**Immunosuppressants**

**I. OVERVIEW**

The importance of the immune system in protecting the body against harmful foreign molecules is well recognized. However, in some instances, this protection can result in serious problems. For example, the introduction of an allograft (that is, the graft of an organ or tissue from one individual to another who is not genetically identical) can elicit a dam- aging immune response, causing rejection of the transplanted tissue. Transplantation of organs and tissues (for example, kidney, heart, or bone marrow) has become routine due to improved surgical techniques and better tissue typing. Also, drugs are now available that more selectively inhibit rejection of transplanted tissues while preventing the patient from becoming immunologically compromised (Figure 40.1). Earlier drugs were nonselective, and patients frequently succumbed to infection due to suppression of both the antibody-mediated (humoral) and cell-mediated arms of the immune system. Today, the principal approach to immunosuppressive therapy is to alter lymphocyte function using drugs or antibodies against immune proteins. Because of their severe toxicities when used as monotherapy, a combination of immunosuppressive agents, usually at lower doses, is generally employed. [Note: Immunosuppressive therapy is also used in the treatment of auto immune diseases. For example, corticosteroids can control acute glomerulonephritis.] Immunosuppressive drug regimens usually consist of anywhere from two to four agents with different mechanisms of action that disrupt various levels of T-cell activation. The immune activation cascade can be described as a three-signal model. Signal 1 constitutes T-cell triggering at the CD3 receptor complex by an antigen on the surface of an antigen-presenting cell (APC). Signal 2, also referred to as costimulation, occurs when CD80 and CD86 on the sur- face of APCs engage CD28 on T cells. Both Signals 1 and 2 activate several intracellular signal transduction pathways, one of which is the calcium- calcineurin pathway, which is targeted by cyclosporine and tacrolimus. These pathways trigger the production of cytokines such as interleukin (IL)-2, IL-15, CD154, and CD25. IL-2 then binds to CD25 (also known as the IL-2 receptor) on the surface of other T cells to activate mammalian tar- get of rapamycin (mTOR), providing Signal 3, the stimulus for T-cell proliferation. Immunosuppressive drugs can be categorized according to their mechanisms of action: 1) Some agents interfere with cytokine produc- tion or action; 2) others disrupt cell metabolism, preventing lymphocyte proliferation; and 3) mono- and polyclonal antibodies block T-cell surface molecule

**II. SELECTIVE INHIBITORS OF CYTOKINE PRODUCTION AND FUNCTION**

Cytokines are soluble, antigen-nonspecific, signaling proteins that bind to cell surface receptors on a variety of cells. The term cytokine includes the molecules known as interleukins (ILs), interferons (IFNs), tumor necrosis factors (TNFs), transforming growth factors, and colony-stimulating factors. Of particular interest when discussing immunosuppressive drugs is IL-2, a growth factor that stimulates the proliferation of antigen-primed (helper) T cells, which subsequently produce more IL-2, IFN-γ, and TNF-α (Figure 40.2). These cytokines collectively activate natural killer cells, macrophages, and cytotoxic T lymphocytes. Clearly, drugs that interfere with the production or activity of IL-2, such as cyclosporine, will significantly dampen the immune response and, thereby, decrease graft rejection.

1. **Cyclosporine**

Cyclosporine [sye-kloe-SPOR-een] is a lipophilic cyclic polypeptide composed of 11 amino acids (several of the amino acids are methylated on the peptidyl nitrogen). The drug is extracted from the soil fun- gus Beauveria nivea. Cyclosporine is used to prevent rejection of kidney, liver, and cardiac allogeneic transplants. Cyclosporine is most effective in preventing acute rejection of transplanted organs when combined in a double-drug or triple-drug regimen with corticosteroids and an anti- metabolite such as mycophenolate mofetil. Cyclosporine is an alternative to methotrexate for the treatment of severe, active rheumatoid arthritis. It can also be used for patients with recalcitrant psoriasis that does not respond to other therapies, and it is also used for xerophthalmia.

**1. Mechanism of action**: Cyclosporine preferentially suppresses cell- mediated immune reactions, whereas humoral immunity is affected to a far lesser extent. After diffusing into the T cell, cyclosporine binds to a cyclophilin (more generally called an immunophilin) to form a complex that binds to calcineurin (Figure 40.3). The latter is responsible for dephosphorylating NFATc (cytosolic Nuclear Factor of Activated T cells). Because the cyclosporinecalcineurin complex cannot perform this reaction, NFATc cannot enter the nucleus to promote the reactions that are required for the synthesis of a number of cytokines, including IL-2. The end result is a decrease in IL-2, which is the primary chemical stimulus for increasing the number of T lymphocytes.

