LEADING ARTICLE

Drugs 2002; 62 (11): 1587-1597
0012-6667/02/0011-1587/$30.00/0

Single-Agent Immunosuppression After Liver Transplantation

What is Possible?

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Abstract

Orthotopic liver transplantation is a life saving and life enhancing procedure. The development of immunosuppressive drugs has contributed to the high rate of success in terms of both patient and graft survival. However, the considerable adverse effects of these therapies are affecting long-term outcomes of transplant recipients. Complications related to immunosuppression are responsible for the majority of deaths in patients surviving more than 1 year.

Therefore, the search for an optimal immunosuppressive regimen has become of paramount importance. The liver has proved to be an 'immunologically privileged' organ, capable in several animal models to be accepted as an allograft without any intervention on the immune system of the recipient. In some human liver allografts acceptance of the new organ is recognised after withdrawal of immunosuppressants, but prior identification of such individuals is not yet possible, thus negating this management option.

Graft-recipient interaction is peculiar in liver transplantation: acute cellular rejection does not always need to be treated, and if it is not severe, appears to be associated with a better survival of both patient and graft.

In the last decade there has been an evolution of immunosuppressive protocols, driven by empirical observation and a deeper understanding of immunological events after transplant. However, most modifications have been made because of the necessity to reduce long-term drug related morbidity and mortality.

Withdrawal of corticosteroids has proven to be safely achievable in most patients, with no deleterious effects on patient or graft survival but with a great benefit in terms of reduction of incidence of metabolic and cardiovascular complications. Long-term 'steroid-free' regimens are therefore now widely used.

Patients with stable graft function can be easily maintained using a single drug usually after 6 or 12 months and usually with a calcineurin inhibitor. The more evolved step of using monotherapy *ab initio* has also proven to be effective in a few studies and needs to be explored further.

In the future new strategies will be designed to help the development of tolerance of the allograft, selectively stimulating instead of suppressing the immune reaction of the recipient.

During the last 2 decades orthotopic liver transplantation (OLT) has become the standard therapy for both acute and chronic liver failure of all aetiologies. The use of immunosuppressive agents, without any doubt, has played a crucial role in the establishment of this technique. Calcineurin inhibitors (CNI) cyclosporin and tacrolimus are the keystones of immunosuppressive regimens used in different centres worldwide.

Nowadays most patients and liver grafts survive beyond the perioperative period, achieving 1-year survival rates of 70 to 90% and 10-year survival of 62%;[1] thus shifting attention to the morbidity and mortality associated with long-term immunosuppression.

In every field of transplantation the main objective has been the induction of graft acceptance in the recipient by achieving control of the immune response, trying to prevent rejection and subsequent organ loss. This has been pursued with the knowledge of the frequent and sometimes potentially unfavourable adverse effects of immunosuppression.

However, within this broad panorama, liver transplantation has several unique features that make it different from transplantation of other solid organs. In contrast to what happens for example to a renal allograft (for which acute rejection is associated with graft loss), it is now accepted that the occurrence of early acute liver graft rejection (and its successful treatment when needed) does not affect patient or graft survival following liver transplantation.[2] Furthermore, mild acute rejection has been reported to be associated with increased patient survival and a trend towards improved graft survival.[3] It could be hypothesised that the occurrence of acute rejection in the setting of controlled alloreactivity exerts a tolerising effect, shielding the graft from further immunological attack.

It is also well recognised that not all liver transplant recipients have an equal propensity to experience acute rejection and predictive risk factors have been described,[3,4] allowing considerable modulation, which so far has not been possible with other solid organ transplants.

In addition, liver transplantation is associated with a high risk of early infections, particularly bacterial ones, which are still a major cause of early death post transplant. This appears to be a consequence of inherent reduction of antibacterial defences, in both acute and chronic liver failure, surgical manipulation of gut and biliary tree, frequent invasive diagnostic procedures after the transplant, and the risk of chest infections as a result of the frequent collapse of lobar segment secondary to diaphragmatic splinting. Logically the 'weakened' host immune system is the ideal playground for bacteria, viruses and a wide spectrum of opportunistic pathogens.

