CALCINEURIN INHIBITOR-INDUCED RENAL ALLOGRAFT NEPHROTOXICITY

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Background. The introduction of the calcineurin inhibitors (CI) cyclosporine and tacrolimus into immunosuppressive protocols initiated a new era in organ transplantation with excellent short-term graft survival. Nevertheless, the chronic nephrotoxicity of these drugs represents a significant adverse factor limiting their long-term use. Patients treated with a CI can be at risk for developing renal failure and this problem is especially pronounced in patients after renal transplantation.

Methods and Results. In a review paper we summarize the clinical aspects, histological manifestations and pitfalls of diagnostics of acute and chronic CI nephrotoxicity in patients after kidney transplantation. We look in detail at the disputed relationship between blood concentrations of cyclosporine and tacrolimus and histological manifestation of toxicity and summarize data showing that for toxic effects, local renal exposure to CI and their metabolites can play a more significant role than systemic exposure. We also include recent views on the pathophysiologic and molecular mechanisms underlying these changes; factors influencing local susceptibility to CI nephrotoxicity are discussed, including variability of expression and activity of P-glycoprotein and cytochrome P450. Last but not least we summarize our own experience with clinically manifest and subclinical forms of nephrotoxicity and their impact on the progression of chronic graft changes.

Conclusions. Owing to their unique effects, CI remain the cornerstone of most immunosuppressive protocols for renal transplantation. Together with optimization of local kidney exposure to CI and their metabolites, efforts to reduce systemic levels as much as possible are the most important preventive measure for reducing toxic renal graft damage.

INTRODUCTION

The introduction of the calcineurin inhibitors (CI), cyclosporine and tacrolimus into immunosuppressive protocols for renal transplantation at the end of the eighties and middle of the nineties led to a significant breakthrough in transplant medicine and the achievement of excellent short-term survival of renal grafts¹. Tacrolimus was then successfully used in liver, pancreas, and heart transplantation². Today, both CIs are pivotal in the prevention of

allograft rejection³ and are part of more than 90% of immunosuppressive protocols used in organ transplantation⁴.

Cyclosporine and tacrolimus differ in their molecular structure and intracellular binding characteristics. Cyclosporine binds to a group of cyclophilin molecules which have high affinity for calcineurin, a key protein phosphatase in the process of T-cell activation. Tacrolimus forms a complex with its cytosol partner FK506-binding protein 12 which also binds to calcineurin. By blocking calcineurin, both CIs inhibit phosphatase controlled translocation of nuclear factor of activated T cells (NFAT) into the nucleus and prevent induction of cytokines and their receptors, required for activation and proliferation of lymphocytes and other immune cells⁵.

Inhibition of the calcineurin-NFAT pathway by cyclosporine and tacrolimus is not specific to immune cells and it can lead to toxic changes in addition to immunosuppressive effects⁶. After kidney transplantation these can manifest as acute or chronic nephrotoxicity. For the acute form, a hemodynamically induced, usually fully reversible disorder of renal function is characteristic. The chronic nephrotoxic effects of CI are already associated with irreversible changes in the form of interstitial fibrosis and tubular atrophy⁷ and are considered to be a significant cause of late dysfunction of transplanted kidneys. An important measure in the prevention of these chronic

ABBREVIATIONS

CI	– Calcineurin inhibitors
NFAT	– Nuclear factor of activated T cells
RAS	– Renin-angiotensin system
NO	– Nitric oxide
COX-2	– Cyclooxygenase-2
TGF-β	– Transforming growth factor beta
C ₀	– Trough level of CI
C ₂	– 2-hour CI post-dose level
AUC _{0 to 12}	– Area under the time-concentration curve
ABCB1	– ATP-binding cassette sub-family B member 1
MDR1	– Multidrug resistance protein-1
NSAIDs	– Non-steroidal anti-inflammatory drugs
ACE	– Angiotensin-converting enzyme

changes is the effort to reduce the CI dose as much as possible and carefully monitor plasma levels. A pitfall in CI use is their variable pharmacokinetics, narrow therapeutic range, and individual susceptibility to their toxic effects⁸. CI serum levels therefore often fail to correlate with the extent of kidney damage and manifestation of toxicity can be non-specific or, especially in the first months after the transplantation, the toxic changes can be clinically silent. Significant side effects associated with cyclosporine and tacrolimus therapy not only impair long-term renal graft function but also the survival of patients. They are also a cause of significant co-morbidity. CI nephrotoxicity can be a significant clinical problem even in non-renal organ transplantation and multiple other conditions requiring treatment with these drugs.

