abstract S22]. IWCLL VIII International Workshop on Chronic Lymphocytic Leukemia; 1999 Oct 29-31; Paris
152. Cooperative Group for the Study of Immunoglobulin in Chronic Lymphocytic Leukaemia. Intravenous immunoglobulinfor the prevention of infection in chronic lymphocytic leukemia: a randomized, controlled clinical trial. N Engl J Med 1988; 319: 902-7
153. Weeks JC, Tierney MR, Weinstein MC. Cost effectiveness of prophylactic intravenous immune globulin in chronic lymphocytic leukemia. N Engl J Med 1991; 325: 81-6
154. Molica S, Musto P, Chiurazzi F, et al. Prophylaxis against infections with low-dose intravenous immunoglobulins (IVIG) in chronic lymphocytic leukaemia: results of a crossover study. Haematologica 1996; 81: 121-6
155. Byrd JC, Hargis JB, Kester KE, et al. Opportunistic pulmonary infections with fludarabine in previously treated patients with low-grade lymphoid malignancies: a role for *Pneumocystis carinii* pneumonia prophylaxis. Am J Hematol 1995; 49: 135-42
156. Cheson BD. Therapy for previously untreated chronic lymphocytic leukaemia: a re-evaluation. Semin Hematol 1998; 35: 14-21

Correspondence and offprints: Professor *Terry J. Hamblin*, Department of Haematology, Royal Bournemouth Hospital, Castle Lane East, Bournemouth, BH7 7DW, England.
E-mail: terjoha@aol.com