L-Asparaginase-based treatment of 15 western patients with extranodal NK/T-cell lymphoma and leukemia and a review of the literature


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Background: Extranodal natural killer (NK)/T-cell lymphoma, nasal type, and aggressive NK-cell leukemia are highly aggressive diseases with a poor outcome.

Patients and methods: We report a multicentric French retrospective study of 15 patients with relapsed, refractory, or disseminated disease, treated with L-asparaginase-containing regimens in seven French centers. Thirteen patients were in relapse and/or refractory and 10 patients were at stage IV.

Results: All but two of the patients had an objective response to L-asparaginase-based treatment. Seven patients reached complete remission and only two relapsed.

Conclusion: These data, although retrospective, confirm the excellent activity of L-asparaginase-containing regimens in refractory extranodal NK/T-cell lymphoma and aggressive NK-cell leukemia. Therefore, L-asparaginase-based regimen should be considered as a salvage treatment, especially for patients with disseminated disease. First-line L-asparaginase combination therapy for extranodal NK/T-cell lymphoma and aggressive NK-cell leukemia should be tested in prospective trials.

Key words: L-asparaginase, NK/T-cell lymphoma

introduction

Extranodal NK/T-cell lymphoma, nasal type, is a rare and severe disease, more frequent in Asia and South America than in Europe and North America. It is thought to arise from natural killer (NK) cells or, occasionally, from a subset of γΔ or αβ cytotoxic T cells and shows a striking association with Epstein–Barr virus (EBV). It is characterized by a CD3ε phenotype, with no surface CD3 or T-cell receptor expression, an activated cytotoxic profile with perforin and granzyme B expression, and common expression of CD56 [1–3].

Usually extranodal NK/T-cell lymphomas primarily involved the nasal cavity or other parts of the upper aerodigestive tract but sometimes occur in extranasal sites without involving the nasal cavity or nasopharynx. The term 'nasal type' has been proposed in the World Health Organisation classification to describe both the disease that arises in the nasal cavity and in the extranasal sites [3, 4]. Aggressive NK-cell leukemia is a rare fulminating form of NK-cell neoplasm that is widely disseminated at diagnosis and mainly affects younger patients [4, 5].

There is no consensus treatment. Localized NK/T-cell lymphomas often respond to radiotherapy [6]. In contrast, patients who have extensive disease or who relapse after radiotherapy have a very poor outcome [7], with a median survival time of only 50 days in case of aggressive NK-cell leukemia [5].

Following an Asian case report published in 2003 [8], we treated a patient with l-asparaginase for refractory NK/T-cell lymphoma and rapidly obtained a complete response. Fourteen other patients, of whom 12 were refractory or in relapse and 10 were at advanced stage (stage IV Ann Arbor), were subsequently treated in seven French centers. Here we report retrospectively our experience in the treatment of these 15 French patients.
patients and methods

From June 2003 to October 2006, 13 patients with extranodal NK/T-cell lymphoma, nasal type, and two patients with aggressive NK-cell leukemia were treated with an L-asparaginase-containing regimen, after being fully informed of the nature and possible adverse reactions of these protocols. Fourteen patients were European and one patient was native of north Africa. The patients were included if they met the following criteria: diagnosis of NK/T-cell lymphoma/leukemia on primary biopsy of the nasopharyngeal region or another extranodal site, malignant cells with a CD3+, CD20− phenotype, a cytotoxic profile, and markers of EBV (EBV encodes small nuclear RNA by in situ hybridization or latent membrane protein 1 by immunohistochemistry). Thirteen of the cases were centrally reviewed by two expert pathologists (BP and PG).

Staging consisted of a complete history taking and physical examination and routine blood tests and serum chemistry before treatment. Computed tomography (CT) of the head, neck, thorax, and abdomen was always used to determine the extent of the primary lesion. Magnetic resonance imaging and whole-body positron electron tomography (PET) were used in three and eight cases, respectively.

All patients received protocols with L-asparaginase (Kidrolase®, OPI, Lyon, France) 6000 U/m², either i.v. daily for 5 (in two cases) or 7 (in six cases) days each month, combined with dexamethasone in six cases, etoposide in one case, and methotrexate in one case or the same dose i.m. on days 2, 4, 6, and 8 in seven cases combined with methotrexate on day 1 (in four cases) and dexamethasone in six cases. Dexamethasone was given at the dose of 40 mg for 4 days, at the beginning of each cycle, in 11 patients and 20 mg in one patient. Methotrexate was given i.v. over 2 h at the dose of 3 g/m² 1 day before the first asparaginase infusion at each cycle, with folic acid rescue. Three patients, who developed anaphylactic reactions to L-asparaginase, subsequently received bimonthly courses of peg-asparaginase (Oncaspar®, OPI).

