Evaluating mechanisms of post-transplant diabetes mellitus

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Abstract

Post-transplant diabetes mellitus (PTDM) is a frequent complication in renal transplantation. While both tacrolimus and ciclosporin are known to be associated with PTDM, the mechanisms underlying this metabolic disturbance and the relative contribution of concomitant corticosteroids have been unclear. At the University Hospital Maastricht, a series of studies have been conducted to investigate these issues. Administering tacrolimus to non-diabetic, dialysis patients was shown to result in a dose-related reduction in insulin secretion without altering insulin resistance. The patients who developed diabetes after transplantation already had impaired glucose metabolism pre-transplant. In a second study, corticosteroid withdrawal from tacrolimus-based immunosuppression reduced insulin resistance without changing insulin secretion. Moreover, reducing tacrolimus blood levels by 30% within the therapeutic window increased both insulin and C-peptide secretion by 24 and 36%, respectively. Accordingly, the effects of tacrolimus on insulin secretion are both dose dependent and reversible. A comparison of the effects of tacrolimus and ciclosporin on glucose metabolism revealed reduced insulin release with tacrolimus at week 3 post-transplant, but for the remainder of the 3 year follow-up there were no significant differences between the two treatment arms. Also, no difference was reported in glucose metabolism following conversion of stable renal recipients from ciclosporin to tacrolimus. Therefore, replacing tacrolimus with ciclosporin in patients experiencing glucose metabolism disturbances is unlikely to be helpful. In a recent study, early corticosteroid withdrawal from tacrolimus-based therapy resulted in a significantly lower incidence of new-onset diabetes mellitus than that achieved with a corticosteroid dose-tapering regimen. In conclusion, corticosteroid minimization plus dose-optimized tacrolimus immunosuppression is likely to be the best option for patients at risk of developing PTDM.

Keywords: ciclosporin; corticosteroid withdrawal; insulin resistance; insulin secretion; post-transplant diabetes mellitus; tacrolimus

Introduction

Diabetes mellitus is a metabolic disorder that is prevalent in the general population, with more than one in five people over 50 years of age presenting with the condition. It is also a recognized complication of organ transplantation. Post-transplant diabetes mellitus (PTDM) is a form of type 2 diabetes mellitus, which is thought to develop in response to a relative insulin deficiency resulting from increased insulin resistance or impaired insulin production, or a combination of both [1]. Transplant recipients are at a particularly high risk of developing PTDM as a consequence of factors additional to those that affect the general population, including the immunosuppressive agents used in transplant management protocols. PTDM is not a separate entity, but a symptom of an underlying metabolic disorder, which is uncovered by immunosuppression.

Here we describe a series of studies undertaken at the University Hospital Maastricht to investigate the effects of immunosuppressants on glucose metabolism. Prevention of PTDM is an important element of successful immunosuppression in renal transplantation, as uncontrolled diabetes is associated with cardiovascular morbidity and mortality [2], which increases the risk of death with a functioning graft [3].

Methods of investigating glucose metabolism

As mentioned, PTDM is thought to occur because of decreased insulin release and/or increased insulin resistance. Given fasting glucose and insulin values
and an intravenous glucose tolerance test (IVGTT), it is possible to determine the mechanisms responsible for PTDM [4]. Insulin resistance can be calculated from fasting values of glucose, insulin and C-peptide; the insulin/glucose ratio; and HOMA-R [homeostasis model assessment of resistance: fasting glucose (mmol/l) \times \text{fasting insulin (mU/l)} \div 22.5].

Insulin secretion can be calculated from the increment of insulin and C-peptide levels after a glucose load, obtained from IVGTT. The insulin sensitivity index ($k_G$) is a parameter determined by insulin resistance and insulin secretion, and expresses the percentage of glucose that is removed from the blood per minute. It is calculated by linear regression from the log-transformed glucose values from 10 to 30 or 60 min after intravenous glucose. A $k_G$ value below 0.8%/min is considered abnormal; values between 0.8 and 1.2%/min indicate borderline glucose intolerance, and values above 1.2%/min are considered normal.

In the prospective studies discussed herein, insulin secretion capacity and insulin resistance were measured, together with other parameters, to assess the mechanisms of PTDM and the relative contributions of various immunosuppressive agents on PTDM.

