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Tacrolimus Interaction with Azole Antifungals in Kidney Transplant Recipients: Is Fluconazole or Clotrimazole a Worse Offender?

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Abstract: ***The utilization of rabbit ant thymocyte globulin induction with tacrolimus-based maintenance immunosuppression may increase the risk for opportunistic fungal infections, particularly oral-esophageal candidiasis. Both are commonly prescribed for deceased donor kidney transplant recipients Tacrolimus (TAC), a calcineurin inhibitor, is metabolized hepatically by cytochrome P450 3A4/5 enzymes. Due to this, a drug interaction can occur with TAC and either fluconazole (FCZ) or clotrimazole (CTMZ). Both are commonly used for antifungal prophylaxis. While both FCZ and CTMZ inhibit CYP3A4/5, systemic absorption of CTMZ is minimal, theoretically limiting the interaction with TAC and reducing the need for dose readjustments. Medical records for adult patients receiving a renal transplant between March 2009 and September 2011 were retrospectively reviewed for TAC dose adjustments following discontinuation of the antifungal agent required to maintain therapeutic TAC blood levels. The change in TAC dose: trough ratio 4-8 weeks after azole discontinuation was greater in patients receiving FCZ compared to CTMZ (FCZ +92.9% vs CTMZ +43.4%, p=0.004). In addition, the proportion of patients requiring ≥30% TAC dose increase was 70% with FCZ versus 45% with CTMZ (p=0.006). The choice of antifungal also did not affect the number of sub-therapeutic TAC levels, the number of patients with sub-therapeutic levels pre- or post-discontinuation, or incidence of biopsy-proven allograft rejection.***

Keywords: ***Tacrolimus, Fluconazole, Clotrimazole, Kidney transplant***

I. INTRODUCTION

Tacrolimus (TAC) is an oral calcineurin inhibitor commonly used as a maintenance immunosuppressant in renal transplant recipients.

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The potent immunosuppression provided by TAC-based maintenance immunosuppression and rabbit antithymocyte globulin induction prevent graft rejection for deceased donor kidney transplant recipients; however, it may also increase the patients' risk for opportunistic bacterial, viral, and fungal infections. To limit the risk of fungal infections, particularly oral-esophageal candidiasis, renal transplant patients at our institution are given anti-fungal prophylaxis for the first 60 to 90 days after kidney transplant. Interactions between TAC and anti-fungal agents are widely accepted, yet significant inter-patient variability in the effect of the interaction exists. The magnitude of interaction between FCZ and CYP3A4/5 substrates is believed to be concentration-dependent, which is determined by FCZ dose as well as renal clearance (the primary route of FCZ elimination). Further, although drug interactions with FCZ are thought to be mediated primarily through CYP3A4, CYP3A5 has also been shown to play a significant role in TAC metabolism.

Prior to August 2010, fluconazole (FCZ) 200 mg daily (adjusted to renal function) for 3 months' post-transplant was our standard anti-fungal prophylaxis regimen for kidney recipients. TAC is metabolized by several CYP450 enzymes, including CYP3A4 and CYP3A5, while FCZ is a known CYP3A4/5 inhibitor. Upon discontinuation of FCZ at post-operative day 90, the patient's dose of TAC generally required an upward titration in the outpatient clinic to maintain therapeutic levels of immunosuppression. The clinician's impression was that this precaution was not being consistently performed in a timely fashion, resulting in prolonged sub-therapeutic levels for numerous patients. Due to this, a switch was made in the anti-fungal regimen to clotrimazole (CTMZ) troches, in hopes of reducing the magnitude of the drug interaction between TAC and FCZ, as well as the urgency for associated dose readjustments. While CTMZ has also been shown to inhibit CYP3A4, and presumably 3A5, this agent undergoes minimal systemic absorption compared to FCZ tablets, theoretically limiting the potential for interaction with TAC and reducing the need for dose readjustments. Several studies have demonstrated an interaction between TAC and FCZ, as well as TAC and CTMZ in renal transplant patients. Authors reported a decrease in the TAC dose of 40-75% was necessary to avoid supra-therapeutic TAC levels when initiating FCZ [1-2]. When CMTZ was initiated, authors reported ~2.4-fold increases in TAC levels in kidney recipients [3] and ~2-fold increase in liver recipients [4], respectively.

Whereas in another case report, authors stated that the TAC level decreased by 60% upon the discontinuation of prophylactic CTMZ [5].