Asparaginase toxicity was monitored by complete blood cell count, coagulation tests, antithrombine, amylase, glycemia, bilirubin, aspartate aminotransferase, and alanine aminotransferase measurements.

response criteria

The treatment responses were assessed using adapted Cheson’s standard criteria [9] to take into account the rarity of lymph node involvement in this type of lymphoma and the frequent PET false positives due to residual inflammatory and infectious lesions, particularly in the nasal cavity. complete response (CR) was defined as no evidence of residual disease; partial response as at least a 50% reduction in tumor burden compared with the beginning of treatment; and no response as less than a 50% reduction in tumor burden or disease progression. Responses were assessed from clinical, radiological, and laboratory studies prescribed by the individual clinicians.

data collection

Data were retrospectively collected on all patients treated with asparaginase for NK/T-cell lymphoma/leukemia in these seven centers with approval from our local ethics committee.

results

baseline characteristics

The characteristics of the 15 patients (10 men, five women) are shown in Table 1. At diagnosis, 12 patients had nasal and three patients extranatal presentation, including two who had aggressive NK-cell leukemia (patients 2 and 7, Table 1). Median age was 45 years (range 38–81 years). Five patients were in relapse and three of them were refractory to salvage chemotherapy. Eight patients were refractory to their first treatment. Only two patients with stage IV disease received the asparaginase-containing regimen as first-line treatment. The median number of therapeutic regimens received by the other patients before inclusion was 1 (range 1–5) including cyclophosphamide, doxorubicin, vincristine and prednisone (CHOP) or CHOP-like regimen in 12 patients.

At the time of inclusion, five patients (33%) had stages I–II disease, and 10 patients (66%) had stage IV disease, with involvement of bone marrow in five patients, the liver in three patients, skin in four patients, the central nervous system in two patients, and an adrenal gland in one patient. The nasal cavity was involved in 10 patients and 11 patients had extranasal involvement, including lymph node involvement in three patients.

Systemic B symptoms were present in 10 patients. Nine patients had a high lactate dehydrogenase (LDH) level.

Four patients had the so-called hemophagocytic syndrome with fever, hepatosplenomegaly, cytopenia, hyperferritiniemia, hypertriglyceridemia, and hypofibrinogenemia.

patient outcomes

All but two of the patients had an objective response to the asparaginase-containing regimen (Table 1).

In three cases the response was only transient, with rapid relapse and death due to progression in two patients who had stage IV disease with bone marrow involvement (patients 5 and 7). The third patient (patient 14), a 80-year-old man who received only one course of l-asparaginase because of hepatitis, but who entered apparent CR, relapsed 8 weeks after the first course. He was then treated with peg-asparaginase (Oncaspar®) with good efficacy and no liver toxicity, but he died of an unrelated cause (complicated diverticulitis) after 4 months.

A fourth patient, a 72-year-old man with a disseminated disease in third relapse, died suddenly 2 months after the first course with an apparent response on his skin localization (patient 1).

Two of the remaining nine patients died from sepsis, 1 and 6 months after the last course of asparaginase, without detectable disease in the first case (patient 10) and in partial remission in the second case (patient 8).

Seven patients were considered to be in CR after the last course of asparaginase-containing treatment. Four of these patients were given intensive treatment with stem-cell support, while three patients received irradiation as consolidation therapy after entering remission (Table 1). Of the four patients who received an intensive treatment, two patients relapsed at 9 and 24 months. Five patients remained alive without recurrence after a median follow-up of 1322 days (721–1549).

elements of asparaginase efficacy

Patient 3. This 51-year-old woman was diagnosed with extranodal NK/T-cell lymphoma localized to the anterior nasal antrum (Figure 1) of the CD3ε+, CD2+, granzyme-B+ CD56+ phenotype. She had no local symptoms apart from nasal obstruction. She was admitted with fever and local
<table>
<thead>
<tr>
<th>No.</th>
<th>Sex/age (years)</th>
<th>Initial stage/site</th>
<th>Initial treatment/relapse treatment</th>
<th>Aspa stage/site</th>
<th>Cycles no./associated treatments</th>
<th>Total dose of aspa (IU)</th>
<th>Consolidation treatment after aspa</th>
<th>Outcome</th>
<th>Follow-up months</th>
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<td>M/72</td>
<td>I/NP</td>
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<td>2/dexa</td>
<td>K: 100 000</td>
<td>ASCT BEAM</td>
<td>NE/DUD</td>
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<td>I/NP</td>
<td>1/metho, dexa</td>
<td>K: 40 000</td>
<td>Rt 40 Gy</td>
<td>CR/NOD</td>
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<td>NE/DOD</td>
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<td>K: 72 000</td>
<td>CR/NOD</td>
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<td>CHOP/IFN/thalid</td>
<td>IIIE/NP</td>
<td>2</td>
<td>K: 120 000</td>
<td>Allograft</td>
<td>PR/DUD</td>
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<td>IV/NP, CNS, eye</td>
<td>3/metho i.v. and i.t., cytarabine i.t. dixa</td>
<td>K: 73 000</td>
<td>Rt 40 Gy</td>
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<td>IV/NP, skin, BM</td>
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<td>K: 162 800</td>
<td>CR/relapse at 9 months, DOD</td>
<td>NE/DUD</td>
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<td>II/NP</td>
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<td>CR/relapse at 9 months, DOD</td>
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<td>Rt 45 Gy</td>
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<td>ESHAP, ASCT BEAM</td>
<td>CR/relapse at 24 months DOD</td>
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<td>K: 40 000; O: 16 000</td>
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<td>K: 50 000</td>
<td>NR/DOD</td>
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<td>2</td>
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</table>

