

An Update on Immunosuppressive Medications in Transplantation

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Introduction

Successful organ transplantation requires the use of immunosuppressive drugs to prevent the host's immune system from rejecting the transplanted organ. Over the past 40 years, immunosuppressive drugs used in clinical transplantation have evolved greatly. During the 1960s and 70s, corticosteroids and azathioprine were the basis of immunosuppressive therapy for organ transplantation. This period was characterized by high incidence of acute rejection and low graft survival rate. The introduction of cyclosporine A in the 1980s represented a major advance in immunosuppressive therapy and substantially improved the 1-year graft survival rate. The so call triple immunosuppressive regimen, consisting of corticosteroids, azathioprine and cyclosporine A, became the standard immunosuppressive protocol for many transplant centers throughout the world. The past 10 years saw the addition of a number of new agents to the armamentarium for immunosuppression, which include a microemulsion formulation of cyclosporine A, tacrolimus, mycophenolate mofetil, rapamycin, daclizumab and basiliximab. Moreover, the recent explosion in our understanding of the cellular and molecular mechanisms involved in the immune response against a transplanted organ has led to the development of several novel immunosuppressive agents. The aim of this article is to provide an update on the established and recently approved immunosuppressive drugs in clinical transplantation and to discuss the future trend of immunosuppressive strategies.

Mechanism of Graft Rejection

In order to understand how various immunosuppressive drugs work, some familiarity with the molecular events involved in the immune response leading to the rejection of a transplanted organ is required. The immune response can be conceptualized as consisting of 3 distinct signals¹ and is shown diagrammatically in Figure 1. The engagement of the T cell receptor (TCR) with the antigenic peptide in the context of self-major histocompatibility complex (MHC) class II molecule provides signal 1. Antigen recognition leads to the

activation of the calcineurin pathway and results in the induction of a number of cytokine genes (e.g. Interleukin-2). Signal 2, or the costimulatory signal, involves the engagement of the CD28 molecule present on the T cell with a member of the B7 family of molecules on the antigen-presenting cell. This synergizes with signal 1 to induce cytokine production. The signal 3 refers to the interaction between the cytokines produced by the activated T cell and their corresponding receptors on the target lymphocytes, which leads to the induction of cell cycling and clonal proliferation through the target of rapamycin (TOR) pathway. De novo purine synthesis is required for the lymphocytes to enter the cell cycle and proliferate.

Established Immunosuppressive Drugs

The immunosuppressive drugs that are currently approved for use in clinical transplantation are listed in Table 1 and their main site of action in relation to the three-signal model of T cell activation is shown in Figure 2.

Azathioprine

Azathioprine is a purine analogue and has been used as an immunosuppressive agent since the

1960s. Azathioprine suppresses the proliferation of B and T lymphocytes through interfering with normal purine synthesis and inhibiting DNA synthesis. The major side effects of azathioprine are bone marrow suppression and hepatotoxicity. Interaction between azathioprine and allopurinol, a xanthine oxidase inhibitor, can lead to excessive marrow toxicity and simultaneous administration of both drugs should be avoided.