As a consequence of the improved short-term survival (due to refinement of surgical techniques and better postoperative management), long-term outcome of patients who have had a liver transplant is becoming the main concern for clinicians who have to deal with the long-term adverse effects of immunosuppressant drugs. During long-term follow up, chronic rejection is currently responsible only for a small proportion of graft loss and some deaths, whereas immunosuppression-related complications as infections, cardiovascular disease, renal failure and *de novo* malignancy account for the most of the cases,[1] particularly when recurrence of primary disease is low such as in non-viral related disease.

Our aim in this short review is to illustrate what has already been done in modulating immunosuppression for liver graft recipients. The potential advantages of less potent but still 'safe' immunosuppressive regimens has led several transplant centres worldwide to use regimens which end up as a single drug, usually reported in carefully studied cohorts of patients in whom corticosteroids were withdrawn some months after transplantation. Only one trial from our centre[5] has compared tacrolimus and cyclosporin as monotherapy *ab initio*, avoiding corticosteroids and other immunosuppressive agents for induction or maintenance.

1. Weaning off Immunosuppression

The 'Eldorado' of liver transplant clinicians is the acceptance of the graft by the recipient without

any long-term pharmacological help. However, although there have been small series of human liver grafts not requiring long-life immunosuppression,[6-8] it is impossible to predict who these individuals will be and the consequences of late rejection are worse than those of early cellular rejection and include death.

In series reporting the attempt to stop anti-rejection treatment (for major drug adverse effects, lack of compliance or by intention in small prospective studies) in patients with stable graft function, complete 'freedom' was achieved in nearly a third of patients.

Takatsuki et al.[8] described a cohort of 63 living donor liver transplant recipients receiving tacrolimus. Twenty six patients were entered in an elective program of withdrawal, while in the remaining 37 the choice was mainly as a result of serious immunosuppression complications, mostly Epstein-Barr-associated post transplant lymphoproliferative disease (30 patients). Criteria for elective withdrawal were an interval of more than 2 years after the transplant with good graft function and no episodes of rejection in the previous 12 months: patients were gradually weaned off tacrolimus. In 24 patients (38.1%, six of them from the elective cohort) tacrolimus could be stopped with a median drug-free period of 23.5 months; 23 patients (36.5%) were still undergoing the weaning process at the time of publication. Rejection occurred in 16 patients (25.4%) after a median interval of 9.5 months (with a range of 1 to 63 months), but all the episodes could be treated reintroducing tacrolimus or with short courses of corticosteroids.

Devlin et al.[6] published a small series including 18 patients in whom immunosuppressants had to be withdrawn because of major adverse effects or immunosuppression-related disorders, with a minimum follow up of 3 years. OLT had been performed at least 5 years before weaning was started and all grafts were functioning; all patients were receiving cyclosporin and azathioprine and 15 of them also corticosteroids. Five patients remained drug-free with normal histological findings in the follow up biopsies; one patient with post-transplant lymphoproliferative and biochemical derangements underwent retransplantation. In the remaining 12 patients, immunosuppression was reinitiated at a median of 3 weeks after initial withdrawal following a flare up in liver function tests; histology showed acute cellular graft rejection in four and an hepatitis-like disorder in eight, with an exacerbation of features already seen in the baseline biopsy in five of these eight. Weaning was again attempted and nine patients were maintained on dosages otherwise considered subtherapeutic.

