Effect of CMV-immunoglobulins (cytotect biotest) prophylaxis on CMV pneumonia after lung transplantation

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--- SUMMARY ---

Lung transplant (LT) recipients among solid organ transplant recipients are at high risk for cytomegalovirus (CMV) infections. We evaluated the effect of CMV-Immunoglobulins (CMV-IG) (Cytotect Biotest) on CMV pneumonia diagnosed in 303 follow-up transbronchial biopsies (TBB) of lung transplant recipients.

24 patients (control group, 155 TBB from 1999 to 2002) received acyclovir for 24 months and 33 recipients (study group, 148 TBB from 2003 to 2008) received a combined CMV prophylaxis consisting of CMV-IG (Cytotect Biotest) for 12 months and a short Ganciclovir or Valganciclovir therapy from 21th to 42th postoperative day followed by acyclovir up to 24 months.

In our study the percentage of pneumonia at first month TBB was similar in the study group vs the control group, 9.1% (3/33) vs 8.3% (2/24), p=0.9 ns, but after the first month the percentage was significantly lower in the study group in the first year at follow-up TBB, 1% (1/99) vs 6.4% (5/78), p=0.048, and in first two years follow-up TBB, 0.8% (1/122) vs 6.5% 8/124), p=0.018 (Statistical analysis: Chi-square test for proportion differences).

Our data suggest a strong efficacy of CMV-IG prophylaxis in reducing CMV pneumonia after first month in lung transplant recipients.

KEY WORDS: Lung transplantation, CMV immunoglobulin, Pneumonia, Prophylaxis

INTRODUCTION

Cytomegalovirus (CMV), a member of the beta herpesvirus group, causes a spectrum of disorders, which includes serious illness in organ transplant recipients (Pereyra *et al.,* 2004; Duncan *et al.,* 1991; Sia *et al.,* 2000). CMV is associated with significant morbidity after transplantation, although the availability of effective antiviral drugs has decreased CMV-related mortality. CMV infection or pneumonitis has also been associated with the development of chronic rejection

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(bronchiolitis obliterans syndrome, BOS) in some, but not all studies (Sharples *et al.,* 2002; Tamm *et al.,* 2004). The endothelium represents the anatomical and functional interface between engrafted donor tissue and the host immune system and interacts dynamically in immunomodulation. This implies an important role for the endothelial cell (EC) in the initiation, maintenance and/or termination of immune interactions affecting the fate of allograft (Pober *et al.,* 1990). CMV does not directly induce HLA class II on infected human EC, but the rejection associated class II induction within the allograft might be a consequence of IFN-gamma release by activated T CD4+ cells in response to CMV infection (Waldman *et al.,* 1993). The rejection process can be hypothesized in three steps:
1) T cell activation by CMV-infected vascular endhotelial cells;

2) cytokine production by activated T cells;
3) cytokine mediated enhancement of graft end-hotelial alloimmunogenicity.

In fact several experimental trials have provided evidence that post transplant CMV infection is involved in the process of chronic allograft rejection in heart and lung transplantation via immune modulation (Everett *et al.,* 1992; Tikkanen *et al.,* 2004).

The aim of treatment has mainly been the reduction of infection and mortality CMV related but many studies have demonstrated a possible role of CMV on rejection (Waldman *et al.,* 1993). CMV pneumonitis was identified to be the stronger independent predictive factor for the later development of BOS (Tamm *et al.,* 2004).

Several groups have intended to find the optimal preventive strategy to avoid CMV infection after lung transplantation demostrating that prolonged valgancyclovir prophylaxis (at least 180 days) following combined prophylaxis together with Ganciclolvir ang CMV-immune globulin (CMV-IG) is safe and effective (Zamora *et al.,* 2004).

