Familial hemophagocytic lymphohistiocytosis (FHL), also known as familial erythrophagocytic reticulosis and erythrophagocytic lymphohistiosirosis, is a rare inherited disorder, whose genetic basis is unknown, characterized by multisystemic infiltration by lymphocytes and histiocytes. The disease occurs during infancy or early childhood with fever, oedema, hepatomegaly and splenomegaly, pan-cytopenia, hypofibrinogenemia, hypertriglyceridemia, and cerebromeningeal infiltration by lymphocytes and histiocytes. The use of cytotoxic therapy, with etoposide or other drug regimens, has achieved sustained remission from this invariably fatal disease. Steroid treatment and intrathecal infusion of methotrexate (MTX) are used to control central nervous system (CNS) involvement and to prevent secondary relapses. Nevertheless, most patients still relapse and ultimately die from disease resistance or secondary infections. The fact that some patients survive for long periods, however, holds out the prospect of improving treatments. Allogeneic bone marrow transplants can bring about long-term remission (up to 8 years) provided an HLA-matched donor is available. In contrast, chemotherapy is often toxic, and sometimes ineffective in treating the primary disease and relapses.

We have tested immunosuppressive agents as alternative primary and maintenance therapy, on the basis of evidence that T cells may play a key role in the disease. Presence of major histocompatibility complex class II (MHC II) (+) T cells, high levels of soluble serum interleukin-2 (IL-2) receptor, and CD8 and γ interferon have been detected in the serum of FHL patients. Moreover, there is anecdotal evidence that cyclosporine A (CSA), a nonmyelotoxic drug that reversibly inhibits T-cell activation (ie, IL-2 production), has a partial or transient effect in FHL and acquired hemophagocytic syndromes. In this pilot study, six consecutive patients were treated with rabbit antithymocyte globulins (ATG) and steroids over 5 days, followed by CSA maintenance therapy. ATG was selected for its lack of myelotoxicity and its high toxicity for T lymphocytes (it induces immunosuppression in graft rejection and in acquired aplastic anemia).
perfusion of fibrinogen, of irradiated packed red blood cells and platelets; fluid intake was restricted; and broad-spectrum systemic antibiotics were administered. During the maintenance phase, the children received intravenous lugs every 3 weeks and oral trimethoprim-sulfamethoxazole.

CASE REPORTS

Patient 1. Patient 1 (AG) is the first child of unrelated parents; her sister died at 14 months from a recurrence of FHL (patient no. 2 in Blanche et al [4]), despite receiving an HLA-identical marrow transplant from the then healthy patient no. 1. At 4.5 years of age, patient AG developed purpuric thrombocytopenia then fever, hepatomegaly, and pancytopenia and exhibited typical biologic manifestations of FHL (including leukocytes in the CSF). Despite a transient remission obtained by chemotherapy (etoposide, high dose intravenous MTX, cytoxan), systemic remission was not achieved. Clinical and biologic manifestations of the disease at this stage are described in Tables 1 through 3. Rabbit ATG (10 mg/kg/d, over 5 days) and methylprednisolone (2 mg/kg/d) were administered 19 days after the last course of chemotherapy and brought about complete remission of clinical and biologic signs within 7 days. Remission was maintained by treatment with low-dose (4 mg/kg/d) CSA, prednisone (2 mg/kg/d), and 1 infusion of VP16 (200 mg/m²/wk). Seizures due to CNS relapse occurred 7 months later, but remission was again obtained with MTX (8 g/m² intravenously). Parental HLA-mismatched T-cell-depleted marrow was infused, after a conditioning regimen comprising VP16, busulfan, and cyclophosphamide but failed to take. A second attempt led to engraftment, although a mild graft-versus-host reaction occurred. The patient died with uncontrolled seizures at day 105. It was not possible to determine if the FHL had reoccurred (Fig 1).

Patient 2. Patient 2 (JN) is the fourth child of unrelated parents of African origin; two siblings died with typical FHL manifestations at 18 and 19 months of age. Patient no. 2 developed FHL at 30 months of age, presenting with fever, edema, and hepatosplenomegaly, but without manifestations of CNS involvement. She had jaundice, pleuritis, pancytopenia, hypofibrinogenemia, hypertriglyceridemia (Tables 1 and 2) and hemophagocytosis in bone marrow smears; no viral infection was found. Numerous activated CD3+ T cells were found in the pleural effusion. Initial treatment consisted of ATG (10 mg/kg for 5 days) with intravenous methylprednisolone (2 mg/kg/d), and 5 IT infusions of steroids and MTX (10 mg each, over 5 weeks). Fever resolved within 2 days, whereas other manifestations, including hemophagocytosis, disappeared within 7 days. Maintenance therapy, comprising CSA (35 mg twice daily orally) and prednisone (15 mg twice daily) was then started. The patient remained well until 6 months after ATG therapy, when she developed fever and hepatosplenomegaly after interruption of maintenance therapy 3 months previously by her parents. Remission was again obtained after the resumption of CSA and steroid treatment. The patient is currently in good condition 22 months after ATG therapy and is being treated with CSA (0.6 mL twice daily) and prednisone (4 mg/d) (Fig 1).