Aspa, asparaginase; CHOP, cyclophosphamide, doxorubicin, vincristine and prednisone; K, Kidrolase®; O, Oncaspar®; DOD, died of disease; DUD, dead unrelated to disease; NOD, no evidence of disease; NP, nasopharynx; BM, bone marrow; CNS, central nervous system; Rt, radiotherapy; CR, complete response; PR, partial response; NE, not evaluated; thali, thalidomide; IFN, interferon; dexa, dexamethasone; metho, methotrexate; DHAP, dexamethasone, cisplatin, and cytarabine; ESHAP, etoposide, methylprednisolone, cytarabine, and cisplatin; ASCT, autologous stem-cell transplantation; BEAM, Bicnu, etoposide, cytarabine, melphalan.
inflammation 14 days after CHOP chemotherapy, and CT showed progression of the nasal tumor (Figure 1). She received a course of L-asparaginase 6000 U/m² i.m. on days 2, 4, 6, and 8 combined with methotrexate 3 g/m² on day 1 and dexamethasone 40 mg from days 1 to 4. The local disorders promptly disappeared. She then received local irradiation (40 Gy). CT (carried out before radiotherapy) (Figure 1) and PET (carried out after radiotherapy) showed a complete response. She is still in CR 37 months after the first course of asparaginase.

**Patient 6.** This 41-year-old man was diagnosed with extranodal NK/T-cell lymphoma localized in the nasal cavity with elevated LDH serum level in November 2002. He received three courses of a high-dose CHOP-like regimen and radiotherapy (40 Gy) and was considered to be in CR in April 2003. He relapsed in November 2003 with fever, maxillary antrum infiltration, and cervical lymphadenopathies. Four weeks after a first course of cisplatin, cytarabine, and dexamethasone (DHAP), the disease progressed with fever, odynophagia, and an increase in cervical node volume. He received a first course of L-asparaginase, 6000 U/m² for 7 days, with 4 days of dexamethasone (40 mg each day, as in the DHAP regimen). He became afebrile after 2 days of asparaginase therapy, and PET showed the complete disappearance of hyperfixation after this first course (Figure 2). He then received two other courses of the same regimen, plus intensive chemotherapy with the bicnu, etoposide, cytarabine, melphalan (BEAM) regimen and stem-cell support. He is well and in persistent CR 50 months after the beginning of asparaginase therapy.

**Patient 10.** This 38-year-old man had stage IV extranodal NK/T-cell lymphoma with palate and skin involvement. The LDH level was 857 units (n < 500 units). After two courses of a CHOP-like regimen, he developed B symptoms and a leukaemic phase with 10 000 lymphomatous cells/mm³. Bone marrow aspiration showed an infiltration by 45% of lymphoma cells. Rapidly after the beginning of the first course of L-asparaginase (6000 U/m² i.v. for 7 days plus 4 days of dexamethasone), B symptoms disappeared and the blood cell counts returned to normal values without any more detectable tumor cell. One week after the second course, he developed septic pneumonia and died, with no detectable lymphoma cells on his blood smears.

### rapidity of responses

The responses were usually very rapid as illustrated by these three case reports. Rapid responses were also seen in patients 1 and 14 on their skin localizations and in patient 2 with a bone marrow biopsy negative after only one course of asparaginase-containing regimen. Patient 7 achieved CR after withdrawal of asparaginase at the beginning of the
second course because of allergy and perforation on his digestive localization. He remained in CR for 6 months before restarting chemotherapy.

toxicity
Ten patients had a fall <60% of the normal value of antithrombin level and were thus treated with antithrombin perfusions (Aclotine®). No documented thromboembolic event was reported. Four patients developed hepatitis (severe in one case) and three had anaphylactic reactions to asparaginase infusion. Three patients developed sepsis, which was fatal in one case (pneumonia), but the relationship with asparaginase exposure was uncertain. Hematopoietic toxicity was mild, neutropenia of grades 4 and 3 occurred in 1 and 3 patients, respectively. No diabetes or pancreatitis was observed.

