Immunosuppressive Therapy and Protocols

Angelo M. de Mattos

The 1990s have seen major steps in the dissection of basic mechanisms of allorecognition, and renal graft survival has achieved unprecedented clinical results. Transplantation has turned into a widespread modality of therapy for patients with chronic renal failure that benefits thousands worldwide. Combinations of immunosuppressive agents have proved to be an effective strategy to inhibit diverse pathways of the multifaceted immune system, allowing the reduction of both dosage and adverse effects of each individual drug. As understanding of the molecular basis of the immune response has expanded rapidly, so have the possibilities for designing therapeutic interventions that are more effective, more specific, and safer than are current treatment options. As we reach the end of the century, several different and innovative approaches will add to this fascinating and complex therapy.

CHAPTER

11

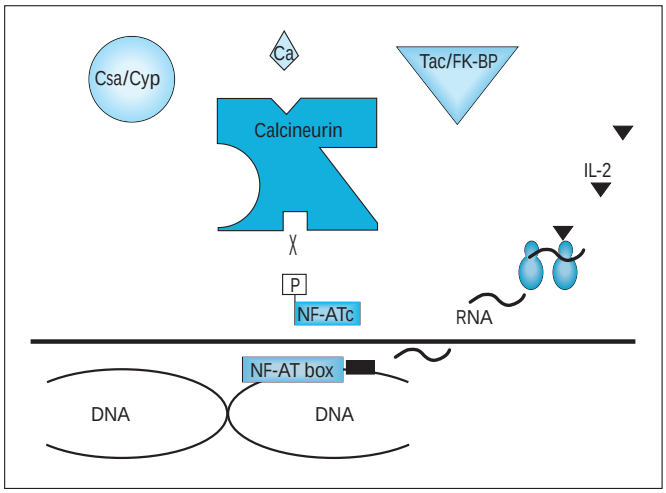


FIGURE 11-1

Mechanism of action for cyclosporine (Csa) and tacrolimus (Tac). The common cytoplasmic target for cyclosporine and tacrolimus is calcineurin. After binding to cyclophillin (Cyp), cyclosporine interacts with calcineurin, inhibiting its catalytic domain. Thus dephosphorylation of transcription factors is prevented, as exemplified by the nuclear factor of activated T lymphocyte (NF-AT). Despite having a different ligand called FK-binding protein (FK-BP), tacrolimus inhibits calcineurin in a similar way. Because phosphorylated transcription factors cannot cross the nuclear membrane, the production of key factors for lymphocyte activation and proliferation (*ie*, interleukin-2, tumor necrosis factor-α, γ interferon, c-myc, and others) is inhibited [1]. NF-ATc—nuclear factor of activated T-lymphocyte-cytoplasmic form; P—phosphorus; Ca—calcium.

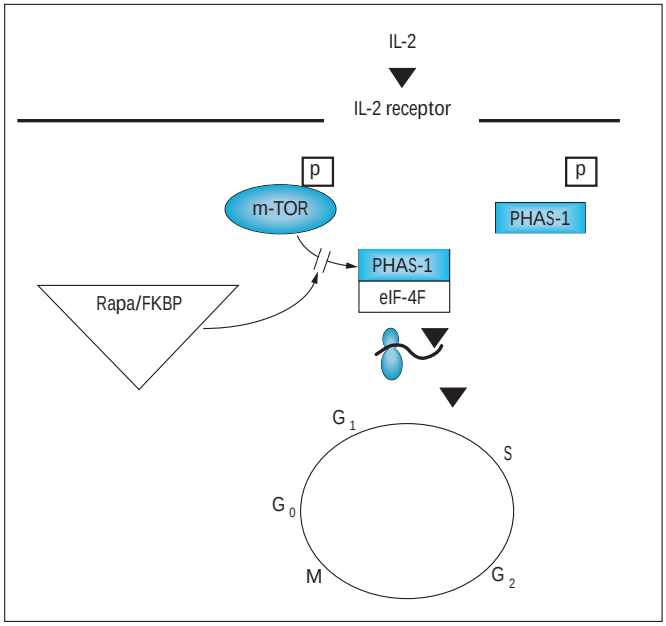


FIGURE 11-2

Proposed mechanism of action for rapamycin (rapa). Rapamycin binds to FK-binding protein (FK-BP). However, the immunosuppressive properties of rapamycin are not due to inhibition of calcineurin. Rapamycin blocks the activating signal delivered by growth factors (exemplified by the interleukin-2 [IL-2] receptor) by blocking the translation of the coding of messenger RNA (mRNA) for key proteins required for progression through the G_1 phase of the cell cycle. In this model the mammalian target of rapamycin (m-TOR, also called FRAP or RAFT1), phosphorylates the translational repressor PHAS-I. Arrest of the cell cycle results, and the proliferation of lymphocytes is thereby inhibited. The full understanding of the mechanism(s) of action of rapamycin is the focus of intense research at this time [2]. elF-4—translation initiation factor belonging to the Ets family; $G_{(0,1,\text{ and }2)}$—quiescent; M—mitosis; S—synthesis.