In this review we summarize the clinical and histological aspects of CI nephrotoxicity, together with the molecular mechanisms and risk factors associated with acute and chronic nephrotoxicity. We also review current preventive and therapeutic guidelines and include our own experience with subclinical and clinically manifest forms of CI nephrotoxicity in a group of 424 protocol biopsies of transplanted kidneys.

ACUTE CI NEPHROTOXICITY

Acute CI nephrotoxicity is characterized by hemodynamically induced renal dysfunction which is nearly always fully reversible. Most data on acute toxicity relates to cyclosporine but tacrolimus effects are believed to be similar. The basis of acute nephrotoxicity is primarily the vascular effects of both CIs but we can also see some symptoms of tubular damage and more rarely symptomatology similar to thrombotic microangiopathy - hemolytic-uremic syndrome.

Acute arteriopathy

Acute hemodynamic effects associated with cyclosporine are mediated by vasoconstriction of afferent arterioles. It has been demonstrated that vasoconstriction is induced by activation of the renin-angiotensin system (RAS) and increase in levels of the vasoconstricting factors, endothelin and thromboxane, as well as suppression of synthesis of vasodilating prostacyclin, prostaglandin E₂, and nitric oxide (NO)⁹⁻¹¹. Interindividual susceptibility to cyclosporine-induced vasoconstriction also plays an important role¹². RAS is activated by direct effect of cyclosporine on juxtaglomerular cells¹³ and indirectly by induced renal arteriolar vasoconstriction, decrease in vasodilating factors levels and increase in endothelin levels¹⁴. Renin-producing cells in afferent arterioles have also been observed in relationship to cyclosporine administration¹⁵. Activation of RAS further intensifies the hemodynamic changes by increasing the production of angiotensin II. Cyclosporine also increases free radical¹⁶ and superoxide¹⁷ production and this reduces the bioavailability of NO¹⁸. Another mechanism by which CI contributes to renal vasoconstriction is a shift of the balance between vasomotor effects of arachidonic acid metabolites (eicosanoids) to-

wards a more pro-vasoconstriction state. The promoter for the COX-2 gene coding for cyclooxygenase-2 contains binding sites for NFAT and NFAT is considered to be the main COX-2 *in vitro* expression stimulator¹⁹. Inhibition of calcineurin/NFAT activity thus reduces COX-2 expression²⁰ which leads to reduction of prostaglandin E₂ production and supports renal vasoconstriction. Selective COX-2 inhibitors have similar effects on renal hemodynamics to CI.

Toxic tubulopathy

Acute CI nephrotoxicity is frequently accompanied by isometric vacuolization of tubular cytoplasm induced by enlargement of endoplasmic reticulum and multiplication of lysosomes^{21,22}. Vacuolization can be a result of relative ischemia caused by vasoconstriction, however the possibility of damage to structure and function of endoplasmic reticulum by inhibition of calcineurin in tubular epithelial cells cannot be ruled out. In this context, it has been demonstrated that cyclosporine induces endoplasmic reticulum stress and cell damage to tubular (and endothelial) cells which can contribute to dedifferentiation and cell death²³⁻²⁵ by induction of pro-apoptotic proteins²⁶. Similar effect on tubular cells was observed with tacrolimus²⁷. Isometric vacuolization is however not a specific finding for CI toxicity, it is also observed in patients on maintenance immunosuppression based on steroids and azathioprine, as well as in purely ischemic damage to the kidney. We also see it in the absence of renal dysfunction and it is probably not related to progression of chronic CI nephrotoxicity²⁸.

