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Thymoglobulin Prevents Chronic Graft-versus-Host Disease, Chronic Lung Dysfunction, and Late Transplant-Related Mortality: Long-Term Follow-Up of a Randomized Trial in Patients Undergoing Unrelated Donor Transplantation

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ABSTRACT

This is an update of a randomized study on antithymocyte globulin (ATG; Thymoglobulin) before transplantation in patients undergoing unmanipulated marrow transplantation from unrelated donors. The median follow-up for surviving patients is 5.7 years. At last follow-up, chronic graft-versus-host disease (GVHD) was scored in 60% of non-ATG and in 37% of ATG patients ($P = .05$), and extensive chronic GVHD was present in 41% and 15%, respectively ($P = .01$). Chronic lung dysfunction was diagnosed in 51% versus 19% of patients ($P = .005$). Forced vital capacity decreased significantly with time in non-ATG patients ($P = .005$), but not in patients who received ATG ($P = .30$). The proportion of patients with Karnofsky scores of ≥90% at 4 years was 57% versus 89% in non-ATG versus ATG patients ($P = .03$). The actuarial 6-year survival for all patients randomized was 31% versus 44% (non-ATG versus ATG; $P = .80$). The cumulative incidence of transplant-related mortality was 51% versus 41% ($P = .70$) and of relapse was 32% versus 40% ($P = .90$). For patients who survived 1 year, transplant-related mortality was 25% versus 3% ($P = .03$), and actuarial survival was 58% versus 85% ($P = .09$). In conclusion, the addition of ATG to cyclosporine/methotrexate provides significant protection against extensive chronic GVHD and chronic lung dysfunction, reduces late transplant mortality, and improves quality of life in patients undergoing unrelated donor transplantation.

KEY WORDS

Leukemia • Bone marrow transplantation • Unrelated donors • Antithymocyte globulin • Chronic lung dysfunction

INTRODUCTION

Chronic graft-versus-host disease (GVHD) remains a major complication of allogeneic bone marrow transplantation (BMT) and has a significant effect on quality of life and late mortality [1,2]. Because of the increased risk of chronic GVHD in patients who receive alternative donor grafts [3], several centers have been using antithymocyte globulin (ATG) during the conditioning regimen or early after transplantation in addition to the conventional cyclosporine/methotrexate prophylaxis [4-9]. These studies have shown that patients who receive ATG have a reduced incidence and severity of acute and chronic GVHD: the reduction is in the order of ≥20% for both acute GVHD grade II to IV and chronic GVHD [10].

These data have been confirmed by 3 randomized studies [11-13]. It is less clear whether survival is affected: in one trial, we were able to confirm a protective effect of ATG on acute and chronic GVHD, but we could show no effect on transplant-related mortality (TRM) and survival at 3 years [13]. The last patient in this study [13] was entered on July 12, 2000, so the minimum follow-up for surviving patients is now >5 years, and the median follow-up is 5.3 years. The aim of this article is to assess the long-term risk of chronic GVHD, chronic lung dysfunction (CLD), quality of life, survival, and TRM in patients randomized to receive or not receive ATG.

PATIENTS AND METHODS

Study Design

The original study has been published [13]: briefly, 109 patients undergoing unrelated donor transplantation were randomized in 4 centers from the Italian Group for Marrow Transplantation to receive or not receive ATG (Thymoglobulin; Genzyme, Cambridge, MA; 7.5-15 mg/kg) in the conditioning regimen. All patients were prepared with conventional cyclophosphamide and total body irradiation, followed by cyclosporine/methotrexate as GVHD prophylaxis [13].

Table 1. *Clinical Data of Patients Receiving or Not Receiving ATG before Transplantation Who Were Alive on Day +100*

No. Patients	No ATG (n = 37)	ATG (n = 38)	*P* Value
No. of patients in trial 1	20	21	
No. of patients in trial 2	17	17	.60
Median age, y (range)	26 (13-51)	27 (16-44)	.80
% with early disease	60%	47%	.20
Diagnosis			
Acute leukemia	11	11	
Chronic myeloid leukemia	25	27	
Myelodysplasia	1	0	.90
Follow-up (d)			
Median	2078	2065	
Range	108-3159	108-3196	
Chronic GVHD (limited + extensive)	60%	37%	.05
Chronic GVHD (extensive)	41%	15%	.01
Chronic lung dysfunction	51%	19%	.005
% with Karnofsky ≥90% at 4 y	57%	89%	.03
Transplant-related deaths	11 (30%)	6 (16%)	.10
Chronic GVHD	9	4	
Respiratory failure	2	0	
Other	0	2	
Relapse-related deaths	7 (19%)	7 (18%)	.50
No. patients alive	19 (51%)	25 (66%)	.10

ATG indicates antithymocyte globulin; GVHD, graft-versus-host disease.

