Treatment of Persistent Rejection with Methotrexate in Stable Patients Submitted to Heart Transplantation

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Objective – To evaluate the use of methotrexate for the treatment of recurrent rejection in heart transplant recipients.

Methods – We studied 6 patients submitted to heart transplantation that showed rejection grade ≥3A (ISHLT) in two consecutive endomyocardial biopsy specimens. The dose was 11.26±3.75mg/week. The evaluated data were: ventricular function, endomyocardial biopsy, white cell count and number of rejection episodes before and after methotrexate administration.

Results – There was a reduction in the number of rejection episodes (5.17±1.47 before methotrexate; 2.33±1.75 after 6 months and 3.17±2.99 after 12 months of treatment, p=0.0193). The ventricular function was normal with ejection fraction of 76.5±4.80 before and 75.6±4.59 after methotrexate (p=0.4859). One patient did not finish the treatment because he showed signs of rejection associated with severe pericardial effusion. Five patients had a reduction in the white cell count (8,108±23.72 before and 5650±1350 after methotrexate, p=0.0961). One pulmonary infection with complete resolution after antibiotic treatment was observed.

Conclusion – Methotrexate in low doses is an effective adjunct therapy in the treatment of recurrent rejection after heart transplantation.

Keywords: heart transplantation, methotrexate, rejection

The first heart transplant in Brazil was performed by Zerbini in 1968. Currently heart transplantation represents a well-established therapeutic option for end-stage heart failure that does not respond to medical treatment.

During the last decade, cyclosporine and other immunosuppressant drugs produced better survival rates and quality of life for heart transplant recipients by reducing the frequency and intensity of rejection episodes.

Despite the advances in immunosuppressant therapy, acute graft rejection is still a major cause of morbidity and mortality in heart transplantation recipients. A acute rejection accounts for 20% and 25% of deaths in the early and late phases after transplantation, respectively. Methotrexate, a folate acid analog, was initially used for the treatment of some malignancies, severe psoriasis, and rheumatoid arthritis, and is now being used as an adjuvant immunosuppressant drug in cases of persistent rejection resistant to ordinary treatment.

The objective of our study was to evaluate the use of low-dose methotrexate in outpatients with persistent rejection grade ≥3A (International Society of Heart and Lung Transplantation).

Methods

We studied six patients, five man (83.3%) and one woman (16.7%), mean age 35±19.5 years, who underwent orthotopic heart transplantation in our Hospital and were followed by the same medical and multidisciplinary team. The diagnoses before heart transplantation were dilated cardiomyopathy (66.6%), ischemic heart disease (16.7%), and Chagas’ disease (16.7%). For baseline characteristics of these patients refer to table I.

All patients were clinically and hemodynamically stable, left ventricular function was normal, and two consecutive endomyocardial biopsies revealed rejection grade ≥3A after oral corticosteroid pulse therapy. Endomyocardial biopsies were analyzed according to the International Society of Heart and Lung Transplantation (ISHLT) criteria.
Persistent rejection was defined as stability or worsening of rejection gradation (≥3A) in two consecutive biopsies 7 to 10 days apart. Patients received two courses of pulse therapy with oral prednisone 1mg/kg per day during four days, followed by 0.2mg/kg reduction every 4 days until the previous maintenance corticosteroid dose was achieved. If no improvement could be demonstrated, patients were selected to receive low-dose methotrexate in addition to the routine three-drug regimen. The immunosuppressant drug regimen is further described in Table I. Routine posttransplantation evaluation for rejection comprises three endomyocardial biopsies performed in weeks 1, 2 and 4 after transplantation. Beginning in week 4 (one month after transplantation) recipients underwent gallium-67 isotope scintigraphy every two weeks up to the third month and thereafter monthly up to the sixth month. If clinical presentation or gallium scintigraphy were suggestive of rejection, patients then underwent endomyocardial biopsy for diagnostic confirmation. Treatment was only initiated after positive diagnostic findings.

The mean administered dose of methotrexate was 11.26±3.75mg/week for 96±14 days, initiated 554±822 days after transplantation.

The analyzed parameters were: ventricular function, endomyocardial biopsy, white blood cell count, and rejection episodes before and after methotrexate administration. Statistical analysis was performed through paired Student's t-test with a significance level of p<0.05.