The aim of this study was to compare the change in the TAC dose: blood level ratio and the associated TAC dose adjustments necessary to achieve therapeutic TAC levels after the discontinuation of either FCZ or CTMZ in adult renal transplant recipients. In addition, we determined the prevalence of therapeutic, supra-therapeutic and sub-therapeutic TAC levels during the restabilization period. We also examined if the choice of anti-fungal agent was associated with differing clinical outcomes, including the overall incidence of graft rejection during the first-year post-transplant, and whether sub-therapeutic levels during the period to restabilization of TAC dosing were associated with an increased incidence of rejection.

II. MATERIAL AND METHODS

A retrospective chart review of adult renal transplant recipients receiving tacrolimus and FCZ or CTMZ anti-fungal prophylaxis was performed. Patients were identified using the Transplant Institute and United Network for Organ Sharing computerized databases. Data was collected from Cerner, Teleresults, Transplant Electronic Chart (post-April 2011), and outpatient paper medical charts or their electronic archive (pre-April 2011).

Approval was obtained from the Institutional Review Board at the University of Tennessee Health Science Center (IRB number 12-02182-XM). Patients were included if they had received a solitary kidney transplant at Methodist University Hospital and received post-transplant immunosuppression with TAC and anti-fungal prophylaxis with either FCZ or CTMZ. Exclusion criteria included receipt of an immunosuppressive agent other than TAC or anti-fungal other than FCZ or CTMZ, a fungal infection requiring therapeutic doses of anti-fungal medications within the first 5 months post-transplant, failure to follow-up in the outpatient transplant clinic for at least 12 months post-transplant, renal allograft loss within the first 3 months post-transplant, documented medication non-compliance, non-continuous use of another potentially interacting medication pre- and post-anti-fungal discontinuation, or fewer than 3 TAC blood levels prior to and in the 4-8 week window after anti-fungal discontinuation.

The primary endpoint was the average change in TAC dose: trough level ratio pre-antifungal discontinuation to the 4–8-week post-discontinuation period for patients receiving FCZ versus CTMZ. Secondary endpoints included comparison of the number of patients with sub-therapeutic TAC levels 4-8 weeks and 6 months after anti-fungal discontinuation, number of sub-therapeutic TAC levels within 8 weeks and 6 months of discontinuation, number of patients requiring ≥30% increase in TAC dose, and the rate of biopsy-proven acute rejection in patients receiving FCZ or CTMZ.

Data was collected on adult renal transplant patients who received their transplant August 2010 – September 2011 (CTMZ group). Data was also collected on an equivalent number of patients who received their transplant in the period immediately prior to August 2010 (FCZ group). The TAC dose: blood trough ratio was calculated using the three TAC trough levels immediately prior to and at least three levels 4-8 weeks after discontinuation of the anti-fungal agent, allowing for at least one-week washout period after discontinuation. For patients with large fluctuations in trough levels, up to five levels will be collected, with the two most extreme values being discarded (values not within 25% of the intra-individual mean). TAC blood levels measured outside of the 9–15-hour trough target window were omitted from collection/analysis, if that information was documented; repeat levels within 2 weeks that are documented as per clinic notes were substituted, if clearly within the pre- vs post- azole period. An additional TAC dose: blood trough ratio was collected at 6-month time post-transplant.

Data was analyzed using IBM SPSS Statistical software. Continuous, non-parametric data was analyzed using the Mann-Whitney U test. Nominal data was analyzed using the Chi Square test. Primary and secondary endpoints were compared among the FCZ and CTMZ groups with a two-sided hypothesis using alpha = 0.05. Estimated glomerular filtration rate (eGFR) was calculated using the Modification of Diet in Renal Diet equation.

III. RESULTS

A. Baseline Characteristics

Sixty patients were enrolled in each cohort. Baseline characteristics were similar in each of the groups (Table 1). At the time of discontinuation, an equivalent number of patients had an eGFR<50 ml/min/1.73m^2, and all patients in both cohorts were on full doses of FCZ (200mg tablets daily) or CTMZ (10mg troches TID). There was no difference in the proportion of African American (AA) or non-AA patients between groups. However, overall, the AA population was 70% in this study.

B. Change in TAC Dosing Post-Anti-Fungal Discontinuation

Patients receiving FCZ had a significantly larger increase in TAC dose: trough level ratio at 4-8 weeks post-discontinuation compared to the pre-discontinuation ratio than patients receiving CTMZ (+93% vs. +43%, p=0.004) (Table 2). The increase in TAC dose (mg TAC/patient weight in kg) was also significantly increased 4-8 weeks after FCZ discontinuation versus CTMZ discontinuation (+53% vs. + 23%, p=0.01).