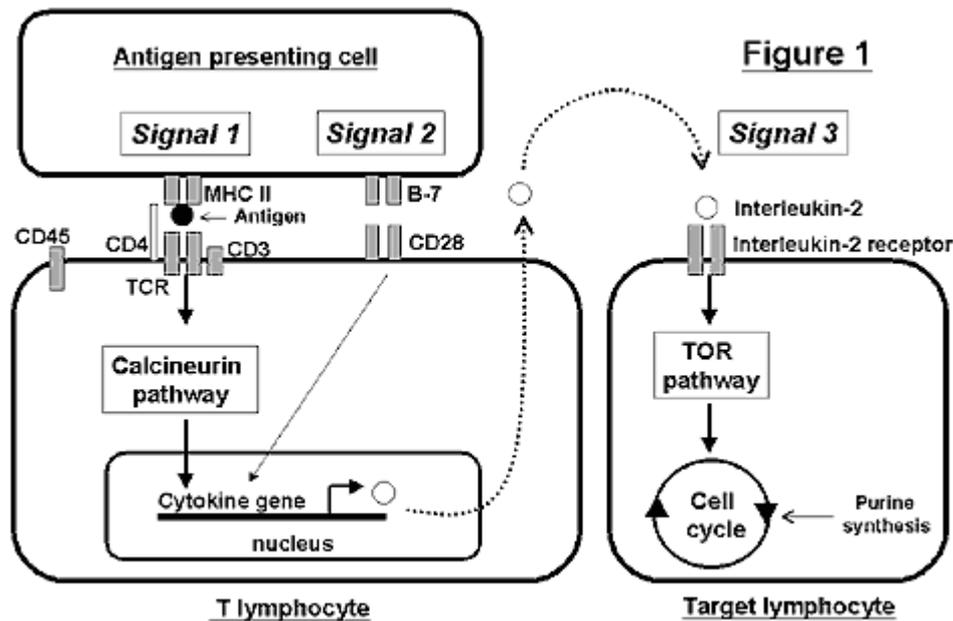


Figure 1. The 3-signal model of T cell activation. (for details, see text)

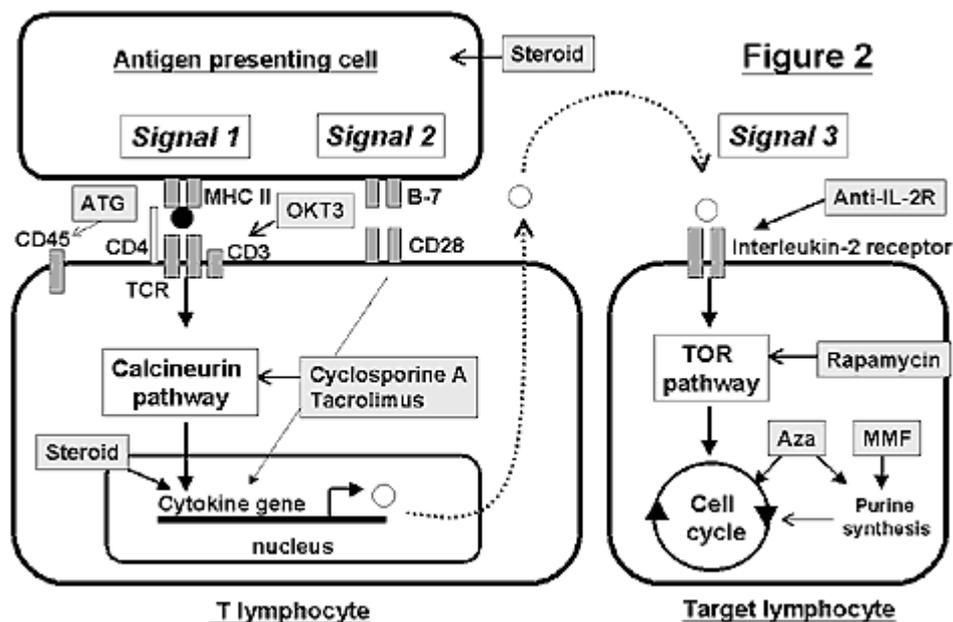


Figure 2. The main site of action of established immunosuppressive drugs. (Abbreviations: ATG: anti-thymocyte globulins; Aza: azathioprine; anti-IL2R: anti-interleukin 2 receptor monoclonal antibodies; MMF: Mycophenolate mofetil)

Table 1. Approved immunosuppressive drugs in clinical transplantation

Corticosteroids
Calcineurin inhibitors
Cyclosporine A
Tacrolimus
Anti-proliferative agents
Azathioprine
Mycophenolate mofetil
Rapamycin
Anti-lymphocyte polyclonal antibodies
ATGAM
Thymoglobulin
Anti-CD3 monoclonal antibodies
OKT3
Anti-interleukin 2 receptor monoclonal antibodies
Daclizumab
Basiliximab

Corticosteroids

Corticosteroids have been used to suppress inflammation and immune-mediated diseases for nearly half a century.

Corticosteroids possess both immunosuppressive and anti-inflammatory properties. Their pharmacological effects are complex and probably represent a wide range of effects on many phases of the immune and inflammatory responses such as inhibition of cytokine production, reduction of adhesion molecule expression, induction of lymphocytes apoptosis and suppression of inflammatory cell activation. Corticosteroids are used for both maintenance immunosuppression as well as for the treatment of acute rejection episodes. Side effects of corticosteroid therapy are numerous and some of them such as cataracts, osteoporosis and avascular necrosis of the femoral heads contribute significantly to the morbidity of transplant patients. Other side effects such as hypertension, hyperlipidemia and diabetes mellitus constitute important risk factors for the development of cardiovascular diseases.