Moreover a beneficial effect of combined prophylaxis in cardiothoracic transplantation (heart, heart-lung and lung transplantation) has been demonstrated using an extensive therapeutic regimen of CMV-IG for at least 3 and 4 post transplant months (Valantine *et al.,* 2001; Weill *et al.,* 2003). CMV-IG in addition to ganciclovir within the first postoperative month is highly effective in reducing CMV infection, in preventing both CMV-related mortality in high-risk lung transplant recipients and development of bronchiolitis obliterans syndrome (BOS). CMV-IG has been used in transplantation for more than a decade. However, this substance has never been evaluated by a prospective randomized clinical trial in lung transplant recipients. Most immunosuppressive and anti-infective drugs have only been proven for safety and efficacy in kidney or liver transplant recipients.

Therefore, no clear recommendations concerning prophylactic regimen and dosage in cardiothoracic transplantation are available from the companies distributing CMV-IG (Ruttman *et al.,* 2006). Aim of the present study is to investigate the role of combined CMV prophylaxis on CMV pneumonia in the first two years following lung transplantation.

MATERIALS AND METHODS

A series of 57 lung transplant recipients was analyzed. Patients were classified regarding the regimen of CMV prophylaxis adopted. A group of 24 patients (control group) received prophylactic regimen based on aciclovir 200 mg bid 3 weeks after transplantation for a 24 month period and ganciclovir treatment in case of shell vial positive viral colture on BAL or tissue viral inclusions at transbronchial biopsy or 3 to 5 times increasing level of antigenemia assay at blood samples. A second group (study group) including 33 recipients received ganciclovir or valganciclovir therapy from 21st day after transplantation for 3 weeks and in case of positivity of shell vial viral colture on BAL or positive tissue inclusion or 3 to 5 times increasing level of antigenemia assay; an additional treatment with CMV-IG (Cytotect Biotest) at day 1, 4, 8, 15, 30 post transplant (1.5 ml/kg body weight) and every month for 1 year after lung transplant (1 ml/kg body weight) was also given intravenously.

Immunosuppressive protocol
Intraoperatively 1 gram of methyl-prednisolone was administered before finishing the atrial anastomosis and was tapered from 40 mg to 25 mg per day within the first week and further reduced to 10-15 mg per day within the first 3 to 6 months posttransplant. Beside corticosteroids maintenance immunosuppression was administered by standard triple therapy including cyclosporine and azatioprine as initial immunosuppression, shifting respectively to tacrolimus and mycophenolate mofetil in case of repeated histologically proven acute rejections (2 A3 or 3 A2, working formulation 1995) or in case of lymphocitic bronchitis-bronchiolitis at the TBB. Therapy using an interleukin-2 receptor antibody (basiliximab) was given in day 1^t and day 4 after transplant.

CMV prophylaxis regimen
The control group received antiviral prophylaxis consisting of aciclovir starting at day 1 with a daily dose of 10 mg per kg body weight bid and anti CMV therapy with i.v. Ganciclovir 5 mg/kg bid in case of positivity of pp65 CMV antigenemia or broncoalveolar lavage or transbronchial biopsy confirming clinical suspect of CMV infection.
In the study group oral ganciclovir 3 grams dai-

ly prophylaxis or valganciclovir (VAL) 900 mg twice daily prophylaxis was given starting on the 21st day after lung transplantation for 3 weeks independently from the CMV matching. Moreover, additional CMV-IG (1.5 ml/kg, 100 mg/ml in the first month and 1.0 ml/kg in the following months) was administered at days 1, 4, 8, 15, 30 and monthly for 1 year. CMV surveillance was performed by using the pp65 CMV antigenemia test twice a week during the initial hospital stay, on every visit or in case that CMV infection or disease were suspected. If clinical CMV infection was suspected, transbronchial biopsy (TBB) and broncoalveolar lavage (BAL) was performed. BAL samples were analyzed by shell vial colture and PCR methods and biopsy speciment for histopatologic evidence of CMV to confirm cllinical infection. In case of CMV infection or clinical disease intravenous GAN and oral Gan or VAL was administered for at least one month.