2**. Pharmacokinetics:** Cyclosporine may be given either orally or by intravenous (IV) infusion. Oral absorption is variable. Interpatient variability may be due to metabolism by a cytochrome P450 (CYP3A4) in the gastrointestinal (GI) tract, where the drug is metabo- lized. Cyclosporine is also a substrate for P-glycoprotein (P-gp), a drug efflux pump, which limits cyclosporine absorption by transporting the drug back into the gut lumen. About 50 percent of the drug is associated with the blood fraction. Half of this is in the erythrocytes, and less than one tenth is bound to the lymphocytes. Cyclosporine is extensively metabolized, primarily by hepatic CYP3A4. [Note: When other drug substrates for this enzyme are given concomitantly, many drug interactions have been reported.] It is not clear whether any of the 25 or more metabolites have any activity. Excretion of the metabolites is through the biliary route, with only a small fraction of the parent drug appearing in the urine.

**3. Adverse effects**: Many of the adverse effects caused by cyclosporine are dose dependent. Therefore, it is important to monitor blood evels of the drug. Nephrotoxicity is the most common and important adverse effect of cyclosporine, and it is critical to monitor kidney function. Reduction of the cyclosporine dosage can result in reversal of nephrotoxicity in most cases, although nephrotoxicity may be irreversible in 15 percent of patients. [Note: Coadministration of drugs that also can cause kidney dysfunction (for example, the aminoglycoside antibiotics) and anti-inflammatories, such as diclofenac, naproxen, or sulindac, can potentiate the nephrotoxicity of cyclosporine. Because hepatotoxicity can also occur, liver function should be periodically assessed.] Infections in patients taking cyclosporine are common and may be life-threatening. Viral infections due to the herpes group and cytomegalovirus (CMV) are prevalent. Lymphoma may occur in all transplanted patients due to the net level of immunosuppression and has not been linked to any one particular agent. Anaphylactic reactions can occur on parenteral administration. Other toxicities include hypertension, hyperlipidemia, hyperkalemia (it is important not to use K+-sparing diuretics in these patients), tremor, hirsutism, glucose intolerance, and gum hyperplasia.

**B. Tacrolimus**

Tacrolimus [ta-CRAW-lih-mus] (originally called FK506) is a macrolide that is isolated from the soil fungus Streptomyces tsukubaensis. Tacrolimus is approved for the prevention of rejection of liver and kid- ney transplants and is given with a corticosteroid and/or an antimetabolite. This drug has found favor over cyclosporine, not only because of its potency and decreased episodes of rejection (Figure 40.4), but also because lower doses of corticosteroids can be used, thus reducing the likelihood of steroid-associated adverse effects. An ointment preparation has been approved for moderate to severe atopic dermatitis that does not respond to conventional therapies.

**1. Mechanism of action**: Tacrolimus exerts its immunosuppressive effect in the same manner as cyclosporine, except that it binds to a dif- ferent immunophilin, FKBP-12 (FK-binding protein; see Figure 40.3).

**2. Pharmacokinetics**: Tacrolimus may be administered orally or IV. The oral route is preferable, but, as with cyclosporine, oral absorption of tacrolimus is incomplete and variable, requiring tailoring of doses. Tacrolimus is subject to gut metabolism by CYP3A4/5 isoenzymes and is a substrate for P-gp. Together, both of these mechanisms limit the oral bioavailability of tacrolimus. Absorption is decreased if the drug is taken with high-fat or high-carbohydrate meals. Tacrolimus is from 10- to 100-fold more potent than cyclosporine. It is highly bound to serum proteins and is also concentrated in erythrocytes. Like cyclosporine, tacrolimus undergoes hepatic metabolism by the CYP3A4/5 isozyme, and the same drug interactions occur. At least one metabolite of tacrolimus has been shown to have immunosup- pressive activity. Renal excretion is very low, and most of the drug and its metabolites are found in the feces.

**3. Adverse effects**: Nephrotoxicity and neurotoxicity (tremor, seizures, and hallucinations) tend to be more severe in patients who are treated with tacrolimus than in patients treated with cyclosporine, but careful dose adjustment can minimize this problem. Development of posttransplant, insulin-dependent diabetes mellitus is a prob- lem, especially in black and Hispanic patients. Other toxicities are the same as those for cyclosporine, except that tacrolimus does not cause hirsutism or gingival hyperplasia. Compared with cyclosporine, tacrolimus has also been found to have a lower incidence of cardio- vascular toxicities, such as hypertension and hyperlipidemia, both of which are common disease states found in kidney transplant recipients. Anaphylactoid reactions to the injection vehicle have been reported. The drug interactions are the same as those described for cyclosporine.