**Mechanistic studies on the influence of tacrolimus and corticosteroids on glucose metabolism**

The first study in the series assessed the effects of tacrolimus alone in non-diabetic patients on dialysis, in order to exclude the confounding influences of surgery and use of concomitant corticosteroids [5]. The 18 patients selected for the study were subjected to IVGTT before and after 5 days of tacrolimus administration (0.15 mg/kg twice daily). Glucose metabolism was studied by measuring $k_G$, insulin resistance, and C-peptide and insulin release. As shown in Figure 1, three patients had a borderline $k_G$, inferring a pre-diabetic state, prior to the administration of tacrolimus. After 5 days of tacrolimus treatment (median trough level was 17.1 ng/ml), $k_G$ decreased in 16 of the 18 patients, from a median of 1.74%/min before tacrolimus exposure to 1.08%/min ($P<0.0001$). Insulin secretion also decreased from a median of 865 to 600 mU/min/l ($P<0.0001$), but there was no significant change in insulin resistance determined by HOMA-R (median of 2.74–2.82 mU/l; $P=0.33$). There was a significant correlation between $k_G$ and tacrolimus trough levels ($P=0.045$).

Seventeen of the 18 patients in this study underwent renal transplantation, 14 of whom were followed-up for a median of 34 months. Three patients developed PTDM, two who had received azathioprine and one who had received mycophenolate mofetil (MMF) in addition to tacrolimus and corticosteroids. Two of the patients had a borderline $k_G$ and one had a low–normal $k_G$ before tacrolimus administration. The results of this study indicate that the decreased $k_G$ seen after 5 days’ treatment with tacrolimus is related to diminished insulin secretion in response to a glucose load. These data also suggest that patients with an abnormal or low–normal $k_G$ may be at increased risk of developing PTDM while receiving tacrolimus-based immunosuppression.

A second study was undertaken to evaluate the relative role of tacrolimus and corticosteroids in the development of glucose metabolism disorders in renal transplant recipients [6]. All 15 stable, non-diabetic, transplant patients included in the study received tacrolimus (0.1 mg/kg twice a day initially, adjusted to achieve target levels <10 ng/ml after the first month) and prednisolone. Fourteen patients also received MMF (1 g/day). Glucose metabolism was evaluated by IVGTT at three time points: before and after corticosteroid withdrawal and after tacrolimus trough level reduction.

After withdrawal of a 10 mg dose of prednisolone, insulin resistance decreased; there was a significant reduction in median fasting C-peptide levels (22%; $P<0.001$) accompanied by reductions in fasting insulin levels, HOMA-R and the insulin/glucose ratio (Figure 2). A reduction in tacrolimus trough levels (from 9.5 to 6.4 ng/ml) resulted in a significant increase in median C-peptide secretion (36%; $P=0.04$).

![Fig. 1. Effect of tacrolimus on the insulin sensitivity index ($k_G$) in 18 non-diabetic patients on dialysis. The shaded area indicates a borderline value of $k_G$, a higher value indicates a normal $k_G$ and a lower value indicates an abnormal $k_G$. Only three patients went on to develop post-transplant diabetes mellitus (PTDM) after receiving tacrolimus, two who had a borderline $k_G$ prior to tacrolimus therapy and one with a low–normal $k_G$. Adapted from van Duijnhooven et al. [5], with kind permission.](image1)

![Fig. 2. Effect of corticosteroid withdrawal and tacrolimus trough level reduction on median fasting blood glucose parameters associated with insulin resistance in 15 stable, non-diabetic, renal transplant patients. Adapted from Boots et al. [6], with kind permission. HOMA-R = homeostasis model assessment of resistance.](image2)
and a 24% increase in insulin secretion ($P = 0.06$), but insulin resistance parameters did not change (Figure 2). Accordingly, the effects of tacrolimus on insulin secretion are both dose dependent and reversible in the majority of patients. These findings are consistent with a higher prevalence of PTDM in the early post-transplant period when high levels of calcineurin inhibitors and corticosteroids are used.

**Comparison of tacrolimus and ciclosporin immunosuppression**

It has been observed previously that the diabetogenic potential of tacrolimus may be greater than that of ciclosporin [7]. A prospective, randomized, longitudinal study has been undertaken to compare glucose metabolism in adult kidney allograft recipients receiving tacrolimus ($n = 11$) vs ciclosporin ($n = 12$) [8]. Tacrolimus was given orally at 0.15 mg/kg twice daily and titrated to achieve trough levels of 10–15 ng/ml during the first 3 months and 7–10 ng/ml thereafter. The starting dose of ciclosporin was 8 mg/kg/day given in two divided doses and titrated to initial trough levels of 100–200 ng/ml followed by 100–150 ng/ml after month 3. All patients received azathioprine 1–2 mg/kg/day until month 3. Prednisolone was administered at a starting dose of 20 mg/day and this was tapered to 5 mg/day from week 6 onwards. Glucose metabolism, studied via IVGTT, was monitored at week 3, months 3 and 6, and at years 1, 2 and 3 post-transplant.