Patient 3. Patient 3 (GS) is the fourth child of unrelated parents; a brother died at the age of 7 weeks due to hemophagocytic lymphohistiocytosis (shown by autopsy). Patient no. 3 developed fever and hepatomegaly at 2 weeks of age, and laboratory test results (Tables 1 and 2) were consistent with FHL; no viral infection was detected. Treatment with ATG (10 mg/kg/d over 5 days) and methylprednisolone (2 mg/kg/d) was started; 5 IT infusions of MTX (6 mg each) were administered over 6 weeks. Clinical and biologic manifestations disappeared after 1 week. Steroid treatment was progressively reduced over 2 weeks, and CSA therapy was started at day 16 (Fig 1).

Two months later, the child received a transplant of T-cell-depleted, HLA-incompatible marrow from his father. The conditioning regimen included treatment with VP16, busulfan, and cyclophosphamide. The patient is currently doing well, without therapy, 24 months after the onset of the disease with full donor marrow engraftment (Fig 1).

Patient 4. Patient 4 (JC) is the second child of unrelated parents of African origin; two siblings died with typical FHL manifestations at 18 and 19 months of age. Patient no. 2 developed FHL at 30 months of age, presenting with fever, edema, and hepatosplenomegaly, but without manifestations of CNS involvement. She had jaundice, pleuritis, pancytopenia, hypofibrinogenemia, hypertriglyceridemia (Tables 1 and 2) and hemophagocytosis in bone marrow smears; no viral infection was found. Numerous activated CD3+ T cells were found in the pleural effusion. Initial treatment consisted of ATG (10 mg/kg for 5 days) with intravenous methylprednisolone (2 mg/kg/d), and 5 IT infusions of steroids and MTX (10 mg each, over 5 weeks). Fever resolved within 2 days, whereas other manifestations, including hemophagocytosis, disappeared within 7 days. Maintenance therapy, comprising CSA (35 mg twice daily orally) and prednisone (15 mg twice daily) was then started. The patient remained well until 6 months after ATG therapy, when she developed fever and hepatosplenomegaly after interruption of maintenance therapy 3 months previously by her parents. Remission was again obtained after the resumption of CSA and steroid treatment. The patient is currently in good condition 22 months after ATG therapy and is being treated with CSA (0.6 mL twice daily) and prednisone (4 mg/d) (Fig 1).

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Table 1. Clinical, Biologic, and Histologic Findings at Initiation of ATG Therapy

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<th>AG (1)</th>
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<th>GS (3)</th>
<th>JC (4)</th>
<th>SE (5)</th>
<th>SC (6)</th>
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parents. He was admitted to hospital at 3 weeks of age with fever, rhinitis, and anemia; pancytopenia was detected. The child required mechanical ventilation for 9 days. His condition progressively improved, and he was discharged 2 weeks later. However, within 15 days, he developed hepatosplenomegaly, pancytopenia, and fever; laboratory findings were typical of hemophagocytic lymphohistiocytosis (Tables 1 and 2). Viral infection was not detected, but activated T cells were found in the blood (Table 3). Prednisolone therapy (3 mg/kg/d) was started, later accompanied by ATG (10 mg/kg/d over 5 days), and 6 IT MTX infusions (6 mg) over 6 weeks; remission was achieved in 1 week. Treatment with steroids was stopped on day 13, after which the child received CSA (3 mg/kg/d). Mild splenomegaly, hypertriglyceridemia (3.55 mmol/L), and neutropenia were observed 1 month later. CSA levels were found to be low, and the dose was consequently increased to 6 mg/kg/d; complete sustained remission was obtained shortly thereafter. Six months after ATG therapy the child is doing well, at home, on a CSA maintenance regimen (6 mg/kg/d) (Fig 1).

**Patient 5.** Patient 5 (SE) is the fourth child of related parents of Moroccan origin. She developed fever and rash at 1 month of age; lethargy, marked hepatosplenomegaly, edema, and jaundice were observed on admission to hospital, and her condition deteriorated despite antibiotic treatment. She had typical biologic manifestations of FHL (Tables 1 through 3), as well as mucositis and severe neurologic illness (including axial hypertonia and grade I/II coma); a cranial CT scan showed diffuse necrotic lesions. She was immediately treated with ATG (10 mg/kg/d over 5 days) and methylprednisolone (5 mg/kg/d intravenously), and 2 IT injections of MTX (3 mg) plus steroids. The hemophagocytic syndrome was controlled in the periphery within 8 days, but her neurologic condition remained poor. Thirty days after ATG therapy, she developed Fusarium sepsis with skin lesions, an enlarged liver and spleen, and hypertriglyceridemia. Her condition deteriorated, and she died with uncontrolled seizures (Fig 1).