Patients

Seventy-five patients survived 100 days after BMT with a median follow-up of >2000 days. Patients randomized to the non-ATG (n = 37) or ATG (n = 38) group were matched for age, disease, and disease phase (Table 1). Each patient was updated for survival, chronic GVHD, CLD, relapse of the original disease, and quality of life assessed by Karnofsky score.

Chronic GVHD and Lung Function

Chronic GVHD was diagnosed according to established clinical criteria as absent, limited, or extensive [14]. Data on respiratory complications were available in 67 patients: 35 in the non-ATG and 32 in the ATG group. CLD [15] was diagnosed when patients complained of breathlessness at rest in the absence of infectious causes: computed tomography of the thorax and spirometric studies—namely, forced expiratory volume in 1 second (FEV_1), forced vital capacity (FVC), and carbon monoxide diffusing capacity—were used to confirm the diagnosis. Typically patients presented with a longitudinal reduction in both FEV_1 and FVC (with the FEV_1/FVC ratio within normal limits or increased), thus suggesting a restrictive defect [16], combined with a decrease in carbon monoxide diffusing capacity. Karnofsky scores, taken at different times after transplantation, were available.

Statistical Analyses

The NCSS package (J. Hinze, Kaysville, UT) was used for χ^2 tables, actuarial survival, cumulative incidence rates, Student *t* tests, Mann-Whitney tests, and 2-dimensional plots. The data are presented as non-ATG patients (controls) versus ATG patients, thus merging ATG 7.5 and 15 mg/kg: this is due to the small number of patients, but differences, when present, are described.

RESULTS

Chronic GVHD

At last follow-up, chronic GVHD was scored in 60% versus 37%, respectively, for non-ATG and ATG patients ($P = .05$). Extensive chronic GVHD was present in 41% versus 15% ($P = .01$; Figure 1).

Chronic Lung Dysfunction

The cumulative incidence of CLD was 51% for non-ATG versus 19% for ATG patients ($P = .005$; Figure 2). In the non-ATG group, there was a significant decrease of FEV_1 beyond 2 years (average Δ of −23%; $P = .02$), and the same was true for FVC (average Δ of −20%; $P = .005$). This was not the case for patients who received ATG (ΔFEV_1, −3%, $P =$

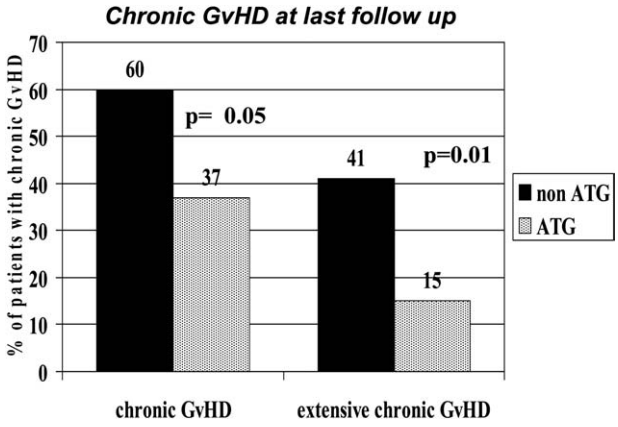

Figure 1. The incidence of chronic graft-versus-host disease (GVHD) at last follow-up is lower in patients who receive antithymocyte globulin (ATG) in the conditioning regimen: this is more significant for extensive chronic GVHD.

.20; ΔFVC, +3%, $P = .30$). The median $FEV_1/\Delta FVC$ ratio was 1.02 and 1.04 for non-ATG and ATG patients, respectively. Bidimensional plots of FVC in the 2 groups, expressed as percentages of predicted values, are outlined in Figure 3. ATG patients had stable FVC with time, whereas non-ATG patients exhibited progressive worsening of FVC, with average FVC of just over 50% 2500 days after transplantation.