Thrombotic microangiopathy - hemolytic-uremic syndrome

Administration of cyclosporine, and less often tacrolimus, represents an important risk factor for development of post-transplant thrombotic microangiopathy - hemolytic-uremic syndrome, a severe but fortunately infrequent form of vasculopathy. The main causative mechanism is endothelial damage due to ischemia caused by vasoconstriction; CI induced hyper-aggregation of platelets can also contribute as activation of prothrombotic factors^{29,30}. Withdrawal of cyclosporine with conversion to tacrolimus or administration of sirolimus is usually a sufficient therapeutic measure^{31,32}. In more severe cases, plasmapheresis, administration of intravenous immunoglobulins may be necessary and recently the positive effect of belatacept as a suitable maintenance immunosuppression in these patients has been reported³³.

CHRONIC CI NEPHROTOXICITY

CI can cause both reversible change in renal hemodynamics and over the long-term lead to development of irreversible damage to parenchyma structures. Typical chronic morphological changes are arteriolar hyalinosis, tubular atrophy, interstitial fibrosis, thickening and fibrosis of the Bowman's capsule, and focal, segmental or global glomerular sclerosis^{34,35}. The etiology of these changes most likely involves a combination of CI induced

hemodynamic changes and direct toxic effect on tubular epithelial cells.

Chronic arteriopathy

A characteristic sign of chronic CI toxicity is the presence of focal or circular lumpy hyaline deposits in the media of afferent arterioles (arteriolar hyalinosis) which replaces necrotic smooth muscle cells. The cause of this cellular damage is unclear, but again can be related to the important role of calcineurin-NFAT in smooth muscle cells³⁶. If the formed deposits are large, they can cause hemodynamically significant narrowing of vascular lumen with subsequent ischemization of renal tissue³⁷. Arteriolar hyalinosis is usually considered to be irreversible, but complete regression of severe cyclosporine-induced arteriopathy has been reported after withdrawal of or reduction in cyclosporine dose³⁸.

Chronic tubulo-interstitial changes

Arteriopathy and narrowing of arteriolar lumen can also be considered as a major contributor to the development of interstitial fibrosis (typically striped) and tubular atrophy in CI therapy. Free radicals and reactive oxygen species are produced because of local ischemia inducing cell apoptosis^{16,39} with damage to tubulo-interstitial cell structures. A significant etiologic factor involved in progression of chronic interstitial changes is also the CI induced increase in production of TGF- β (transforming growth factor beta). This seems to be stimulated in part by decrease in NO release⁴⁰ and in part by increase of local angiotensin II concentration⁴¹. TGF- β supports development of interstitial fibrosis by inhibiting degradation and supporting production of proteins of the extracellular matrix and by induction of epithelial mesenchymal transition^{42,43}. It is also involved in progression of chronic graft vasculopathy⁴⁴. The role of TGF- β in the pathogenesis of chronic changes is supported by the fact that blocking its effect via anti-TGF- β antibodies, led to an amelioration of morphologic manifestations of nephrotoxicity and stabilization of renal function in a mice model⁴⁵. Activation of RAS also contributes to the progression of chronic tubulo-interstitial changes. Besides its direct hemodynamic effect, it stimulates secretion of aldosterone which itself worsens interstitial fibrosis by increased production of growth factors (e.g. TGF- β) and reactive oxygen species and by inhibition of extracellular matrix degradation⁴⁶.

Chronic glomerular changes

Chronic glomerular damage associated with CI treatment occurs as a result of glomerular ischemia due to severe arteriolar hyalinosis⁴⁷. Global glomerular sclerosis is most common⁴⁸ but we can also see focal segmental fibrous changes caused by hyperfiltration of residual glomeruli⁴⁹. Eventually, atubular glomeruli can be detected in case of tubular disruption.

ELECTROLYTE DISORDERS

Disorders of ion homeostasis, such as hyperkalemia, hypomagnesemia, hyperuricemia, and hyperchloremic metabolic acidosis often occur as a result of impaired tubular function⁵⁰. Hyperkalemia in calcineurin blockade relates in part to inhibitory effects on Na⁺-K⁺ATPase in collecting ducts^{51,52} and to the decreased number of mineralocorticoid receptors⁵³ and distal tubular acidosis. Cyclosporine also has a significant effect on magnesium metabolism. Decrease in magnesium re-absorption due to reduction of paracellin-1 expression in the thick ascending limb cells of the loop of Henlé often leads to severe hypomagnesemia^{54,55} which in itself facilitates development of chronic interstitial fibrosis⁵⁶. Cyclosporine is also one of the leading causes of post-transplant hyperuricemia due to its inhibitory effect on tubular secretion of uric acid⁵⁷.