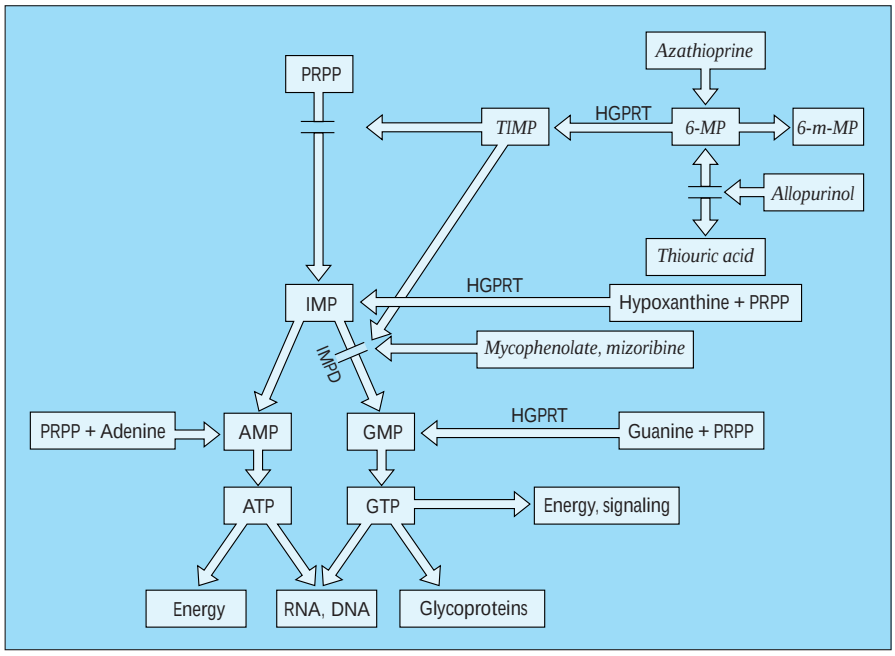


FIGURE 11-3

Mechanism of immunosuppression of azathioprine and mycophenolate mofetil (MMF). Azathioprine and MMF prevent lymphocyte proliferation by way of inhibition of purine base synthesis, thus resulting in decreased production of the building blocks of nucleic acids (*ie*, DNA and RNA). Azathioprine is metabolized to 6-mercaptopurine (6-MP), which is further converted to 6-ionosine monophosphate. This molecule inhibits key enzymes in the *de novo* pathway of purine synthesis (adenosine monophosphate [AMP] and guanosine monophosphate [GMP]). MMF is metabolized to mycophenolic acid, which is a noncompetitive inhibitor of the enzyme that converts inosine monophosphate (IMP) to GMP. The depletion of GMP may have effects other than inhibition of nucleic acid production. Some events of T-lymphocyte activation are independent of guanosine triphosphate (GTP), as is the assembling of certain adhesion molecules. ATP—adenosine triphosphate; HGPRT—hypoxanthine-guanine phosphoribosyl transferase; IMPD—inosine-monophosphate dehydrogenase; PRPP—phosphoribosyl pyrophosphate; 6-m-MP—6-methyl-mercaptopurine; TIMP—thioinosine monophosphate. (*Adapted from* de Mattos and coworkers [3,4].)