Quality of Life

The proportion of patients with a Karnofsky score of ≥90% in the non-ATG versus ATG patients was 28% versus 44% at 1 year ($P = .20$), 62% versus 63% at 2 years ($P = .90$), 56% versus 95% at 4 years ($P = .005$), and 57% versus 89% beyond 4 years ($P = .03$; Table 1).

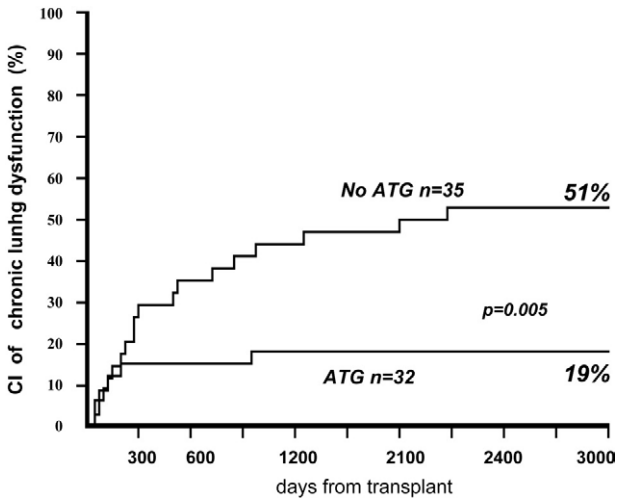

Figure 2. There was a significantly higher cumulative incidence (CI) of chronic lung dysfunction in patients randomized to the non-ATG arm.

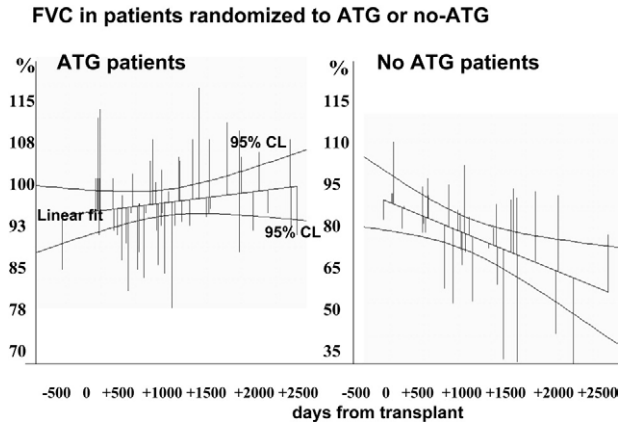

Figure 3. Bidimensional plots of forced vital capacity (FVC) expressed as percentage of predicted in patients who received or did not receive ATG and days after BMT. The linear fit for trend and 95% confidence limits (CL) are shown. FVC (percentage of predicted) and days from BMT are displayed on the y- and x-axis, respectively. Vertical bars represent determinations with 95% CL. ATG patients (58 determinations in 22 patients) show a relatively stable FVC (percentage of predicted), whereas the non-ATG group (38 determinations in 16 patients) shows a continuous decline in this parameter with time.

Survival and TRM

The actuarial 6-year survival for all patients randomized is 31% versus 44% in the non-ATG and ATG groups ($P = .80$; Figure 4A), and in patients surviving 1 year, it is 58% versus 85%, respectively ($P = .09$), with more late events in the non-ATG group (Figure 4B). Similarly, the overall cumulative incidence of TRM is 51% (non-ATG) versus 41% ($P = .70$; Figure 5A). In patients surviving 1 year, it is 25% versus 3% ($P = .03$; Figure 5B).

Causes of Death

Relapse-related deaths were 19% in the non-ATG and 18% in the ATG group ($P = .50$). Other causes of death are outlined in Table 1.

DISCUSSION

We have shown in this study that ATG given before transplantation (1) provides significant protection against chronic GVHD in patients undergoing unrelated donor BMT with cyclosporine/methotrexate prophylaxis, (2) reduces the risk of CLD, (3) improves quality of life, and (4) reduces late TRM.