discussion
Patients with extranodal NK/T-cell lymphoma have a cumulative 5-year survival probability of ~40% [4]. Although involved-field radiotherapy seems to be the preferred option for localized disease, this therapy remains poorly effective [6], with the suggestion that high-dose irradiation associated to combination chemotherapy with P-glycoprotein (P-gp)-unrelated agents may give better results [10, 11]. Chemotherapy protocols generally used to treat lymphomas of other histological subtypes have very little efficacy in patients with disseminated disease or disease recurrence after radiotherapy [7, 12, 13]. Tang et al. [13] reported a 2-year overall survival rate of 23.6% and a median survival of 5 months among patients with stages III and IV disease. In another study, Chim et al. [7] reported that only one patient entered CR after salvage chemotherapy with various regimens among 24 patients who failed to achieve CR following a first line of combination chemotherapy.

The mechanisms of extranodal NK/T-cell lymphoma resistance to conventional chemotherapy are not fully understood, but could be related to the frequent expression of P-gp by lymphoma cells, which is responsible for the multidrug resistance phenotype [14].

L-asparaginase is not affected by multidrug resistance and has an original antitumoural mechanism: tumor cells unable to synthesize L-asparagine die when their stores of L-asparagine are depleted by L-asparaginase. Two independent groups demonstrated that L-asparaginase reduced NK-cell activity on normal NK cells in vitro [15] and induced apoptosis of tumoural NK cells, whereas chemotherapy agents currently used to treat lymphoma patients had no activity [16]. In the former study [16], L-asparaginase was tested on two NK-cell tumor-derived cell lines and on samples from six patients with NK/T-cell lymphoma (n = 4) or leukemia (n = 2). L-asparaginase induced apoptosis of the two cell lines and of five of the six patient samples with a strong correlation among asparaginase synthetase expression, in vitro sensitivity, and the clinical response to L-asparaginase [16].

The first report of the clinical efficacy of L-asparaginase in extranodal NK/T-cell lymphoma was published by Yong et al. [17]. This group, at the Peking cancer hospital, has most experience with L-asparaginase in this setting and has published three reports totaling 33 patients [17–19]. All the patients were resistant to first-line treatment with CHOP, which yielded CR in 13 (23.8%) of the initial population of 46 patients. In this group of 33 patients with refractory disease, 17 patients (51.5%) entered CR with a regimen consisting of L-asparaginase 6000 U/m² i.v. on days 1–7, vincristine 1.4 mg/m² on day 1, and dexamethasone 10 mg on days 1–7, with 28-day cycles. The 5-year overall survival rate was 55.9%. In contrast, only 1 of 24 patients entered CR after treatment with conventional salvage chemotherapy in Chim’s report [7].

We obtained similar impressive responses to L-asparaginase. Seven (46%) of our 15 patients entered CR after L-asparaginase-containing treatment. Three of these seven patients had stage IV refractory disease when L-asparaginase was started, and five patients remain in continuous CR with a follow-up of 43 months. Of the seven patients who reached...
CR, four received high-dose chemotherapy with stem cell transplantation and three received consolidation radiotherapy after completion of asparaginase therapy. We cannot know what would have been the durability of the response without these consolidation therapies but to obtain CR in these patients without asparaginase would have been unlikely [7].

In the latest report from Yong et al. [19], as in our present series, 1-asparaginase was well tolerated, with hepatitis and allergy as the main adverse effects. Two patients in the Chinese cohort and three patients in our study had allergic reactions to asparaginase. These reactions can be life threatening and may lead to treatment failure.

Besides these two series of 33 and 15 patients and another recently published reporting six patients [20], the literature contains only a few case reports [8, 21–26]. Table 2 summarizes all published results of 1-asparaginase treatment for extranodal NK/T-cell lymphoma and aggressive NK-cell leukemia.

In conclusion, the results of this French cohort confirm the efficacy of 1-asparaginase on extranodal NK/T-cell lymphoma and aggressive NK-cell leukemia. These results support the use of 1-asparaginase-containing regimens as salvage treatment for refractory or relapsing patients. We have launched a prospective multicenter trial to better determine the response rate to 1-asparaginase combined with methotrexate and dexamethasone and to identify factors predictive of treatment outcome (ClinicalTrials.gov Identifier: NCT00283985). Data from clinical and laboratory studies may already justify trials using 1-asparaginase in combination therapy for disseminated extranodal NK/T-cell lymphoma in first line as the one began in Japan with 1-asparaginase associated to etoposide and multidrug resistance-unrelated agents [20].

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references