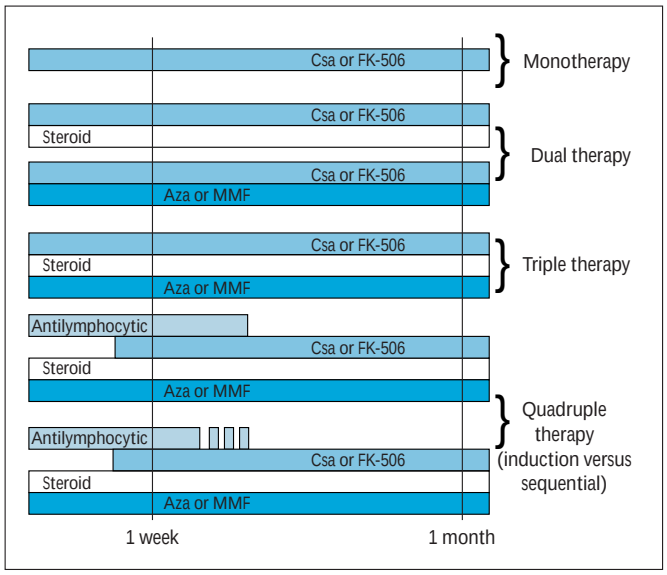
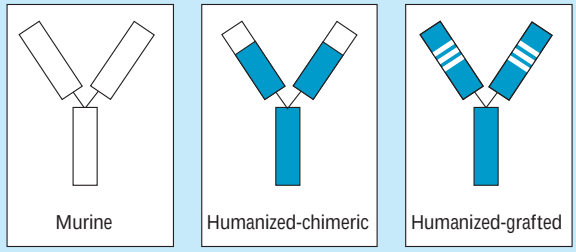


FIGURE 11-4

Summary of strategies for combining immunosuppressive agents. Currently, monotherapy (usually cyclosporine [Csa]) is not used in the United States. Dual therapy (involving cyclosporine or tacrolimus) is used commonly in Europe. Most centers in the United States use triple or quadruple therapy (induction or sequential). Some centers continue the induction with the antilymphocytic biologic agent for a predetermined period (usually 10–14 days), overlapping with the initiation of cyclosporine (or tacrolimus). Alternatively, the biologic agent is discontinued and cyclosporine (or tacrolimus) begun as soon as the graft function reaches a determined threshold, resulting in no overlap of these two agents. In living donor transplants, azathioprine (Aza) is commonly begun a few days before surgery. [5]. FK-506—tacrolimus; MMF—mycophenolate mofetil.

A



B

Monoclonal antibody	Type	Target
Muromonab OKT3	Murine	CD3
Anti-Tac	Murine	IL-2R (CD25)
SDZ-CHIB	Murine/Human	IL-2R (CD25)
T10B9	Murine	TCR
BMA 031	Murine	TCR
WT 32	Murine	CD3
Anti-ICAM 1	Murine	CD54
33B3-1	Rat	IL-2R (CD25)

FIGURE 11-5

Evolution of monoclonal antilymphocytic antibodies. Monoclonal antibodies are the result of complex genetic engineering techniques. **A**, Differences among murine, chimeric, and "humanized" antibodies. Attempts to reduce side effects, improve efficacy, and decrease xenosensitization are the main reasons for development of these modifications on the murine molecule. **B**, The different monoclonal antibodies, their classification regarding the molecular structure, and their targets. Muromonab OKT3 (Ortho Pharmaceutical, Raritan, NJ) is the only monoclonal antibody commercially available at this time [6]. CD3— monomorphic membrane co-receptor present in T-lymphocytes; IL-2R—interleukin-2R; TCR—T-cell receptor.