The paper by Mazariegos[7] reports the experience from Pittsburgh University. They first observed five patients who stopped immunosuppressant therapy because of non-compliance: three remained well at 14, 15 and 17 years after stopping drugs, one was killed in a road accident after 11 years off immunosuppression and the last patient underwent retransplantation because of recurrent hepatitis C infection after 9 drug-free years (in both the last patients there was no histological evidence of rejection). These data supported a prospective drug withdrawal program including 95 patients who were >5 years post OLT, with no episodes of rejection in the previous 2 years and in the baseline biopsy, and without vascular or biliary complications or recurrence of original disease. Weaning was gradual; 18 patients (19%) had been off drugs with a median follow up of 35.5 months, 37 patients were still in the weaning process. Rejection was histologically documented in 21 patients (with chronic rejection suspected in three) and clinically, without biopsy, in further seven patients: all of them were restarted on their previous drugs. Twelve patients were excluded because of non-compliance,[8] recurrent primary biliary cirrhosis (PBC),[2] renal failure and pregnancy (one each). Patients receiving cyclosporin-based regimens had a significantly lower rate of success for complete drug withdrawal at 1-year post-weaning. Interestingly, the drug-free patients failed to show a significant improvement in renal function or hypertension.

Taking all reports into account the data on empirical withdrawal does not make this a current nor

viable management strategy, as individual response cannot be predicted. No data are available as to whether human leucocyte antigen (HLA) matching or lymphotoxicity assays might help in identifying those who remain well without immunosuppression.

Some of the authors[6] have suggested that the primary disease leading to transplantation could exert a strong influence on the capacity of the recipient to overcome immunosuppression: patients affected by immunological or viral disease would be more likely to experience graft dysfunction.

At the moment a realistic goal is the reduction of stable immunosuppressive regimen to the minimum required to prevent rejection, carefully balancing the gain in "protective effect" with the unavoidable price to pay in terms of complications. In most centres patients are usually started on a double- or triple-drug regimen comprising cyclosporin or tacrolimus plus corticosteroids and azathioprine, or recently mycophenolate mofetil substituted for azathioprine.

The great majority of episodes of acute cellular rejection occur in the first weeks after transplant, so this is thought to be the time frame when a more vigorous effort is required to preserve the graft. However, in the early peri- and postoperative period these patients are particularly prone to the risk of infectious complications carrying high morbidity and mortality rates.

We believe that a substantial percentage of liver transplant recipients are over-immunosuppressed. The earlier protocols, translated from experience in renal transplantation, did not reflect the way immunosuppression is managed for OLT which is a much more dynamic process, requiring for each patient a continuous evaluation of immunosuppression needs balanced with the risk of infection.

Renal dysfunction is another frequent complication following liver transplantation. Pre-operatively renal impairment, especially hepatorenal syndrome, can be present. Post-operatively, sepsis, antibacterial therapy and graft dysfunction can influence the development of *de novo* renal failure, but CNI-induced nephrotoxicity is the most important and common cause of long-term dysfunction.

The grade of nephrotoxicity is correlated to the dose of CNI, particularly early dose administration, and to the cumulative quantity of drug over the follow up period. In a retrospective analysis of 883 liver transplant recipients,[9] severe chronic renal failure developed in 4% of patients surviving more than 1 year: serum creatinine values 3 months post OLT and cyclosporin concentrations at 1 month after transplant were identified as risk factors for development of renal failure. In another series[10] of 834 patients, the rate of end stage renal disease (ESRD) was 5.4%; considering the actuarial rate, the incidence of ESRD at 13 years was 9.5%. Multivariate analysis showed serum creatinine values at 4 weeks, 3 months and 1 year to be independent risk factors for the development of ESRD. Furthermore, patients with ESRD who underwent renal transplantation had a much worse outcome when compared to patients undergoing primary renal transplant in the same time frame at the same institution, with survival rates at 10 years of 42.6 versus 75.1%. These observations stress the point that prevention of long-term complications has to be kept in mind from the beginning.

2. Corticosteroid Withdrawal

In the process of reconsidering the strategy for immunosuppression, the first step has been a change in the use of corticosteroids: complications of this treatment[11] in liver transplant recipients can be even more dramatic, as the liver disease already predisposes to several complications, for example pre-existing osteoporosis, diabetes mellitus and bacterial infections.

