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AZT+METHOTREXATE IN HIGH-GRADE HIV-RELATED NON-HODGKIN LYMPHOMAS: INTERIM REPORT ON FEASIBILITY AND TOLERANCE

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ABSTRACT

Background. **3'azido-3'deoxythymidine (AZT) is a thymidine analogue useful in the treatment of AIDS. We have previously demonstrated that AZT can possess significant antineoplastic activity when combined with *de novo* thymidylate synthesis inhibitors, such as 5-fluorouracil or methotrexate (MTX). We report here on the feasibility and tolerance of the combination AZT+MTX in the treatment of HIV-related non-Hodgkin lymphomas (NHL).**

Patients and Methods. **IV-positive patients with high-grade NHL were treated, at weekly intervals, with three consecutive courses of MTX 1 g/m^2 plus increasing doses of oral AZT (2, 4 and 6 g/m^2) and folinic acid rescue.**

Results. **From July 1992 to June 1994, 16 patients were enrolled in the trial after giving informed consent. Grade III-IV neutropenia was observed after 22% of the courses, but was prevented by G-CSF administration in 34/59 courses. Grade III-IV anemia was observed after 15% of the courses. No non hematological toxicity was observed.**

Conclusions. **The combination AZT+MTX was extremely well tolerated in this series of HIV-related NHL patients. If these results are confirmed in terms of antineoplastic efficacy, the protocol will deserve wider application.**

Key words: AIDS, non-Hodgkin lymphoma, AZT, methotrexate

AZT (3'azido-3'deoxythymidine) is a thymidine analogue that is clinically useful in the treatment of ARC/AIDS.[1] After intracellular phosphorylation by thymidine kinase, the drug acts as a false substitute for physiological nucleosides at the level of HIV reverse transcriptase; this prevents the formation of DNA copies of the viral (RNA) genome and has proven effective in halting retroviral replication.[2] AZT represents a poor substrate for human DNA polymerase α and thus has not been shown to be active as an antineoplastic drug. It has been observed, however, that the cytotoxic activity of AZT can be increased by combination with 5-fluorouracil (5FU)[3] both *in vitro*, in human cell lines, and *in vivo*, in murine models. The biochemical rationale for this synergism is that phosphorylation, metabolism and incorporation of AZT into DNA are favored when *de novo* thymidylate biosynthesis is inhibited. Phase I clinical trials of AZT in combination with 5FU in non-HIV cancer patients showed good tolerance and safety.[4,5] More recent studies by our group have evaluated the combination of methotrexate (MTX) and AZT. *In vitro*, on colon carcinoma cell lines[6] and leukemic cell lines,[7] the antineoplastic activity of AZT was substantially increased by MTX. Biochemical analysis revealed that this enhanced cytotoxic effect is related to increased incorporation of AZT into DNA. *In vivo* studies in murine models have demonstrated a reduction in tumor growth;[8] furthermore, the toxicity of this combination (both hematological and

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non hematological) has been greater than that displayed by either drug alone.

In this study we aimed at investigating the efficacy of the combination AZT+MTX in HIV-related non-Hodgkin lymphomas (NHL). The choice of MTX rather than 5FU was based upon previously reported data that showed a 20% CR rate in poor-risk NHL treated with MTX in monochemotherapy.[9,10] We report here on the tolerance and feasibility of this protocol.

Patients and Methods

Patients

The study included 16 adult HIV seropositive patients with high-grade NHL, either newly diagnosed or pre-treated (relapsed/refractory). Exclusion criteria were: performance status > 2 (WHO grading system); hemoglobin (Hb) < 8g/dL; white blood cell (WBC) count < 2.0×10^9/L; abnormal hepatic, cardiac or renal function; active opportunistic infections. Before treatment, patients underwent conventional staging procedures including bone marrow trephine biopsy, CT scan, chest X-ray and abdominal ultrasonography.

Treatment plan

Treatment consisted of three consecutive courses of MTX 1 g/m^2 (days 1, 8 and 15) + oral AZT at 2 g/m^2 (days 1, 2 and 3), 4 g/m^2 (days 8, 9, 10) and 6 g/m^2 (days 15, 16 and 17). Leucovorin 50 mg was administered twice daily on days 1-2, 8-9 and 15-16. From the eleventh patient on, the treatment was continued with three more courses at the maximum dose of AZT in case of complete or partial response.

Supportive treatment

Every kind of supportive treatment was allowed, including antibiotic and antifungal (oral amphotericin B or nistatine) prophylaxis and granulocyte-colony stimulating-factor (G-CSF, 5 μg/kg), according to the physician's discretion. Packed RBC or platelet transfusions were given when Hb dropped < 7.5 g/dL or platelet count < 20×10^9/L.

Clinical and laboratory evaluation

Physical examination, WBC count and differential with lymphocyte subpopulations, Hb, platelet count, liver and renal function and serum electrolytes were evaluated prior to treatment and once weekly. HIV1 core antigen was monitored by anti-p24 enzyme immunoassay (Du Pont, France). A complete restaging procedure was performed after three courses.