At 6 months after FCZ or CTMZ discontinuation, there was a borderline significant difference between TAC dose: trough level ratio (+89% vs. +57%, p=0.046) but no difference in TAC mg/kg dose (+41% vs. +44%, p=0.773), when compared to the pre-discontinuation ratio and dose. From 4-8 weeks to 6 months' post-discontinuation, there was no difference in the TAC dose: trough level ratio in patients on FCZ or CTMZ (+0.6% vs. +11%, p=0.16). However, patients post-CTMZ had a significantly larger increase in TAC mg/kg dose than patients post-FCZ in the 4-8 weeks to 6-month time (-4.3% FCZ vs. +14% CTMZ, p=0.001).

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We also examined the proportion of patients requiring ≥30% increase in TAC dose after the anti-fungal medication was discontinued. We chose 30% as our cut-off because that was the recommended empiric increase in TAC dose at the time of anti-fungal discontinuation in the protocol from our institution's transplant clinic. More patients required ≥30% increase in TAC dose within the first 8 weeks with FCZ than with CTMZ when compared to the pre-discontinuation dose (70% vs. 45%, p=0.006). However, by the 6-month time point, there was no longer a difference between the cohorts (63% (FCZ) vs. 55% (CTMZ), p=0.35).

Due to the potential effect of renal function on the clearance of FCZ, we examined whether an eGFR <50 ml/min/1.73m^2 affected the change in TAC dose: trough level ratio (Table 3). All patients were on the full dose of FCZ (200 mg PO daily) and CTMZ (10 mg PO TID) at the time of anti-fungal discontinuation. However, we found that there was no difference in the percent change in the ratio between patients with normal or impaired renal function.

C. Proportion of Sub-Therapeutic Levels Pre- and Post-Anti-Fungal Discontinuation

The anti-fungal used for prophylaxis did not affect the number of patients with at least one sub-therapeutic TAC level before or after discontinuation (Table 4). There was also no difference in the proportion of sub-therapeutic TAC levels while patients were on FCZ or CTMZ or after the medication was stopped.

D. Biopsy-Proven Acute Rejection

The rate of biopsy-proven kidney acute rejection following anti-fungal discontinuation was not different in the two cohorts. There were 2 episodes of rejection in the group receiving FCZ and 3 episodes in the group receiving CTMZ (p=1.0). All 5 episodes were found to be acute T-cell-mediated rejection.

IV. DISCUSSION

Both FCZ and CTMZ are commonly used in many kidney transplant protocols to prevent fungal infections in the first few months post-operatively. At our institution we recently changed from FCZ to CTMZ to try to limit the required dose adjustments for TAC. Since CTMZ is administered as troches, it is believed to be less systemically absorbed than FCZ tablets, thereby limiting the magnitude of the drug interaction with TAC. We found that by switching to CTMZ, the increase in TAC dose necessary to keep the trough level within the therapeutic range 4-8 weeks after the discontinuation of CTMZ was significantly less than when patients received FCZ. However, by 6 months post-anti-fungal, the difference between the cohorts had become non-significant. Interestingly, renal function did not affect the change in TAC dose: trough level ratio, even in the FCZ group. We chose to use TAC dose: trough level ratio as the measure of TAC dose adjustments. According to the protocol at our institution, TAC trough goals typically decrease at post-transplant month 3, which is also when the anti-fungal medication was discontinued. By using the ratio, we were able to control for absolute changes in TAC dose, due to the lower goal TAC level. In addition, each patient was able to serve as their own control to determine the proportional change pre- and post-anti-fungal discontinuation, which provided a clear measure of the pharmacokinetic interaction between TAC and the anti-fungal agent. Despite the difference in TAC dose: trough level ratio change, there was no difference in the number of patients that had at least one sub-therapeutic TAC level within 8 weeks and at 6 months after discontinuing FCZ or CTMZ. Notably, although there was no difference between the groups, 80% of patients on FCZ and 65% of those on CTMZ had ≥1 sub-therapeutic level in the first 2 months following anti-fungal discontinuation. In addition, choice of anti-fungal did not affect the incidence of biopsy-proven acute rejection in either of the cohorts after patients stopped taking FCZ or CTMZ. While the authors of several of the articles make specific recommendations on TAC dose adjustments based on their results, significant questions remain. Of note, most studies have included a relatively small proportion of AA patients, leading to uncertainty regarding the applicability of the findings to the patient population at our institution, of which two-thirds are AA. In addition, none of the published studies have directly compared the effects of FCZ and CTMZ on TAC dosing. We found that AA patients had significantly higher TAC dose requirements than non-AA patients. This difference may likely be due to the higher prevalence of the CYP3A5*1 allele expression in the AA population as previously reported [6-7]. This study is not without limitations. We may have lacked a sufficient number of patients for meaningful differences in some of our parameters. As this was a retrospective study, we were limited in the number of patients available for data collection. We were also unable to control confounders, such as concomitant disease states, which may influence metabolism and dose changes in TAC for an individual patient. This was primarily an outpatient study, and it may not be possible to identify all patients who were non-compliant with their medication regimen, if this is not documented or the patient does not admit to non-compliance, or if trough levels were not measured 12-hours post-TAC dose. In addition, the impact of other concurrent medications on the interaction between TAC and anti-fungal agents was not examined. Importantly, the renal transplantation protocol at our center includes long-term steroid use post-transplant, which has been shown previously to decrease the bioavailability of TAC [8]. Due to the retrospective nature of this study, we were not able to obtain blood samples to identify genotypes for patients in this study.