Statistical analysis

Statistical analysis was performed using Chi-square test for proportion differences.
P- values less than 0.05 were considered to indicate statistical significance.

RESULTS

In our study in first year TBB (months 1,3,6,9,12) the percentage of CMV pneumonia in study group was lower, 3% (4/132) vs 6.8% (7/102), p=0.17 ns, as in the first two years TBB (months 1, 3, 6, 9, 12, 18, 24), 2.5% (4/155) vs 6.7 (10/148), p=0.08 ns but the percentage of pneumonia at first month TBB was similar in study group vs control group, 9.1% (3/33) vs 8.3% (2/24), p=0.9 ns. In the study group CMV pneumonia were seen mainly in the first month TBB. Therefore we analyzed the percentages after the first month: the percentage was significantly lower in the study group in first year TBB (months 3, 6, 9, 12), 1% (1/99) vs 6.4% (5/78), p=0.048, and in first two years TBB (months 3, 6, 9, 12, 18, 24), 0.8% (1/122) vs 6.5% (8/124), p=0.018.

DISCUSSION

Due to the high mortality rate associated with CMV disease in the transplantation setting the optimal strategy for the control of CMV infection is the prevention of overt disease. The two main approaches for prevention of CMV disease are prophylaxis and preemptive therapy with currently avaible antiviral compounds. Both strategies are able to reduce the incidence and severity of CMV disease. The possible role played by CMV-IG remains unclear.

Our data indicate that CMV-IG in addition to a short ganciclovir or valganciclovir therapy in the first two postoperative years after lung transplantation were effective in reducing CMV pneumonia.

In particular we found a protective effect from third month TBB for two years but we did not find efficacy in first month follow-up TBB.

The effect seems to last for at least 12 months after the first year of combined prophylaxis suggesting some "long-lasting" protective effect in the second year on CMV pneumonia. It is not clear if this effect is either just direct, or, at least in part, indirect. The direct effect could be mainly IG mediated, as can be supposed by the efficacy in 3, 6, 9, 12 months TBB when CMV-IG was the only prophylaxis administered. The indirect effect could be related to a reduction of the number of acute rejection and lymphocytic bronchitis/bronchiolitis (AR and LB), as we already demonstrated (Solidoro *et al.,* 2008).

In fact we investigated the role of combined CMV prophylaxis on AR and LB in the first year following lung transplantation in 46 patients.

Twenty-five recipients (control group) receiving a total of 107 Transbronchial Biopsies (TBB), had CMV prophylaxis consisting of acyclovir for at least 24 months; 21 recipients (study group) receiving a total of 73 TBB, had CMV prophylaxis consisting of ganciclovir or, since 2005, valganciclovir associated with CMV-IG (Cytotect Biotest) as in present study. In the study group the number of episodes of A2 and A3 acute rejections (AR) needing pulse steroids therapy and lymphocytic bronchitis (LB) were significantly lower than in the control group: 6% (5/73) vs 17% (19/107), p=0.04 and 2% (2/73) vs 11% (12/107), p=0.04, respectively. A logistic regression analysis was performed using the presence/absence of AR as dependent variable and combined CMV prophylaxis, LB, CMV pneumonia as independent variables. The result was a strong significant relationship between combined CMV prophylaxis and reduc-

tion in prevalence of AR (OR 3.25, CI 1.12-9.40, p0.03) (19).

The data of our studies seem to suggest a role of CMV-Ig prophylaxis in reducing CMV pneumonia and both AR and LB, risk factors for chronic rejection.

However both the absence of effect on first month TBB and the reduction of CMV pneumonia from the third month in lung transplantation need more studies to better define the immunological CMV-related mechanisms and their relationship with acute and chronic rejection; the goal is the correct definition of the best cost saving strategy to prevent CMV infections and reduce drug related side effects.

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