Corticosteroids have been withdrawn from most long-term immunosuppressive regimens. In the first years after the development of liver transplantation, corticosteroids were withdrawn only when the risk of further rejection was considered less. Following this, studies documented withdrawal, sometimes as early as 2 weeks after transplant. The bulk of evidence in studies with more than 10 patients (table I)[12-21] demonstrates that corticoste-

Table I. Studies on corticosteroid withdrawal in liver transplant recipients

Study	Study design	Immunosuppression	No. pts	Time of withdrawal	Follow up (m)	Acute rejection, no. pts (%)	Chronic rejection, no. pts (%)	Steroid restarted, no. pts (%)	Deaths, no. pts (%)	Lost grafts, no. pts (%)
Padbury[14]	R	CyA+AZA	168	>3m	28	7 (4.5)	6 (3.9)	14 (8.3)	20 (12)	17 (10)
Punch[15]	P	CyA+AZA	51	>1y	13.8	2	0	6	0	0
McDiarmid[13]	RCT	CyA+AZA	33	>1y	19.7	2 (6)	0	NR	0	0
		CyA+AZA+S	31		17.6	2 (6.5)	0	NR	0	0
Tchervenkov[17]	P	CyA	39	>1y	12[a]	3 (9)[a]	0	1	0	0
Fraser[18]	R	CyA+AZA	96	>3m	24.3 ± 1	8 (8.3)	3 (3)	0	14 (14)	4 (4)
		CyA+AZA+S	18		8.4	7 (39)	3 (17)	0	8 (44)	2 (22)
Stegall[16]	P	CyA	28	>2y	12	2 (7.1)	0	NR		
		CyA+S	24			1 (4.2)	0	NR		
Gomez[12]	P	CyA	72	>1y	23.2 ± 8.1	0	0	0	0	0
		CyA+AZA	14			0	0	0	0	0
Belli[19]	RCT	CyA	54	3m	38 ± 15	2 (4)[b]	0	1	11 (20)	
		CyA+S	50		42 ± 15	3 (8)[b]	1 (3)[b]		9 (18)	
Stegall[20]	RCT	CyA+MMF	36	<1 (14d)	6	15 (46)[c]	0	0	2 (6)	1
		TAC+MMF	35			11 (42.3)[c]	0	3 (8.5)	4 (11.4)	1
Tisone[21]	RCT	CyA+AZA+S	22	<3m	16		0	0	6	0
		CyA+AZA	23				0	0	5	0

a Evaluated in 33 patients with >3m follow up.

b Patients who changed therapy (n = 12) excluded from analysis – none had rejection before or after the change.

c 13 patients who did not follow the protocol were excluded from analysis.

AZA = azathioprine; **CyA** = cyclosporin; **MMF** = mycophenolate mofetil; **NR** = not reported; **P** = prospective evaluation; **R** = retrospective evaluation; **RCT** = randomised controlled trial; **S** = corticosteroids; **TAC** = tacrolimus.

roid withdrawal does not increase patient or graft loss, but on the other hand reduces the rates of long-term complications such as hypertension, development of diabetes mellitus or hypercholesterolaemia.

Following the introduction of new immunosuppressive drugs, several centres have developed new 'steroid-free' protocols avoiding their use even in the induction phase and restricting their indication to treatment of acute rejection; however, they have substituted corticosteroids for a new immunosuppressive agent, for example, mycophenolate mofetil,[20,22,23] antithymocyte globulin or anti-interleukin–2 agents,[24] so that overall immunosuppressive potency is not necessarily reduced.

An Italian centre[21] conducted a pilot study comparing two different schedules of immunosuppression based on cyclosporin plus azathioprine with or without corticosteroids, not given *ab initio* in one group, and gradually tapered and then stopped by the end of the third month post OLT in the second group. Protocol biopsies at 1-week post-OLT showed a similar incidence of acute cellular rejection between the two groups (moderate to severe rejection: 60% in corticosteroid-treated patients, 55% in patients without corticosteroids). In this study most patients underwent spontaneous resolution and only four (10%, two in each group) were treated. At 1 month there were only two further rejection episodes (one in each group). This patient group adds further to the evidence that early cellular rejection does not require treatment in every patient in liver transplantation.