CI NEPHROTOXICITY DIAGNOSTIC OPTIONS

The diagnosis of clinically manifest form of acute CI nephrotoxicity relies on correct evaluation of the clinical picture. It should be considered in the case of acute impairment of graft function by at least 20% above baseline values with concurrent elevation of cyclosporine levels > 400 ng/ml or tacrolimus >20 ng/ml. Reduction of the dose and normalization of CI levels in such a case should be followed by improvement of graft function. In the case of subclinical acute toxicity in grafts with very good function, the only option for detection is a protocol biopsy of the transplanted kidney. Biopsy represents an essential means for determining chronic nephrotoxicity and at the same time provides important information about the degree of irreversible changes of individual parenchyma structures.

Unfortunately, no other non-invasive procedure or sufficiently sensitive and specific marker is currently available for early diagnosis and monitoring of toxicity. In the early post-transplantation period, serum creatinine level does not reflect the degree of damage to the graft well⁷. Likewise, the increased count of CD 146+ circulating endothelial cells in peripheral blood is not specific as a marker of vascular toxic damage to the graft^{58,59}. In our pilot study which prospectively monitored histological and ultrasound findings of very well functioning grafts affected by subclinical rejection⁶⁰, no sufficiently sensitive and specific parameter for toxicity was identified (unpublished data).

COMPARISON OF TOXIC EFFECTS OF BOTH CI

Owing to their identical mechanism of action associated with inhibition of calcineurin, both CIs have similar nephrotoxic characteristics and induce similar histological damage⁶¹. There is some evidence that tacrolimus has a lower nephrotoxic potential than cyclosporine, especially for lower vasoconstricting properties⁶² and profibrogenic characteristics⁶³; latter data however confirm similar

effects of both CIs on the development of histological toxic changes⁴⁸, progression of chronic interstitial alterations^{64,65}, and level of oxidative stress^{66,67}.

In renal transplantation, the precise analysis of long-term results of graft survival based on comparison of the toxic effects of cyclosporine and tacrolimus is much more difficult to perform, because of the interference with graft rejection. The preference of tacrolimus to cyclosporine in current immunosuppressive protocols is based, despite similar nephrotoxic profile, on its more favorable metabolic profile, better anti-rejection effects, and better long-term function and survival of the graft^{68,69}. A similar conclusion was reached by the SYMPHONY study, currently the largest randomized multicenter study comparing different immunosuppressive protocols, which showed the benefit of low dose tacrolimus over cyclosporine in terms of both long-term graft function and graft survival⁷⁰.

There are also differences in the non-renal toxic effects of cyclosporine versus tacrolimus⁷¹ which often influence the choice of CI in a particular patient. Post-transplant diabetes mellitus, neurotoxicity, cephalgia, diarrhea, and hypomagnesemia were observed more commonly after tacrolimus treatment. Cyclosporine is associated especially with gingival hyperplasia, hirsutism, higher LDL cholesterol and triglycerides levels^{68,72}. The causes of these differences between cyclosporine and tacrolimus are not clear.

FACTORS INFLUENCING DEVELOPMENT OF CI NEPHROTOXICITY

The main goal of immunosuppressive therapy after kidney transplantation is to prevent rejection complications and at the same time toxic effects of the medication used. This is mainly achieved by monitoring blood concentrations. However, to maintain cyclosporine and tacrolimus concentrations in the recommended range is complicated by their much varied individual absorption, distribution, metabolism and elimination. For these reasons there is a poor correlation between the CI dose and subsequent blood concentration⁷³.

Therapeutic drug monitoring of CI

Cyclosporine and tacrolimus blood levels are routinely established as fasting levels 12 hours following prior application (C_0). For tacrolimus, this analysis is considered sufficient for checking the adequacy of dosing⁷⁴. However, for cyclosporine, the correlation between C_0 levels and the total exposure measured as area under the time-concentration curve (AUC_{0-12}) is relatively poor. Measurement of cyclosporine levels two hours after administration (C_2) correlates more closely with exposure^{75,76} but the use of C_2 in routine outpatient practice is complicated. Furthermore, there are no conclusive studies showing a significant benefit of C_2 measurements compared to C_0 in prevention of rejection or toxicity and no recommended target values for C_2 have been set.