**Results**

As shown in Table I, methotrexate was administered 554±822 days after transplantation, with a starting dose of 7.5±1.58mg/week, an average dose of 11.26±3.75mg, for 96±14 days. Patients had an average of 5.17±1.47 episodes of rejection before methotrexate, 2.33±1.75 episodes after six months, and 3.17±2.99 episodes after 12 months of treatment (p<0.0193) (fig. 1). Three patients (50%) had no rejection after methotrexate interruption, 2 patients (33.33%) had rejection grade 1A and one patient was refractory to treatment persisting in rejection grade 3A.

Left ventricular function remained within normal limits. Left ventricular rejection fraction was 76.5±4.8% before and 75.6±4.59 after treatment with methotrexate (p=0.4859) (fig. 2).

One patient did not complete the treatment because he had severe pericardial effusion on echocardiogram associated with signs of fatigue. The patient was admitted to the hospital and responded well to intravenous corticosteroid pulse therapy. In five patients, a reduction occurred in white cell blood count from 8,108±2,372 before to 5,650±1,350 after treatment with methotrexate (p<0.0961) (fig. 3). We observed one episode of infection with white blood cell count elevation and signs of pulmonary infection. The patient's status improved after the administration of antibiotics in addition to methotrexate.

Survival rates were 100% in the first year and 83.33% by the end of the fifth year. One patient died 44 months after transplantation due to acute rejection. Survivors remained clinically stable.

**Discussion**

The folic acid analog methotrexate, initially used for the treatment of some malignancies, severe psoriasis, and rheumatoid arthritis, has been used as adjuvant medication to the routine three-drug immunosuppressive therapy (cyclosporine, azathioprine, prednisone) in cases of persistent rejection.10,12,13.

The use of methotrexate was proposed in 1988 by Constanzo-Nordin et al., when they reported improvement in 9 out of 10 episodes of persistent rejection after methotrexate administration.11,12.

Methotrexate's beneficial effects are due to its action in cellular and humoral immune response, reducing B lymphocyte ability to secrete antibodies in response to specific and nonspecific stimulation. In addition, methotrexate interferes with cytokine production through interleukin secretion and reduces purine synthesis causing diminution in lymphocyte proliferation.11,13,15,16.

Only one patient in the study did not reach the end of the proposed treatment because of fatigue, leg edema and severe pericardial effusion demonstrated by chest radiogram and echocardiogram. He was admitted to the hospital to receive corticoid pulse therapy.

Patients in this study were clinically and hemodynamically stable, and it is known that such patients have lower mortality. However, the question of whether or not to treat rejections in cases like these remains unsolved. There are clinical reports describing patients treated without pulse therapy for rejection grade 2 and also reports from groups that defend control of rejection at any price considering the re-

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**Table I - Patients identification**

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<thead>
<tr>
<th>#</th>
<th>Sex</th>
<th>Etiology</th>
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<tr>
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<td>M</td>
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<td>26/6/85</td>
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<tr>
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**Table II - Immunosuppressant therapy**

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<th>#</th>
<th>Azathioprine (mg/kg/day)</th>
<th>Prednisone (mg/kg/day)</th>
<th>Cyclosporine (mg/kg/day)</th>
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Table III - Results after methotrexate (MTX)

<table>
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<tr>
<th>#</th>
<th>MTX Start (days)</th>
<th>Rjct episodes before MTX (6 mo)</th>
<th>Rjct episodes after MTX (6 mo)</th>
<th>Rjct grade after MTX</th>
<th>Use of MTX</th>
<th>Initial dose MTX (mg/wk)</th>
<th>Avrg dose MTX (mg/wk)</th>
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MTX - methotrexate; Rjct - rejection; Gr - rejection grade; wk - week; Avrg - average.

In conclusion, methotrexate is a safe and effective adjuvant immunosuppressant therapy, promoting reduction in rejection episodes, with mild side effects and is capable of preserving myocardial function.

Limitations of this study: Although the treatment was administered to a preselected group of patients, this was not a randomized clinical trial for it lacked a control group of patients using corticoid pulse therapy, and therefore we could not compare the efficacy and safety of each regimen.

The number of patients in our study was small because persistent rejection is less frequently seen with the currently available three-drug immunosuppressant regimen. Most cases of rejection subside after the first course of corticoid pulse therapy.
References