Results

From June 1992 to June 1994, sixteen patients were considered elegible for the study (Table 1) and were subsequently enrolled in the AZT + MTX trial after giving informed consent. Their median age was 35 years; 8/16 were diagnosed as having full-blown AIDS prior to lymphoma, and 15/16 had fewer than 200 CD4/mm^3 (10/16 fewer than 100 CD4/mm^3). Histological diagnoses were: Burkitt (5 cases), immunoblastic (2 cases), anaplastic large cell lymphoma (3 cases),

Table 1. Clinical characteristics of the patients.

Total number	16
M/F	10/6
Median age (range)	35 yrs (23-49)
Risk group:	
IVDU	8
Homosexual	4
Heterosexual	4
Prior AIDS 8/16	
Prior AZT treatment	10/16
CD4 < 200 mm^3	15/16
Histology:	
Burkitt	5
Anaplastic large cells	3
Immunoblastic	2
Centroblastic	4
High-grade unclassifiable	2
Stage:	
I	1
II	4
III	0
IV	11
Previous treatment:	5/16
F-MACHOP	1
CVP	3
αIFN (Kaposi sarcoma)	1

centroblastic (4 cases), high-grade unclassifiable (2 cases); 11/16 patients were in stage IV, with bone marrow involvement in 7, hepatic involvement in 2, CNS and pleural involvement in 1 case. Five out of 16 patients had been previously treated: 4 for lymphoma (1 with F-MACHOP and 3 with CVP protocol) and 1 for Kaposi's sarcoma (α-interferon). Table 2 reports the data concerning the toxicity experienced with AZT + MTX. Leukopenia with grade III-IV neutropenia (PMN < 0.5×10^9/L) was observed in 13/59 courses (22%); grade III-IV anemia (Hb < 8 g/dL) occurred in 8/59 courses (15%), while platelet count never dropped below 50×10^9/L. Neutropenia was the main factor in determining delays in administering the following course (8 courses were delayed, with a mean delay of 18.6 days). In order to improve the hematological tolerance of this protocol, G-CSF was added after 34/59 courses. In none of these cases was it necessary to postpone the treatment with AZT + MTX. Out of the 14 patients evaluable for response, 8 (57%) obtained a complete remission and 4 (28%) showed a partial response. The follow-up, however, is presently too short to draw any conclusions about the efficacy of this therapeutic regimen. No differences in hematological toxicity were observed between high-risk (CD4 < 200, prior AIDS) and low-risk patients.

Table 2. Hematological toxicity.

Patient	*Courses*	*Neutropenia (grade III-IV)*	*Anemia (grade III-IV)*	*G-CSF (# of courses)*	*Delay (# of courses)*
1	3	1/3	1/3	0/3	1/2
2	3	1/3	1/3	0/3	1/2
3	1	1/1	1/1	0/1	NE
4	3	1/3	2/3	2/3	1/2
5	3	3/3	1/3	0/3	2/2
6	3	3/3	1/3	0/3	2/2
7	3	1/3	0/3	0/3	1/2
8	3	0/3	0/3	2/3	0/2
9	3	1/3	0/3	2/3	0/2
10	3	0/3	0/3	2/3	0/2
11	3+3	0/6	0/6	5/6	0/5
12	3+3	0/6	1/6	5/6	0/5
13	1	1/1	0/1	1/1	NE
14	3+3	0/6	0/6	5/6	0/5
15	3+3	0/6	0/6	5/6	0/5
16	3+3	0/6	0/6	5/6	0/5
TOTAL	59	13/59	8/59	34/59	8/43

Non hematological toxicity was not associated with this protocol. Furthermore, we did not observe any opportunistic infections except for one case of *herpes genitalis* (Pat. #14) which promptly resolved with local therapy. From the sixth patient on, the treatment was carried out in an outpatient setting.

Discussion

Approximately 3-8% of HIV-infected individuals develop NHL during the course of their disease.[11,12] Compared to lymphomas occurring in non HIV-infected populations, HIV-related NHLs display several peculiar characteristics, including presentation in advanced stage, frequent extranodal involvement, *B* symptoms and histological features of high-grade malignancy.[13,14] AIDS itself affords patients a poor tolerance to chemotherapy due to pre-existing cytopenia, low performance status and, above all, the frequent occurrence of opportunistic infections.[15] For these reasons earlier studies with dose-intensive regimens[16,17] were associated with a high incidence of therapy-related deaths and life-threatening infections. More recent studies with less intensive regimens have yielded contrasting results in terms of tolerance, especially in high-risk patients, including ones with prior AIDS, CD4 count < 200/mm^3 or poor performance status.[18,19] Moreover, several authors noted a cumulative toxicity of antiviral and antineoplastic therapy and emphasized the need to withdraw AZT or switch to dideoxycytidine (ddC) while treating the lymphoma.[19,20] The novelty of our study consists in being able to continue administering AZT throughout the entire treatment period. Even though the number of treated patients is small, we have shown that the combination AZT+MTX is extremely well tolerated since leukopenia, the major side effect, was effectively prevented by routine use of G-CSF. In no case were opportunistic infec-

tions or evidence of non hematological toxicity such as oral mucositis, a common side effect of MTX, observed.[21] If these results are confirmed in terms of anti-tumor efficacy in a larger series of patients, the combination AZT+MTX could represent a useful therapeutic approach for HIV-related NHL. This also represents a further achievement in the treatment of lymphoproliferative disorders.[22,23]

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