It is therefore difficult to infer CI nephrotoxicity from C_0 or C_2 levels. In our group of 424 protocol biopsies in which clinically manifest and subclinical nephrotoxic

changes were evaluated over a one-year follow-up, C_0 levels of cyclosporine and tacrolimus did not differ significantly between groups with toxicity and normal histological finding. Moreover, cyclosporine C_2 levels ranging within the selected therapeutic limits did not prevent the development of toxic changes, either⁷⁷. Rowshani et al. also demonstrated that even with low target AUC in month 6 and year 1 after the transplantation there is significant fibrogenic potential⁶⁵, which suggests that neither such detailed monitoring approach can eliminate interindividual differences in sensitivity to CI, probably genetically induced.

Pharmacokinetics and pharmacogenetics of CI

Genetic variability affecting expression and function of metabolizing isoenzymes of cytochrome P450 (especially CYP3A4 and CYP3A5) and transport P-glycoprotein which is associated with single-nucleotide polymorphisms of coding genes is responsible for interindividual differences in intestinal absorption, bioavailability, and anti-rejection and toxic effects of cyclosporine and tacrolimus^{78,79}. Following intestinal absorption, more than 90% of cyclosporine and tacrolimus is bound to plasma proteins; in full blood both are highly concentrated in erythrocytes. Metabolic processing of CI by hydroxylation and demethylation by CYP3A4, CYP3A5 (as well as CYP3A7) takes place mostly in the liver and gastrointestinal tract. Only small amounts of the maternal compound appear in urine and stool, most metabolites are then eliminated in bile and to some extent also in urine.

The effect of single-nucleotide polymorphism of genes for CYP3A and P-glycoprotein on CI pharmacokinetics still remains unclear. According to some authors, the CYP3A4*1B allele which increases expression of CYP3A4 is associated with significantly higher clearance of cyclosporine compared to homozygotes with normal (wild-type) allele CYP3A4*1^{80,81}. Other studies however failed to confirm this association^{82,83} obviously by reason of low population frequency of CYP3A4*1B allele and difficulties in acquiring adequate numbers of subjects for valid statistical analysis. In carriers of at least one wild-type CYP3A5*1 allele, cyclosporine administration was associated with significantly lower AUCs compared to CYP3A5*3 homozygotes with low CYP3A5 enzyme activity, where adequate levels of cyclosporine could be achieved with lower doses⁸⁴⁻⁸⁷. However these findings have not been confirmed by other authors again⁸⁸⁻⁹⁰. In a paper by Crettol et al. the pharmacokinetics of cyclosporine were evaluated in expressors of CYP3A7. Carriers of CYP3A7*1C genotype (which was shown to be a marker of CYP3A7 expression) required 1.4 to 1.6 times higher daily doses per body weight than noncarriers during the first year after transplantation⁸⁶. Concerning the influence of polymorphism in ABCB1 [ATP-binding cassette subfamily B member 1, formerly known as MDR1 (multidrug resistance protein-1)] gene for P-glycoprotein on pharmacokinetics of cyclosporine, significantly lower concentrations and AUCs were demonstrated in carriers of MDR1 1236 wild-type allele compared to homozygotes with 1246 T allele⁹¹. Another study documented significantly lower AUC and C_2 levels in carriers of MDR1 3435 T allele

in examination done on day 3 after the transplantation, however this difference was not significant one month later⁹². Some other large scale studies did not confirm any association between polymorphisms of MDR1 and pharmacokinetics of cyclosporine^{93,85}.

The significance of CYP3A and ABCB1 (MDR1) polymorphisms was also studied for tacrolimus. Despite of limitations associated with low population CYP3A4*1B allele frequency, lower C_0 concentrations of tacrolimus were demonstrated in its carriers in 3rd and 12th month after transplantation, compared to CYP3A4*1 homozygotes⁹⁴. Patients with CYP3A5*3 polymorphism and missing enzyme activity exhibited higher blood levels and required lower maintenance doses of tacrolimus compared to the CYP3A5*1 variant^{95,96,90,97}. For MDR1 and tacrolimus pharmacokinetics, carriers of 2677T or 3435T alleles were found to have higher average levels of tacrolimus compared to 2677 G homozygotes (GG) and 3435 C homozygotes (CC)^{98,99}. This association was not confirmed by other authors^{100,101} and the significance of other variants of the ABCB1 (MDR1) gene for pharmacokinetics of tacrolimus is unclear⁹⁷.