However, corticosteroid weaning or withdrawal can be difficult in patients with underlying autoimmune liver disease, who might be at an increased risk of developing recurrent disease.[25,26] In addition, some patients do have cellular rejection after complete withdrawal of corticosteroids. However, this appears to be rare; only a few of the studies in table I document this ranging from 0 to 8% and it is not clear if these patients had autoimmune disease.

This point further underscores the concept that liver transplant recipients are not a homogeneous population. Each treatment regimen should be tailored to the individual patient, taking into account risk factors for rejection, indications and contraindications to immunosuppressive drugs, the condition of the donor graft, the presence of renal dysfunction and risk of infection.

Patients transplanted for viral-induced cirrhosis are the subgroup who could gain most from corticosteroid withdrawal and reduction of immunosuppression. A glucocorticoid-responsive element has been long ago identified in the hepatitis B virus (HBV) genome and its stimulation increases HBV transcription and expression of viral gene products.[27,28] Furthermore, immunosuppression results in an enhanced HBV replication in patients with chronic HBV infection undergoing therapy for cancer.[29] Hepatitis C virus (HCV) reinfection after OLT seems to have a much more rapid and aggressive course, with 20% of patients developing cirrhosis within 5 years after transplantation. The association between HCV recurrence and immunosuppression, as cause and effect seems logical, but has not yet been clearly outlined.[30] In our own cohort of 59 patients we found that the development of more severe fibrosis was significantly associated with triple or double than single agent initial immunosuppression.[31]

3. Single Drug Therapy

Single drug therapy can be considered as an evolution from the previous schedules, that is, to use only one drug from the beginning.

There is only one prospective randomised trial[5] from our unit reporting the use of monotherapy with either cyclosporin or tacrolimus *ab initio*, which included 64 patients. There was no statistical difference between the two groups in patient 1-year survival rates (cyclosporin = 78%, tacrolimus = 85%). Both drugs were administered at the standard dosage used in combination regimens: a total of 0.1 mg/kg/day divided in two daily doses for tacrolimus and 10 mg/kg/day for cyclosporin also divided in two daily doses.

An extended follow-up[32] (with a median duration of 878 days) including 68 patients showed an actuarial 4-year survival for patients and grafts, re-

spectively, of 74 and 73% for the cyclosporin group and 79 and 77% for the tacrolimus group. Over this prolonged period, maintenance monotherapy was unchanged in 87% of tacrolimus-treated and in 70% of cyclosporin-treated patients. There were 22 episodes of acute rejection treated with corticosteroid boluses in 29 patients in the cyclosporin group and 23 episodes in 27 patients in the tacrolimus group; two patients (2.94%) experienced chronic rejection (both in the cyclosporin group). Eight patients (21.6%) in the cyclosporin group and four (12.6%) in the tacrolimus group had no rejection on protocol biopsies. Eight patients in the cyclosporin group were switched to the tacrolimus group because of resistant rejection[7] or nephrotoxicity; two patients were switched from the tacrolimus to the cyclosporin group because of neurotoxicity and haemolysis. Additional immunosuppression (mycophenolate mofetil or azathioprine plus prednisone) was required in two patients in each group.

Further evidence supporting the use of monotherapy safely comes from trials of corticosteroid withdrawal in which the comparison group was on a single drug, usually a standard dose of CNI. In the largest study, Belli et al.[19] described the long-term follow up of a cohort of 104 patients, 54 of whom were randomised 6 months after the transplant to cyclosporin monotherapy, while the remaining 50 were maintained on cyclosporin plus corticosteroids. Only two patients in the cyclosporin group experienced acute rejection due to drug malabsorption, which resolved with corticosteroid boluses but without the need for long-term treatment; one patient was shifted to long-term corticosteroids because of severe pruritus. At 5 years, patients survival rates did not significantly differ between the two groups (cyclosporin plus corticosteroids = 82%, cyclosporin = 77%), whereas the prevalence of hypertension (58 *vs* 17%; p = 0.00002) and diabetes (25 *vs* 6%; p = 0.007) at 12 months was significantly reduced in the monotherapy group.