**Patient 6.** Patient 6 (SC) is the seventh child of consanguineous parents of African origin. Two of her siblings died at 3 months of age with typical FHL, which had been unsuccessfully treated with steroids. Patient no. 6 was admitted at 70 days of age with fever, lethargy, hepatosplenomegaly, purpura, and hypertonia (Tables 1 through 3). No viral infection was detected. She was treated with ATG (10 mg/kg/d over 5 days), methylprednisolone (5 mg/kg/d intravenously, progressively withdrawn over 3 weeks), and 5 IT infusions of MTX (3 mg) over 12 days. Complete remission was consequently increased to 6 mg/kg/d; complete sustained remission was obtained shortly thereafter. Six months after ATG therapy the child is doing well, at home, on a CSA maintenance regimen (6 mg/kg/d) (Fig 1).
was obtained 7 days after starting ATG therapy; no side-effects were observed. Continuous intravenous CSA was started (5 mg/kg/d), and subsequently replaced by oral administration at day 20 (10 mg/kg/d in two doses). The girl was discharged after 28 days, and is currently (3 months later) doing well on oral CSA (25 mg/d), corresponding to a blood concentration approximately 150 ng/mL 4 hours after intake (Fig 1).

**DISCUSSION**

Treatment with ATG and steroids rapidly induced systemic remission of 6 ascertained cases of FHL: 5 patients showed complete remission, which was maintained by CSA administration (with or without steroids) for up to 22 months.

Immunosuppressive treatment caused no detectable side-effects in 5 of the patients; 1 developed Fusarium sepsis, but it could not clearly be attributed to the treatment. This absence of side-effects contrasts with the adverse effects of FHL chemotherapy. ATG treatment was accompanied by a transient decrease in T-cell counts, but these were reestablished (>1,000/μL) in 5 patients 2 months after treatment; a secondary T-cell immunodeficiency was not detected.

Clinical improvement was noticeable within 2 days and systemic remission occurred within 1 week. This effectiveness cannot be attributed solely to the accompanying steroid therapy for two reasons: (1) in one patient (no. 1) FHL was resistant to steroids and chemotherapy (etoposide, vincristine, and cytoxan); (2) previous studies have shown that steroid therapy alone at best induces transient and partial clinical and biologic improvement. Thus, it appears that treatment with ATG and steroids may bring about remission more effectively than etoposide or other cytotoxic drugs, and may have fewer side-effects. The choice of ATG dose was empirical, based on previous regimens of ATG use in the treatment of aplastic anemia. Because the regimen used appeared efficient, we did not intend to modify it, although no information is available on what the optimal dose might be.

As in previous studies, poor penetration of ATG through the blood-brain barrier remains an obstacle, as indicated by the CNS relapse in one patient 7 months after treatment, and the death of another patient from severe CNS disease. This problem was addressed by using IT MTX infusions.
similar to those used in previous treatment regimens. Long-term CNS remission of FHL thus remains a concern in patients treated with the proposed protocol.

Remission was maintained using the immunosuppressive drug CSA, which blocks T-cell activation, is nonmyelotoxic, and has been reported to control FHL at least transiently (bringing about remission directly, or maintaining etoposide-induced remission in patients with ill-defined hemophagocytic syndromes). In the present study, CSA (intravenously at 3 to 5 mg/kg in continuous infusion or 8 to 10 mg/kg/d orally) appeared effective, and had no side-effects. Notably, one patient who relapsed after her parents' decision to stop maintenance therapy entered a second remission after CSA resumption. In another patient (no. 4), mild symptoms of FHL reappeared simultaneously with low trough levels of CSA and were subsequently eliminated by increasing the dose of CSA. The apparent safety and efficacy of this T-cell–targeted immunosuppression strategy suggests that the original choice of treatment regimen, despite being based on previous experience in other areas was reasonable.

Nonmyelotoxic immunosuppressive treatment of FHL could be used to bring about remission before allogenic bone marrow transplantation (which demands a healthy HLA-identical sibling donor), or it could be used as a possible alternative to marrow transplants in cases in which no matched donor is available. However, in the absence of other prospects for the treatment of FHL, immunosuppression can at least be envisaged as a relatively safe treatment option.

The effectiveness of the combined ATG/steroid/CSA therapy for FHL further supports the hypothesis that T cells are at the root of the disease. The involvement of T-cell activation and proliferation in the disease is supported by the high serum levels of lymphokine (interferon γ), and the occurrence of circulating activated T cells with high levels of soluble IL-2 receptor and CD8. As T cells trigger macrophage activation via cytokines or cell contact, it is tempting to incriminate a primary T-cell dysfunction, e.g., abnormally regulated activation leading to secondary macrophage activation. Although the effectiveness of our therapy supports this hypothesis, a direct effect on macrophages cannot be ruled out. Efficacy of immunosuppressive therapy in FHL holds out the prospect of more specifically T-cell–targeted therapies.

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REFERENCES

Treatment of familial hemophagocytic lymphohistiocytosis with antithymocyte globulins, steroids, and cyclosporin A

JL Stephan, J Donadieu, F Ledeist, S Blanche, C Griscelli and A Fischer