Tissue concentrations of CI

As was already mentioned, not even precise monitoring of CI levels prevents development of chronic nephrotoxicity. One of the possible reasons is the fact that local CI concentrations in the renal graft do not correspond with the systemic levels, but rather are much higher^{102,103}. In their study, Podder et al. documented that higher local concentrations of cyclosporine significantly correlate with reduction of renal function and development of histological signs of nephrotoxicity¹⁰⁴. Studies of tissue expression and function of P-glycoprotein further elucidated the relationship between local exposure and nephrotoxicity. This transporter is expressed on the apical membrane of tubular epithelial cells and is the main protein responsible for excretion of tacrolimus and cyclosporine¹⁰⁵. It has been demonstrated that cyclosporine-induced up-regulation of P-glycoprotein is indirectly related to incidence of arteriolar hyalinosis, interstitial fibrosis, and periglomerular fibrosis¹⁰⁶. On the other hand, if the expression or function of this protein is reduced, cyclosporine may accumulate. It has been documented that single-nucleotide polymorphism of ABCB1 (MDR1) in the kidney donor associated with presence of the T allele at position 3435 affecting the expression and function of P-glycoprotein is associated with development of chronic cyclosporine nephrotoxicity¹⁰⁷. Whether P-glycoprotein expression is also affected by tacrolimus is still to be shown. It will also be necessary to confirm if there is a relationship between ABCB1 (MDR1) polymorphism and local renal concentrations of CI.

Toxic effects of CI metabolites

Studies aimed at evaluation of biologic effects of CI metabolites focus mostly on their immunosuppressive characteristics; very little is known about their toxic effects. Roby et al. have shown that blood levels of cyclosporine metabolites significantly exceed levels of the

maternal drug and several of these metabolites reduced glomerular filtration rate in a rat model¹⁰⁸ and led to tubular vacuolization¹⁰⁹. In a study done in recipients of transplanted liver, a positive association between cyclosporine metabolite levels in blood and renal dysfunction was observed¹¹⁰. However, the effects of these metabolites on tubular function, expression of TGF- β , and other aspects of CI nephrotoxicity are currently unknown. The nephrotoxic potential of tacrolimus metabolites has not been studied yet, however suspicion for their toxicity is supported by the fact that recipients of kidneys who express CYP3A5 (carriers of polymorphism CYP3A5*1) have higher risk of development of chronic CI nephrotoxicity compared to non-expressors (CYP3A5*3/*3)^{111,112}. Our knowledge is insufficient for a final verdict as to whether cyclosporine and tacrolimus metabolites contribute to development of nephrotoxicity. Hypothesis on their toxic effects will have to be verified by controlled prospective studies with measurements of CI levels and levels of their metabolites not only in blood but also in renal tissue.

Local factors increasing risk of CI toxicity

Interindividual variability in sensitivity to CI nephrotoxicity can be influenced by local renal factors independent of systemic or renal levels of cyclosporine, tacrolimus or their metabolites. Higher susceptibility to toxic damage to the older native kidneys or kidneys from older donors is probably related to alteration of autoregulation of renal perfusion and presence of pre-existing chronic vascular changes. Such predisposed kidneys also tend to have reduced capacity for natural cellular reparation¹¹³ which is especially dangerous in CI accelerated aging of cells and formation of reactive oxygen species¹¹⁴.

Chronic abuse of non-steroidal anti-inflammatory drugs (NSAIDs) facilitates the progression of chronic changes by a mechanism similar to the one by which these drugs increase susceptibility to CI nephrotoxicity^{115,116}. Some other factors also represent risk for nephrotoxicity. Polymorphism of TGF- β in codon 10 was associated with progressive dysfunction of native kidneys in recipients of transplanted hearts treated with cyclosporine or tacrolimus¹¹⁷. In case of recipients of transplanted kidneys, this knowledge could help identify subjects who are more susceptible to CI nephrotoxicity because they received a graft from a carrier of this polymorphism and their immunosuppressive therapy could be adjusted accordingly. The presence of polymorphism in gene for ACE (angiotensin-converting enzyme) (DD genotype) is also reported to be associated with shorter long-term graft survival in high-risk recipients of transplanted kidneys due to higher susceptibility to CI nephrotoxicity¹¹⁸.