**C. Sirolimus**

Sirolimus [sih-ROW-lih-mus] is a macrolide obtained from fermentations of the soil mold Streptomyces hygroscopicus. The earlier name, and one that is sometimes still used, is rapamycin. Sirolimus is approved for use in renal transplant ation, to be used together with cyclosporine and corticosteroids, allowing lower doses of those medications to be used, thereby lowering their toxic potential. The combination of sirolimus and cyclosporine is apparently synergistic because sirolimus works later in the immune activation cascade. To limit the long-term side effects of the calcineurin inhibitor, sirolimus is often used in calcineurin inhibitor withdrawal protocols in patients who remain rejection free during the first 3 months posttransplant. The antiproliferative action of sirolimus has found use in cardiology. Sirolimus-coated stents inserted into the cardiac vasculature inhibit restenosis of the blood vessels by reducing proliferation of the endothelial cells. In addition to its immunosuppres- sive effects, sirolimus also inhibits proliferation of cells in the graft inti- mal areas and, thus, is effective in halting graft vascular disease.

**1. Mechanism of action**: Sirolimus and tacrolimus bind to the same cytoplasmic FK-binding protein, but instead of forming a complex with calcineurin, sirolimus binds to mTOR, interfering with Signal 3. The latter is a serine-threonine kinase. [Note: TOR proteins are essential for many cellular functions, such as cell-cycle progression, DNA repair, and as regulators involved in protein translation.] Binding of sirolimus to mTOR blocks the progression of activated T cells from the G1 to the S phase of the cell cycle and, consequently, the proliferation of these cells (see Figure 40.5). Unlike cyclosporine and tacrolimus, sirolimus does not owe its effect to lowering IL-2 pro- duction but, rather, to inhibiting the cellular responses to IL-2.

**2. Pharmacokinetics**: The drug is available only as oral preparations. Although it is readily absorbed, high-fat meals can decrease the drug’s absorption. Sirolimus has a long half-life (57 to 62 hours) com- pared to those of cyclosporine and tacrolimus, and a loading dose is recommended at the time of initiation of therapy, but only requires once daily dosing. Like both cyclosporine and tacrolimus, sirolimus is metabolized by the CYP3A4 isozyme and interacts with the same drugs as do cyclosporine and tacrolimus. Sirolimus also increases the drug concentrations of cyclosporine, and careful blood level monitoring of both agents must be done to avoid harmful drug toxicities. The parent drug and its metabolites are predominantly eliminated in feces.

**3. Adverse effects**: A common side effect of sirolimus is hyperlipidemia (elevated cholesterol and triglycerides), which can require treatment. The combination of cyclosporine and sirolimus is more nephrotoxic than cyclosporine alone due to the drug interaction between the two, necessitating lower doses. Although the administration of sirolimus and tacrolimus appears to be less nephrotoxic, sirolimus can still potentiate the nephrotoxicity of tacrolimus, and drug levels of both must be monitored closely. Other untoward problems are headache, nausea and diarrhea, leukopenia, and thrombocytopenia. Impaired wound healing has been noted with sirolimus in obese patients and those with diabetes, which can be especially problematic imme- diately following the transplant surgery and in patients receiving corticosteroids.

**D. Everolimus**

Everolimus [e-ve-RO-li-mus] (another mTOR inhibitor) was recently approved by the U.S. Food and Drug Administration for use in renal transplantation in combination with low-dose cyclosporine and corti- costeroids. It was originally approved in 2009 for second-line treatment in patients with advanced renal cell carcinoma.

**1. Mechanism of action**: Everolimus has the same mechanism of action as sirolimus. It inhibits activation of T cells by forming a complex with FKBP-12 and subsequently blocking mTOR.

**2. Pharmacokinetics**: Everolimus differs from sirolimus in its pharma- cokinetic profile. Everolimus is rapidly absorbed, attaining maximal concentrations in 1 to 2 hours post dose, but absorption is decreased with high-fat meals. Everolimus is a substrate of CYP3A4 and P-gp and, thus, is subject to the same drug interactions as previously mentioned immunosuppressants. Everolimus avidly binds erythocytes, and monitoring of whole blood trough concentrations is recommended. It has a much shorter half-life than does sirolimus at 30 ± 11 hours and requires twice-daily dosing. Everolimus increases drug concentrations of cyclosporine, thereby enhancing the nephrotoxic effects of cyclosporine, and is, therefore, recommended to be used with reduced doses of cyclosporine.

**3. Adverse effects**: Everolimus has similar side effects to sirolimus, includ- ing hyperlipidemia, impaired or delayed wound healing following transplantation, and enhanced nephrotoxicity in combination with higher doses of cyclosporine. An additional adverse effect noted with everolimus is angioedema, which may increase with concomitant use of angiotensin-converting enzyme inhibitors. There is also an increased risk of kidney arterial and venous thrombosis, resulting in graft loss, usually in the first 30 days posttransplantation.