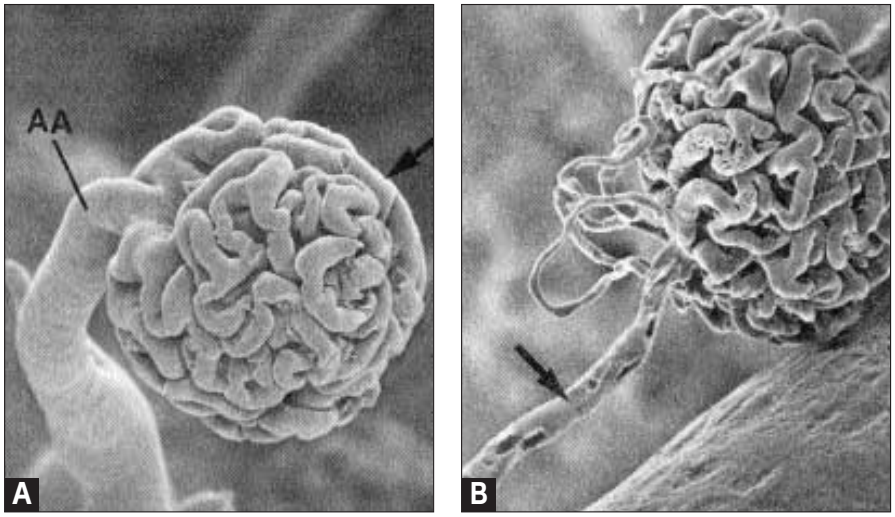


FIGURE 11-6

Experimental model of the vasoconstrictive effect of cyclosporine. Some of the acute nephrotoxicity of cyclosporine is due to the significant yet reversible vasoconstrictive effect of the drug. **A**, Scanning electron micrograph of glomerulus of a rat not exposed to cyclosporine. *Arrow* indicates glomerular capillary loop. AA—afferent artery. **B**, After 14 days of cyclosporine treatment, the entire length of an afferent arteriole shows narrowing (magnification × 500). *Arrow* indicates afferent artery. (*From* English and coworkers [7]; with permission.)

AGENTS USED IN RENAL TRANSPLANTATION

Drug	Dosage	Adverse reactions	Cost
Cyclosporine Sandimmune (Sandoz Pharmaceuticals, East Hanover, NJ)	Starting dose: 7–10 mg/kg/d in 2 divided doses Maintenance: based on blood levels	Nephrotoxicity, hypertension, gingival overgrowth, hirsutism, hepatotoxicity, neurotoxicity, hypomagnesia, hyperkalemia	Gelcaps: $1.61/25 mg; $6.42/100 mg Liquid: $6.41/100 mg, orally
Neoral (Sandoz Pharmaceuticals, East Hanover, NJ)	Starting dose: 7–10 mg/kg/d in 2 divided doses Maintenance: based on blood levels	Same	Gelcaps: $1.44/25 mg; $5.77/100 mg Liquid: $6.38/100 mg, orally
	IV Csa equals one third of oral Csa; IV cyclosporine is given by continuous infusion over 24 h		$113.32/100 mg, IV
Azathioprine Imuran (Glaxo Wellcome, Research Triangle Park, NC)	Starting and maintenance dose: 1–3 mg/kg/d; IV dose equals half of oral dose Decrease dose by half for 50% decrease in leukocyte count	Leukopenia, anemia, thrombocytopenia, hepatitis, pancreatitis, alopecia, skin cancer, aplastic anemia (rare)	$1.29/50-mg tablet $101.18/100-mg vial, IV
Azathioprine (Roxane Laboratories, Columbus, OH)	Hold dose for leukocyte count of <3000		$1.16/50-mg tablet
Azathioprine sodium (injectable) (Bedford Laboratories, Bedford, OH)			$81.60/100-mg vial, IV
OKT3 (Ortho Pharmaceutical, Raritan, NJ) Muromonab-cd3	Induction: 2 mg/d (low-dose) 5 mg/d (standard) Rejection treatment: 5 mg/d Hold (delay) dose for weight gain >3% or temperature >39°C Increase dose based on CD3+ cell count and CD3 density (suggested) Discontinue if anti-OKT3 antibody titer >1:1000	Cytokine release syndrome: fever, chills, chest pain, dyspnea, wheezing, noncardiogenic pulmonary edema, nausea, vomiting, diarrhea, headache, aseptic meningitis, seizures, skin rash	$672.00/5-mg vial
Antithymocyte globulin Atgam (Upjohn Co, Kalamazoo, MI)	Starting dose: 15–30 mg/kg/d Decrease (or hold) dose for leukocytes <3000 or platelets <100,000	Leukopenia, thrombocytopenia, fever, chills, skin rash, back pain, headache, nausea, vomiting, diarrhea, horse serum sickness	$262.24/250-mg vial
Prednisone (various manufacturers) Deltasone (Upjohn Co, Kalamazoo, MI)	Starting dose: 500 to 1000-mg infusion for 3–5 d Maintenance: taper schedule (variable)	Fat redistribution, increased appetite, weight gain, hyperlipidemia, hypertension, peripheral edema, hyperglycemia, skin atrophy, poor healing, acne, night sweats, insomnia, mood changes, blurred vision, cataracts glaucoma, osteoporosis	$0.02–$0.05/5-mg tablet Methylprednisolone, IV $17.88–$35.50/500-mg vial
FK-506, tacrolimus Prograf (Fujisawa USA, Inc, Deerfield, IL)	Starting dose: 0.15–0.3 mg/kg/d in 2 divided doses Avoid IV (0.05–0.1 mg/kg/d as a continuous infusion over 24 h) Maintenance: based on blood levels	Nephrotoxicity, hypertension, hepatotoxicity, pancreatitis, diabetes, seizures, headache, insomnia, tremor, paresthesia	$2.39/1-mg caplet $11.97/5-mg caplet $222.00/5-mg ampule, IV
Mycophenolate mofetil CellCept (Roche Laboratories, Nutley, NJ)	Starting dose: 2–3 g/d orally in 2 divided doses (IV preparation in clinical trials) Maintenance: based on GI and bone marrow toxicities	Nausea, vomiting, diarrhea, leukopenia, anemia, thrombocytopenia	$2.04/250-mg caplet $4.08/500-mg tablet $102.00/500-mg, IV
Daclizumab (Roche Laboratories, Nutley, NJ)	1 mg/kg/d every 2 wk for a total of 5 doses	Reported same as placebo	$418.20/25 mg, IV
Simulect (Novartis Pharmaceuticals Inc., East Hanover, NJ)	20 mg/d, given on days 0 and 4 post transplant	Reported same as placebo	$1224.00/20mg, IV