PREVENTION AND THERAPY OF CI NEPHROTOXICITY

Complete elimination, minimization of doses or early withdrawal of CI

It is likely that some recipients of transplanted kidneys could profit from immunosuppressive protocols from

which CI would be completely eliminated or administered only at low doses. Such practice would be suitable especially in cases of kidney transplantation from older donors and in presence of risk polymorphisms of genes for TGF- β , ACE, and enzymes and proteins involved in transport and metabolism of CI, primarily CYP3A5 and ABCB1 (MDR1). Complete exclusion of CI from immunosuppressive protocols in the immediate post-transplant period will not ensure adequate prophylaxis of acute graft rejection¹¹⁹. However, after a period of several months from the transplantation, the optimal choice for such high-risk patients could be conversion from a CI to less nephrotoxic drugs¹²⁰ or alternatively minimization of CI doses with maintenance of concentrations at the lower end of the therapeutic range. Repeated protocol biopsies of the graft can be used to confirm the safety of such methods from the aspect of subclinical rejection changes or persisting subclinical toxic changes; biopsy is currently the only reliable method for monitoring the adequacy of immunosuppressive therapy. In a paper published earlier we presented our recommendation for management of CI therapy and optimal schedule of protocol biopsies of transplanted kidneys⁷⁷.

Alternative options

In a situation when presence of CI in the immunosuppressive protocol is required to ensure optimal transplant results, certain preventative measures can be used to limit their nephrotoxic effects. Administration of calcium antagonists as vasodilators removes the renal vasoconstriction which, it would appear, plays a key role in development of acute and chronic toxicity. Beneficial effects of calcium antagonists on preservation of plasma flow and glomerular filtration rate in transplanted kidneys were demonstrated for nitredipine, lacidipine^{121,122} and nifedipine¹²³.

In a single study, inhibition of RAS by ACE inhibitors has limited development of cyclosporine nephrotoxicity¹²⁴ and administration of angiotensin II blockers led to a significant reduction in plasma levels of TGF- β and endothelin^{125,126}. In an experiment, application of spironolactone eliminated many of the RAS- and aldosterone-induced effects occurring during CI therapy¹¹; studies confirming these finding in humans are however not available.

Other therapeutic approaches are promising for the prevention of CI nephrotoxicity and progression of chronic changes, e.g. magnesium supplementation in CI induced hypomagnesemia¹²⁷, administration of statins¹²⁸ and antioxidants^{129–132} or application of an anti-TGF- β antibodies⁴⁵. However, no controlled studies with these approaches are available.

CONCLUSION

Introduction of cyclosporine and tacrolimus into immunosuppressive protocols significantly improved short term results of organ transplantations. However, the chronic nephrotoxicity of these agents negatively affects renal graft survival, limiting their long-term administra-

tion. Pathophysiologic mechanisms behind CI nephrotoxicity are only partially elucidated and especially the question of whether inhibition of calcineurin-NFAT pathway is the main effect responsible for toxicity still remains unsolved. For the differences in interindividual sensitivity to CI, local renal factors are probably more important than systemic exposure to cyclosporine or tacrolimus. It is primarily the variability in expression and activity of P-glycoprotein and CYP3A4/5, effect of polymorphisms in genes for TGF- β and ACE, but the autoregulation and reparation potential of the donor kidney also plays a role just as other factors facilitating toxicity, such as salt depletion and high-risk concomitant pharmacotherapy. Since CI remain, despite their nephrotoxic effect, the mainstay of immunosuppressive protocols, their use needs to be optimized. The main measure to prevent nephrotoxicity is the effort to reduce systemic levels and keep local renal exposure to CI and their metabolites as low as possible. Repeated protocol biopsy seems to be a good tool for the confirmation of the safety of such methods from the aspect of subclinical rejection changes or persisting subclinical toxicity. Currently it represents the only reliable method for checking the adequacy of immunosuppressive therapy.

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