Gomez et al.[12] described corticosteroid withdrawal in 86 OLT recipients with stable graft function and no rejection 1 year after the transplant. Seventy two patients were maintained on cyclosporin monotherapy and fourteen required azathioprine to allow CNI dose reduction because of nephrotoxicity. After a mean follow up of 23 months, there had been no episode of rejection nor need to resume corticosteroid treatment.

In a Canadian cohort[17] of patients with a mean follow up of 12 months, 39 of 42 patients could be maintained on cyclosporin monotherapy, stopping azathioprine and corticosteroids. Only three of 33 patients with a follow up >3 months and subtherapeutic cyclosporin concentrations experienced an episode of acute rejection, which was successfully treated.

Another small report[16] includes 28 patients receiving cyclosporin monotherapy after corticosteroid withdrawal with a mean follow up of 12 months. Four (14.2%) were treated with corticosteroid boluses for rejection (biopsy-proven in two) and three (10.7%) needed to resume long-term corticosteroid treatment because of generalised symptoms and colitis (one of the three).

A series from Denver, which was reported within another study, included patients (OLT between 1998 and the end of 1999), for whom the main immunosuppressive agent was either tacrolimus or cyclosporin. Corticosteroids were used only in the first 14 days after OLT, leaving patients on CNI monotherapy from that point onward. One-year patient and graft survival were[31] 94 and 89%, respectively.[33]

Long-term follow up of patients treated with tacrolimus-based protocols in Pittsburgh showed that nearly 70% of liver recipients were stable on monotherapy, while 26% needed corticosteroids or azathioprine (8/82 patients, 9.7%) at 84 months after OLT.[34] Similarly, in a clinical trial[35] in 84 patients of tacrolimus low-dose (0.1 mg/kg/day), which is now the dose commonly used, 74% remained on tacrolimus monotherapy without the need for corticosteroids at 1 year.

4. Future Options

Although the present optimal immunosuppressive regimen is not yet completely defined, the liver transplantation community is already exploring new agents.

4.1 Sirolimus

Sirolimus (rapamycin) is one such new agent which inhibits T-cell proliferation between the G1 and S phases, interacting with a class of kinases known as mammalian target of rapamycin (mTOR) and blocking signal transduction. What makes sirolimus a potentially promising drug is the absence of nephrotoxicity and the antifibrotic and anti-smooth muscle cell proliferation effect shown by it in animal models. Similarly to previous immunosuppressive drugs, most of the clinical experience with sirolimus has been obtained in renal and cardiac transplantation. In liver transplantation it has been used as part of triple-drug regimens (with mycophenolate mofetil and corticosteroids[36]) for patients in whom CNI were contraindicated because of renal dysfunction, or following CNI toxicity, and secondly in association with CNI as a steroid- and CNI-sparing agent,[33,37] and thirdly as rescue therapy for rejection.[38]

In the first study,[36] with a maximum follow up of only 7 months, nine of 14 patients received triple therapy without CNI (sirolimus, mycophenolate mofetil, corticosteroids). Of these nine, three had to stop sirolimus and start CNI and in the six who continued sirolimus, four had no rejection. In the other five of 14 who had early toxicity from CNI and sirolimus was substituted, one had to go back on CNI and one had cellular rejection. At the time of reporting, 11 patients were receiving maintenance immunosuppression either with three or four drugs. One had died and two were not taking sirolimus but tacrolimus alone.

In a consecutive series of 52 of 56[37] patients surviving transplantation who were given low-dose tacrolimus (trough target 5 μg/L), sirolimus and corticosteroids, only eight (15%) had rejection but no protocol biopsies were performed. Patients were weaned off corticosteroids between 3 and 6 months. Creatinine levels did not increase over 1 year. Five patients stopped sirolimus (one intolerance, four discontinuation).