The only significant difference between the two treatment groups was seen at 3 weeks post-transplant, at which time the median increment of C-peptide secretion was shown to be 57% lower with tacrolimus than with ciclosporin-based therapy, and the median increment of insulin secretion was 48% lower ($P < 0.05$). Notably, this divergence disappeared thereafter (Figure 3), and there were no significant differences between tacrolimus- and ciclosporin-treated patients for any of the glucose metabolism parameters during the rest of the 3-year follow-up. In addition to showing tacrolimus and ciclosporin equivalence in terms of glucose metabolism after the early post-transplant period, these findings indicate that the long-term use of tacrolimus or ciclosporin is not associated with chronic, cumulative effects on pancreatic $\beta$ cells.

In support of the finding regarding tacrolimus and ciclosporin equivalence are the results of a fourth study, which examined the effect on glucose metabolism of converting 16 stable, ciclosporin-treated patients to tacrolimus after completion of randomized, controlled trials [9]. This study showed that the mean $k_C$ remained stable 9 weeks after conversion to tacrolimus (from 1.28 to 1.41%/min; $P = \text{NS}$). This is an important finding, showing that glucose metabolism is not altered after converting patients from ciclosporin to tacrolimus. Therefore, it is unlikely that renal transplant recipients who experience glucose metabolism disturbances >3 months post-transplant will benefit from changing treatment from tacrolimus to ciclosporin.

**Relative contribution of tacrolimus and corticosteroids to PTDM**

The data presented herein suggest that patients at risk of PTDM would benefit from immunosuppressive regimens that eliminate corticosteroids and/or employ reduced doses of a calcineurin inhibitor. However, both interventions may potentially increase the risk of acute rejection, which is a primary concern. As well as directly damaging the graft, rejection episodes necessitate bolus corticosteroid doses, which are in themselves diabetogenic.

Findings from a recently completed prospective study suggest that early corticosteroid withdrawal from tacrolimus-based immunosuppression is feasible without increasing the risk of acute rejection [10]. The study included 62 renal transplant recipients randomized to undergo corticosteroid cessation after day 7 post-transplant ($n = 28$) or to receive a corticosteroid dose reduction protocol in which the dose was tapered over a period of 3 months prior to being withdrawn ($n = 34$). All patients received tacrolimus at a starting dose of 0.10–0.15 mg/kg twice daily, adjusted to achieve trough levels of 15–20 ng/ml in the first 2 weeks, 10–15 ng/ml during weeks 2–4 and gradually reduced to 5–7 ng/ml after month 6. After a median follow-up of 2.7 years, two patients in the corticosteroid stop group vs 10 patients in the corticosteroid taper arm developed new-onset diabetes mellitus, defined as the use of oral antidiabetic medication or insulin at any time after transplantation. This difference between the treatment groups reached statistical significance ($P = 0.04$). While all patients in the taper group developed diabetes in the first 4 months post-transplant, patients in the stop group developed diabetes after the first year. Later development of PTDM may be due to mechanisms consistent with diabetes mellitus in the general population rather than a direct effect of immunosuppressive therapy or transplantation [4]. Importantly, the early elimination of corticosteroids did not put patients at increased risk of PTDM. 

![Fig. 3. A 3 year follow-up comparing insulin release in renal transplant patients randomized to tacrolimus-based therapy ($n = 11$) and ciclosporin-based treatment ($n = 12$) [8].](image-url)
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of new-onset diabetes mellitus, defined as a 25% increase in acute rejection in the two treatment arms [10].

The feasibility of this approach has been confirmed recently in multicentre trials in Europe and the USA. In a large, open, European study, kidney transplant patients were randomized to treatment based on tacrolimus plus MMF in combination with either daclizumab (n = 260) or corticosteroids (n = 278) [11]. At 6 months post-transplantation, the incidence of new-onset diabetes mellitus, defined as ≥30 days insulin dependence in patients not diabetic pre-transplant, was reported to be just 0.4% in the corticosteroid-free group vs 5.4% in the corticosteroid maintenance group (P = 0.001). Likewise, a US study of 301 renal transplant patients followed-up for 3 years found that early corticosteroid withdrawal (at post-operative day 6) from an immunosuppressive regimen comprising antithymocyte globulin, tacrolimus or ciclosporin, in addition to either MMF or sirolimus, resulted in a PTDM incidence of 0% [12]. In both studies, there was no evidence of a significant increase in acute rejection or graft loss with corticosteroid avoidance and withdrawal.