The first finding relates to the effect of ATG on chronic GVHD, already described in the original study [13] and in keeping with other retrospective studies [4-9]. These studies, involving almost 1000 patients, have all shown a reduction in the order of 20% to 30% of chronic GVHD in patients who receive ATG [10], although retrospective studies

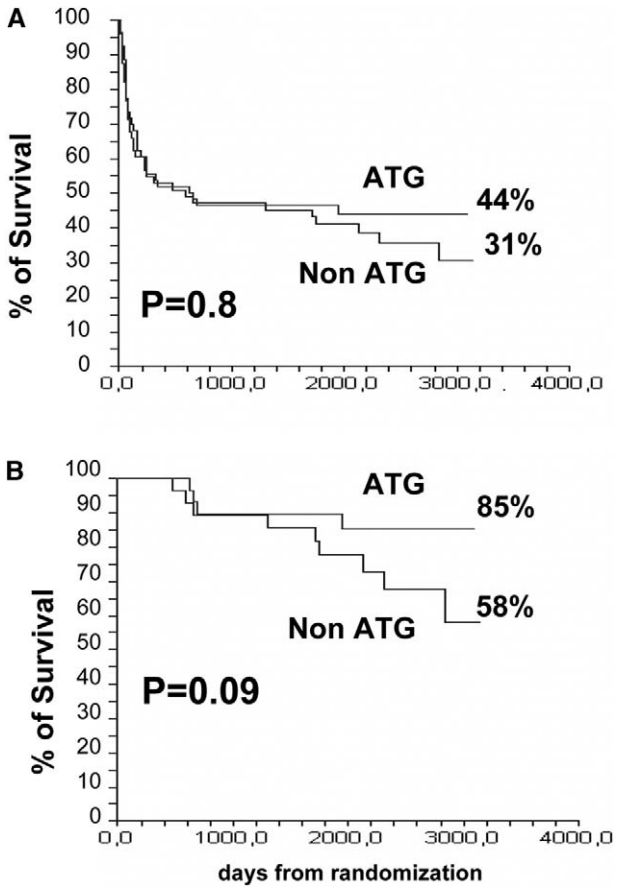

Figure 4. A, Actuarial survival from transplantation of patients randomized to the ATG arm (n = 56) or non-ATG arm (n = 53). B, Actuarial survival of patients randomized to the ATG arm (n = 29) or non-ATG arm (n = 28) who were alive 1 year after transplantation.

can be biased by selection of patients for one treatment or the other. We believe that the strengths of this study are the prospective randomized nature of the trial and the long median follow-up of >2000 days. The small number of patients is a drawback, especially when they are broken down according to the dose of ATG received (15 or 7.5 mg/kg): nevertheless, the results showed a significant difference when ATG patients were compared with controls. We are showing in this article chronic GVHD as assessed at last follow-up, because this takes into account treatment and responses. The protective effect of ATG on chronic GVHD included the limited form, but more so the extensive form, of the disease: this is important because it is the persistence of the latter that exposes patients to a high risk of severe complications [2], including chronic lung dysfunction.

CLD can be restrictive, obstructive, or mixed [15]: the restrictive form seems to be more sensitive to immunosuppressive therapy, whereas the obstructive form is not [15]. In this study, the FEV_1/FVC ratio of our patients was close to 1 or more, thus suggesting that the restrictive defect predominated. Despite this, we have seen little response to immunosuppressive therapy: some of these patients have had progressive lung dysfunction necessitating chronic oxygen supplementation and eventual admission to an intensive care unit. CLD has been associated with chronic GVHD [17]. Actually, CLD could be considered a form of lung GVHD. In this study, the cumulative incidence of CLD was significantly higher in non-ATG patients compared with ATG patients. Bidimensional plots of FVC, expressed as percentages of expected values, indicate 2 separate patterns of evolution: the ATG patients show relatively stable average FVC, whereas the non-ATG group shows a continuous decline of FVC with time that is suggestive of a progressive restrictive defect. Two patients died recently of respiratory insufficiency in intensive care units, and 2 additional patients have severe restrictive defects that necessitate oxygen supplementation: all of these are in the non-ATG group. As a consequence, ATG patients, who survive their transplantation with less