Polyclonal Anti-lymphocyte Globulins and OKT3

ATGAM and thymoglobulin are polyclonal anti-lymphocyte globulins, derived from horse and rabbit respectively. The immunosuppressive effect of these polyclonal antibodies is mediated mainly through interacting with a variety of surface markers (e.g. CD45, CD3, CD4) on the lymphocytes. OKT3 is a murine monoclonal antibody against the CD3 complex of molecules on the surface of T lymphocytes. Polyclonal anti-lymphocyte globulins and OKT3 are mainly used for the treatment of severe acute rejection. They are also used for induction immunosuppression in sensitized patients and in patients with delayed graft function. As both polyclonal anti-lymphocyte antibodies and OKT3 significantly impairs cell-mediated immunity, patients receiving these antibodies are predisposed to opportunistic infections (especially CMV infections) and malignancies.

Cyclosporine A

Cyclosporine A, a cyclic 11 amino acid peptide, has been an essential component in most immunosuppressive protocols in transplantation for nearly 2 decades. Cyclosporine A binds to cyclophilin, a ubiquitous intracellular protein, to create the active drug complex. The cyclosporine-cyclophilin complex then inactivates calcineurin, a pivotal enzyme in the T cell receptor-signaling pathway. Calcineurin inhibition suppresses the transcription of pro-inflammatory cytokine genes (e.g. Interleukin-2, interferon- γ).

The conventional formulation of cyclosporine A has the disadvantage of variable solubility and dependency on bile for absorption. During the past few years, a new microemulsion formulation of cyclosporine A has been introduced, which is better absorbed and gives more consistent bioavailability.² The most important side effect of cyclosporine A is nephrotoxicity. Other significant side effects include hypertrichosis, gum hypertrophy, hypertension, hyperuricemia, hyperkalemia and hyperlipidemia. As cyclosporine A has a narrow therapeutic index, monitoring of cyclosporine blood levels is essential to minimize toxicity.

Tacrolimus

Tacrolimus is a macrolide antibiotic, which resembles cyclosporine A in its mechanism of action. Like cyclosporine A, tacrolimus binds to an immunophilin, FK-binding protein (FKBP) in the cytoplasm. The resultant tacrolimus-FKBP complex then interacts with calcineurin and inhibits its action in the same manner as cyclosporine A.

Tacrolimus has been used widely in liver transplantation since the early 1990s.³ Recent clinical trials in renal transplant patients have established that tacrolimus was equivalent to cyclosporine A in terms of patient and graft survival outcomes and might be superior in terms of prevention of acute rejection.⁴ Tacrolimus has also been used as rescue therapy for refractory rejection in renal transplant patients on cyclosporine-based therapy.⁵

The side effect profile of tacrolimus is slightly different from that of cyclosporine A. While both tacrolimus and cyclosporine A are equally nephrotoxic and can cause hyperkalemia and hyperuricemia, certain side effects such as gum hypertrophy, hirsutism and hyperlipidemia are more commonly observed with cyclosporine A whereas tremor and glucose intolerance are more commonly seen with tacrolimus.

Mycophenolate Mofetil

Mycophenolate mofetil has been developed as a replacement for azathioprine for maintenance immunosuppression. It acts by inhibiting inosine monophosphate dehydrogenase, a key enzyme in the de novo purine synthesis pathway, thereby limiting the proliferation of B and T lymphocytes. Three large multicenter clinical trials have established the efficacy of mycophenolate mofetil in addition to cyclosporine and steroids in the primary prevention of acute allograft rejection in renal transplant patients as compared to treatment with azathioprine or placebo.⁶ These studies show that mycophenolate mofetil significantly reduce the incidence of acute rejection in the first year after transplantation and the need for intensive immuno-suppression to treat rejection. Mycophenolate mofetil may also be useful in treating acute cellular rejection and reversing refractory acute rejection in renal transplant patients.⁷ There are experimental data to suggest that mycophenolate mofetil may help to prevent chronic allograft rejection. Mycophenolate mofetil has also been used in cardiac and liver transplantation with encouraging results.^{8,9}

The major side effects of MMF are gastrointestinal upset (particularly diarrhea), increased risk of tissue invasive cytomegalovirus infection, leukopenia and mild anemia. Mycophenolate mofetil does not cause hyperlipidemia, nephrotoxicity or hepatotoxicity.