These studies underline the importance of selecting the optimum immunosuppressive regimen in patients at risk of PTDM. Although a regimen based on a combination of tacrolimus and corticosteroids has the potential to increase the risk of PTDM, it is evident from the findings of the steroid minimization studies that tacrolimus therapy minus corticosteroids results in a very low incidence of PTDM. Thus, it may be postulated that tacrolimus-based immunosuppressive regimens employing early corticosteroid withdrawal or corticosteroid avoidance will be of considerable benefit to transplant recipients at risk of PTDM.

Management of PTDM at the University Hospital Maastricht

As discussed, early elimination or total avoidance of corticosteroids from tacrolimus-based regimens, as well as lower doses/blood levels of tacrolimus minimize the development of post-transplant glucose metabolism disorders. On the basis of these findings, we at the University Hospital Maastricht have developed an algorithm for managing PTDM in renal transplant patients receiving tacrolimus-based immunosuppressive therapy (Figure 4) [4].

During the first month post-transplant, it is important to maintain therapeutic levels of tacrolimus (10–20 ng/ml) to minimize the risk of acute rejection. After month 1, and in the absence of graft rejection, the procedure involves rapid corticosteroid withdrawal followed by a progressive reduction in tacrolimus trough levels to ~5 ng/ml at month 3. These measures should result in improvement or reversal of glucose metabolism disturbances in most patients. At this time, it is important to taper concomitant insulin and/or hypoglycaemics to prevent hypoglycaemia. If patients do not respond to the management protocol, insulin resistance should be determined and, if high, the involvement of infection or obesity should be considered and treated as appropriate. For any remaining patients who still show signs of PTDM, changing the immunosuppressive regimen may be an option, although the benefit of this is questionable because of a need to reintroduce corticosteroids and the risk of acute rejection. Conversion from tacrolimus to ciclosporin is illogical because the potential for glucose disturbance after month 3 post-transplant is equivalent for both agents. Replacing tacrolimus with an adjunctive agent such as MMF or sirolimus is a possibility, but again the risk of acute rejection presents as a major limitation to this approach.

Conclusions

On the basis of these studies, it can be concluded that tacrolimus and ciclosporin tend to reduce insulin release, while concomitant use of corticosteroids increases insulin resistance. The effects of tacrolimus on insulin release are dose related and reversible, even after 2 years; thus, reducing the dose of tacrolimus by ~30% within the target range has been shown to result in a 24% increase in insulin production. There is also good evidence showing that early withdrawal or avoidance of corticosteroids from tacrolimus-based regimens results in a very marked reduction in the incidence of PTDM [10–12]. These findings are consistent with a higher prevalence of PTDM in the early post-transplant period when high levels of calcineurin inhibitors and corticosteroids are used. Importantly, a comparison of tacrolimus and ciclosporin has shown that long-term glucose metabolism is not significantly different between the two immunosuppressants, and that for tacrolimus-treated patients who experience persistent glucose dysregulation >3 months post-transplantation, no value is to be gained by switching therapy to ciclosporin. Indeed, conversion...
from tacrolimus to ciclosporin may have detrimental consequences on blood pressure and cholesterol levels, as demonstrated recently by Ligtenberg et al. [13]. In this study, converting patients from ciclosporin to tacrolimus led to reductions of 10 mmHg in daytime mean arterial blood pressure (MAP; from 114 to 104 mmHg) and of 1 mmol/l in total cholesterol (from 6.1 to 5.1 mmol/l). These beneficial changes were reversed when patients converted back to ciclosporin (daytime MAP increased to 113 mmHg; total cholesterol increased to 6.0 mmol/l). In conclusion, while both tacrolimus and ciclosporin can induce glucose metabolism disturbance and PTDM in at-risk patients, corticosteroid avoidance or early withdrawal is the best available preventative measure. Corticosteroid minimization together with dose-optimized tacrolimus cornerstone immunosuppression is likely to be the most favourable option in this patient cohort.

Conflict of interest statement. The authors conducted several studies for Fujisawa, Novartis and Roche; J.P. van Hooff has served as consultant to Fujisawa, Novartis and Wyeth.

References