Cost to the pharmacist based on the average wholesale price listing in *Red Book*, 1997 [8].

CD3—monomorphic membrane co-receptor present in T-lymphocytes; Csa—cyclosporine; GI—gastrointestinal.

Adapted from de Mattos and coworkers [3,4].

FIGURE 11-7

A summary of the immunosuppressive agents currently used in human renal transplantation is given. Dosages and costs are subject to local variation.

CLINICALLY RELEVANT DRUG INTERACTIONS WITH IMMUNOSUPPRESSIVE DRUGS

Drug	Effect	Mechanism
Cyclosporin A and tacrolimus		
Diltiazem	Increased blood levels	Decreased metabolism (inhibition of cytochrome P-450-IIIA 4)
Nicardipine		
Verapamil		
Erythromycin	Increased blood levels	Decreased metabolism (inhibition of cytochrome P-450-IIIA 4)
Clarithromycin		
Ketoconazole	Increased blood levels	Decreased metabolism (inhibition of cytochrome P-450-IIIA 4)
Fluconazole		
Itraconazole		
Methylprednisolone (high dose only)	Increased blood levels	Unknown
Carbamazepine	Decreased blood levels	Increased metabolism (inhibition of cytochrome P-450-IIIA 4)
Phenobarbital		
Phenytoin		
Rifampin		
Aminoglycosides	Increased renal dysfunction	Additive nephrotoxicity
Amphotericin B		
Cimetidine	Increased serum creatinine	Competition for tubular secretion
Lovastatin	Decreased metabolism	Myositis, increased creatine phosphokinase, rhabdomyolysis
Azathioprine		
Allopurinol	Increased bone marrow toxicity	Inhibiting xantine oxidase
Warfarin	Decreased anticoagulation effect	Increased prothrombin synthesis or activity
ACE inhibitors	Increased bone marrow toxicity	Not established
Mycophenolate mofetil		
Acyclovir-ganciclovir (high doses only)	Increased levels of acyclovir-ganciclovir and mycophenolate mofetil	Competition for tubular secretion
Antiacids	Decreased absorption	Binding to mycophenolate mofetil
Cholestyramine	Decreased absorption	Interferes with enterohepatic circulation

ACE—angiotensin-converting enzyme.
Adapted from de Mattos and coworkers [3,4].

FIGURE 11-8

Clinical relevant drug interactions with immunosuppressive agents. Close monitoring of drug levels is required periodically with concomitant use of drugs with potential interaction. Drug level monitoring is clinically available for cyclosporin A and tacrolimus. Monitoring of non-immunosuppressive drug level is also important when used with potential interacting immunosuppressive agents.

NEW IMMUNOSUPPRESSIVE AGENTS UNDERGOING CLINICAL TRIALS

Agent	Mechanism of action
Rapamycin	Inhibition of cytokine action (downstream of interleukin-2 receptor and other growth factors)
Leflunomide	Inhibition of cytokine action (expression of or signaling by way of interleukin-2 receptor)
	Inhibition of DNA and RNA synthesis (pyrimidine pathway)
Brequinar	Inhibition of DNA and RNA synthesis (pyrimidine pathway)
Deoxyspergualin	Unknown (related to heat-shock proteins?)
SKF-105685	Unknown (stimulation of suppressor cells?)
Mizoribine	Inhibition of DNA and RNA synthesis (*de novo* purine pathway)
CTLA-4Ig	Blockage of T-cell co-stimulatory pathway

FIGURE 11-9
Proposed mechanisms of action of new immunosuppressive drugs currently undergoing clinical or preclinical trials in organ transplantation [9].

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