Rapamycin

Rapamycin has recently been approved for the prophylaxis of rejection in kidney transplant patients in the United States. Rapamycin binds to the same immunophilin as tacrolimus, namely FKBP, to become active. It acts by inhibiting a key enzyme known as target of rapamycin (TOR), which is involved in the signal transduction pathway from cytokine/growth factor receptors to the nucleus to initiate cell cycling. This results in the inhibition of proliferation of activated lymphocytes. A phase III clinical trial comparing rapamycin to azathioprine for the reduction of acute rejection in renal transplant patients has recently been published.¹⁰ This study showed that the incidence of acute rejection was reduced in the rapamycin treated group. However, the rapamycin treated group also

showed evidence of increased cyclosporine A nephrotoxicity, presumably due to a drug interaction between rapamycin and cyclosporine A. The major side effects of rapamycin are hyperlipidemia and thrombocytopenia. Experimental data suggest that rapamycin may prevent the development of graft atherosclerosis, a hallmark of chronic rejection.

Anti-interleukin 2 Receptor Monoclonal Antibodies

Anti-interleukin 2 receptor monoclonal antibodies act by binding to a chain of the interleukin-2 receptors on activated T lymphocytes and render them unresponsive to interleukin-2, thereby preventing interleukin-2 induced clonal expansion of activated T lymphocytes. Anti-interleukin 2 monoclonal antibodies are used in the induction phase after renal transplantation. Two anti-interleukin 2 receptor monoclonal antibodies, namely daclizumab (fully humanized) and basiliximab (chimeric) have gone through phase III clinical trials in renal transplant recipients.¹¹ Both drugs (in combination with cyclosporine A) reduced the frequency of acute rejection by about one third. Anti-interleukin 2 receptor monoclonal antibodies are very well tolerated with virtually no side effects. The use of these antibodies has recently been extended to heart transplant patients.¹²

New Immunosuppressive Drugs

There are a number of new agents currently undergoing Phase I or II clinical trials, which are showing promise as future immunosuppressive agents in clinical transplantation. These include CTLA4Ig, a fusion protein that blocks the CD28 costimulatory pathway,¹³ anti-LFA-1 monoclonal antibodies which block adhesion molecules on the T cell surface,¹⁴ anti-CD4 monoclonal antibodies (OKT4) which disrupts the interaction between the T cell receptor and the MHC-antigenic peptide complex,¹⁵ FTY720, a synthetic myriocin analogue, which alters lymphocyte homing or trafficking pathways¹⁶ and allotrap, a synthetic peptides derived from MHC class I molecules with immunomodulatory properties.¹⁷

Future Trend in Immunosuppression

The availability of several new immunosuppressive drugs such as mycophenolate mofetil, anti-interleukin 2 receptor antibodies and rapamycin has increased the choice of immunosuppressive drugs for use in clinical transplantation. The question is which combination of drugs would give maximum efficacy and minimal toxicities. Several large multicenter trials are currently underway to address this issue. It is likely that no standard immunosuppressive protocol would fit all patients and that the prescription of immunosuppressive drugs for our patients would have to be individualized. Moreover, as the long-term use of corticosteroids and cyclosporine A is associated with significant side effects, the advent of these newer agents has allowed clinical trials to be conducted to test the possibility of early steroid withdrawal and reduction or total avoidance of cyclosporine A.¹⁸

The use of the newer immunosuppressive drugs has reduced the incidence of acute rejection after organ transplantation to a very low level. Moreover, some of these agents such as mycophenolate mofetil or rapamycin, have been shown in experimental models to be capable of preventing the development of chronic rejection. Whether these effects will translate into better long-term graft survival for the patients remain to be proven by prospective clinical trials with sufficient period of follow up.

The immunosuppressive drugs currently in use are still relatively non-specific and are therefore associated with significant immune and non-immune toxicities. The search for the "ideal" immunosuppressive drug continues. Ultimately, it is hoped that with better understanding of the immune response involved in the rejection of an transplanted organ, novel immuno-suppressive strategies can be developed which will modulate the host immune response in such way that long-term graft acceptance without the need for non-specific immunosuppression can become a reality.

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