V. CONCLUSION

In summary, we found that there was a smaller change in the TAC dose: trough level ratio after discontinuation of CTMZ than with FCZ. In addition, significantly more patients required ≥30% increase in the TAC dose after discontinuing FCZ when compared to CTMZ. The drug interaction between CTMZ and tacrolimus was less significant than with FCZ. Despite this, there was no significant effect on the prevalence of biopsy-proven acute cellular rejection between groups.

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Ethical Approval and Consent to Participate	No, the article does not require ethical approval and consent to participate with evidence.
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Authors Contributions	All authors have equal participation in this article.

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Table 1: Patient Demographics and Baseline Characteristics

	FCZ (n=60)	CTMZ (n=60)	p-value
Male	30 (50%)	31 (52%)	NS
African-American (AA)	40 (67%)	44 (73%)	NS
Non- AA	20 (33%)	16 (27%)	NS
Age (years)	48 ± 13	49 ± 14	NS
eGFR <50 ml/min/ 1.73m2	27 (45%)	28 (47%)	NS
Average Dose	200mg daily (100%)	10mg TID (100%)	N/A

Table 2: Comparison of Average Changes in TAC Dose: Trough Level Ratios and Dose Requirements Pre- and Post-Antifungal Discontinuation

	FCZ (n=60)	CTMZ (n=60)	p-value
Change TAC dose : trough level ratio			
Pre- to 4-8 weeks post-D/C	+93% (52 - 150)	+43% (3.0 - 115)	0.004
Pre- to 6 months post-D/C	+89% (37-134)	+57% (17-102)	0.046
Change TAC mg/kg			
Pre- to 4-8 weeks post-D/C	+53% (4.7 - 96)	+23% (-15 to 59)	0.01
Pre- to 6 months post-D/C	+41% (-7.7 to 81)	+43.5% (-5.8 to 82)	0.77
≥30% increase in TAC dose			
Pre- to 4-8 weeks post-D/C	70%	45%	0.006
Pre- to 6 months post-D/C	63%	55%	0.36

Table 3: Impact of Renal Function on TAC dose: Trough Level Ratio

	FCZ (n=60)			CTMZ (n=60)		
	eGFR <50	eGFR >50	p-value	eGFR <50	eGFR >50	p-value
Change TAC dose : trough level ratio						
Pre- to 4-8 weeks post-D/C	+92% (58 - 173)	+94% (36 - 125)	0.19	+48% (5.6 - 101)	+38% (3.6 - 124)	0.4
4-8 weeks to 6 months post-D/C	-0.5% (-24 to 17)	+1.6% (-24 to 22)	0.9	+11% (-5.5 to 44)	+5.7% (-23 to 41)	0.48

Table 4: Sub-Therapeutic TAC Blood Levels Pre- and Post- Antifungal Discontinuation

	FCZ	CTMZ	p-value
# patients with sub-therapeutic levels			
Pre- D/C	21/60 (35%)	26/60 (43%)	0.35
4-8 weeks post- D/C	48/60 (80%)	39/60 (65%)	0.07
# sub-therapeutic levels			
Pre- D/C	23/180 (13%)	36/180 (20%)	0.09
4-8 weeks post- D/C	123/345 (36%)	75/243 (31%)	0.26

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