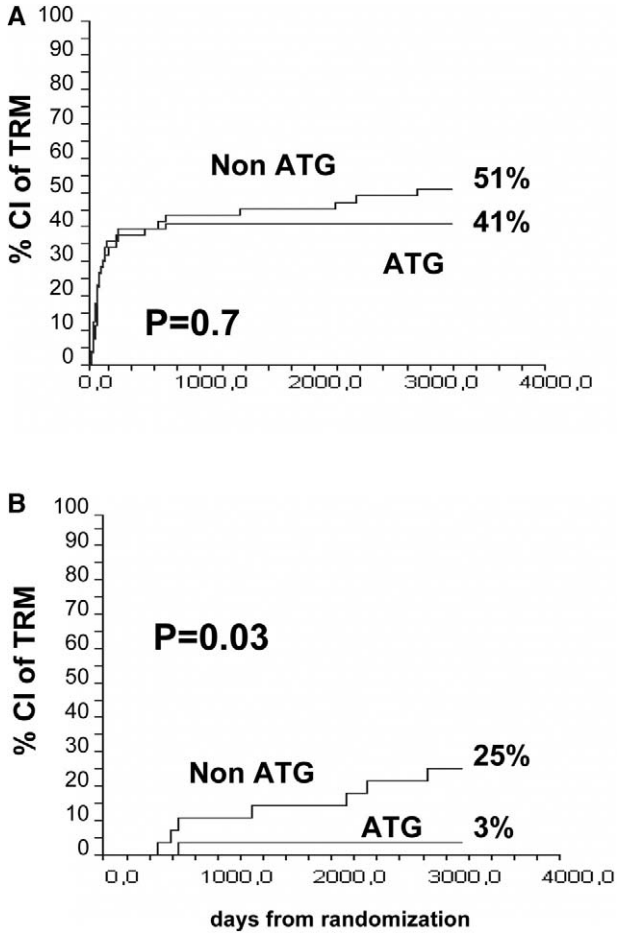

Figure 5. A, Cumulative incidence (CI) of transplant-related mortality (TRM) of patients randomized to the ATG arm (n = 56) or non-ATG arm (n = 53). B, Actuarial 9-year survival of patients randomized to the ATG arm (n = 29) or non-ATG arm (n = 28) who were alive 1 year after transplantation.

chronic GVHD and fewer respiratory problems, have a better quality of life, as indicated by Karnowsky scores ≥90%, and this has become more evident as time goes by.

If the form of chronic GVHD prevented by ATG is clinically relevant, then one should see an effect on TRM: indeed, TRM was comparable at the time of the original publication [9] and also in this study up to 1 year after transplantation. With longer follow-up, TRM starts to diverge 1 to 2 years after transplantation, and 2000 days after transplantation ATG patients have a lower rate of TRM per year as compared with non-ATG patients. Relapse-related deaths were comparable, as was long-term survival, with a trend in favor of ATG patients, although the numbers are small.

Are these data sufficient to suggest that ATG should be used in all patients undergoing an unrelated donor transplantation? Many centers would disagree, and this is primarily based on the results of ex vivo T-cell depletion: that is, removal of T cells from the graft before it is infused into the patient. A large International Bone Marrow Transplantation Registry study has shown that there is no overall advantage for ex vivo T-cell depletion over unmanipulated grafts [18], despite the reduction of GVHD, the principal cause of failure in allogeneic transplantation. Second, ATG is not without side effects, and drawbacks include an increased risk of early bacterial infections [10], Epstein-Barr virus reactivation, and Epstein-Barr virus–associated lymphoproliferative disease [19]; there also may be an increased risk of relapse associated with a reduced graft-versus-leukemia effect [20]. Third, there have been very few prospective randomized studies, usually with short follow-up, and there has been little time to observe late events. In this study, despite small numbers of patients, we have shown that ATG can prevent chronic GVHD and CLD, but more importantly, we are suggesting that it may take 4 to 5 years before chronic GVHD produces its negative effects on quality of life and mortality.

If this is true, this study would also suggest that a 2-drug combination, such as cyclosporine/methotrexate, does not provide sufficient protection in the unrelated donor setting against chronic GVHD and late complications. This may be relevant at a time when more unrelated transplantations are being performed and when peripheral blood is increasingly used as a stem cell source. Also, prospective trials comparing ATG patients versus controls are difficult to set up, given the very long time it takes for planning and follow-up to capture late events. We are currently working on protocols involving different ATG timing and schedules [18], and perhaps this triple combination—ATG, cyclosporine, and methotrexate—may be compared with other 3-drug combinations.

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