In another study[33] with a short mean follow up of 124 days, patient survival was 92%. Cellular rejection (presumptive or biopsy-proven) occurred in ten of 33 (30%) patients receiving sirolimus with either tacrolimus or cyclosporin (both groups had corticosteroids for the first 3 days). The authors compared this with an historical control group in which surprisingly 37% of patients with acute rejection required muromonab-CD3 (OKT3) compared to only 3% of sirolimus-treated patients.

It is difficult to establish the value of sirolimus from these protocols but it further emphasises that corticosteroids can be withdrawn very soon after liver transplantation, if indeed used at all as we believe.

In the last study,[38] 16 patients with chronic rejection were treated with sirolimus and corticosteroids (n = 8) or sirolimus, tacrolimus and corticosteroids (8). Unfortunately it is not reported in which group the eight patients rescued with reversal of ductopenia belonged to.

Currently, there is only one experience of 15 patients reported,[39] in which sirolimus was used as monotherapy *ab initio* in only four patients (two died, one had cyclosporin added and one had a follow-up of only 118 days; three had cellular rejection); and from 3 months in 11 patients who continued sirolimus having stopped cyclosporin and corticosteroids (three died and one changed to tacrolimus, and two had rejection), with a range of follow up between 178 and 806 days.

4.2 Daclizumab and Basiliximab

Another new class of immunosuppressants is the interleukin (IL)-2 receptor (IL-2R) antagonists, daclizumab and basiliximab. These act more specifically than CNI on a receptorial subunit expressed only on activated T cells and thus selectively inhibiting their proliferation.

After proving effective in reducing rejection rates in renal transplantation, daclizumab has been

evaluated in a few small non-randomised studies of induction therapy for liver transplant recipients. In particular, it has been used in those patients with pre-operative renal failure, or at risk of developing postoperative renal dysfunction, but always in combination with other immunosuppressants (mycophenolate mofetil, tacrolimus, corticosteroids),[40-44] with contrasting results. These pilot studies all suggested that induction therapy with daclizumab in combination with non-CNI drugs followed by CNI administration could be carried out safely. However, a prospective pilot study[42] of CNI-free immunosuppression using daclizumab with mycophenolate mofetil and corticosteroids was stopped because of a very high rate of acute cellular rejection (seven of seven enrolled patients, with four patients experiencing steroid-resistant rejection requiring muromonab-CD3 therapy).

Basiliximab, as part of a quadruple induction protocol, has been reported to decrease the rate of acute rejection,[45,46] both steroid-sensitive and steroid-resistant, but there was also an increased rate of chronic rejection. Thus, there is a safety issue with the use of IL-2 receptor antagonists.

On the basis of the sequence of events in graft rejection in OLT, IL-2R antagonists could play a role in immunosuppression at the very beginning, leaving the stage to CNI when they can be introduced without excessive risk of infection or renal dysfunction, that is, monotherapy for the first to second week with IL-2R antagonism and then monotherapy with CNI. This regimen needs to be investigated in randomised clinical trials.

5. Conclusions

Currently the optimal immunosuppressive regimen following liver transplantation is not defined, but we believe that centres should follow an inverse maxim of less is more, that is, less immunosuppressive potency for more long-term gain of good quality survival. Ultimately the induction of tolerance may be the future possibility and reality.[47,48]

It is known from animal models that the early interaction between donor leucocytes and recipient immune cells are fundamental in the development of spontaneous graft acceptance; immune activation is required for this process. CNI and corticosteroids acting at various levels prevent the development of tolerance in several models.[49-51] Future approaches will probably be based on delayed introduction of CNI or their use at a dosage currently considered to be subtherapeutic, using in the very early post-transplant phase drugs that allow an early immunologic activation to induce the tolerising process. More selective treatment of cellular rejection will also be a useful strategy. However, it will take some time before such protocols are evaluated in properly designed randomised trials in liver transplant recipients.

Acknowledgements

No sources of funding were used to assist in the preparation of this manuscript; and the authors have no potential conflicts of interest.

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