Comparison of the influence of cyclosporine and tacrolimus on the pharmacokinetics of prednisolone in adult male kidney transplant recipients

Troels K. Bergmann (1,2), Nicole M. Isbel (3), Katherine A. Barraclough (3), Scott B. Campbell (3), Brett C. McWhinney (4), Christine E. Staatz (1).

(1) School of Pharmacy, University of Queensland, Brisbane, Australia; (2) Department of Clinical Pharmacology, Aarhus University Hospital, Aarhus, Denmark; (3) Department of Nephrology, University of Queensland at the Princess Alexandra Hospital, Brisbane, Australia; (4) Department of Chemical Pathology, Pathology Queensland, Royal Brisbane and Women's Hospital, Brisbane, Australia.

Corresponding author:

Troels K. Bergmann

Department of Clinical Pharmacology, Aarhus University Hospital, Aarhus, Denmark.

Wilhelm Meyers Alle 4

8000 Aarhus C. Denmark.

Ph: +45-61704711

Email: tbergmann@health.sdu.dk

Short title: Cyclosporine prednisolone interaction study

This is the postprint version of the following article: Bergmann, Troels K., Isbel, Nicole M., Barraclough, Katherine A., Campbell, Scott B., McWhinney, Brett C. and Staatz, Christine E. (2014) Comparison of the influence of cyclosporine and tacrolimus on the pharmacokinetics of prednisolone in adult male kidney transplant recipients. Clinical Drug Investigation, 34 3: 183-188. doi:10.1007/s40261-013-0162-1

Abstract

Background and objective

Cyclosporine has been observed to precipitate Cushingoid features in kidney transplant recipients already on prednisolone. Some pharmacokinetic studies have demonstrated increased prednisolone exposure in patients on cyclosporine therapy compared to azathioprine, whereas other studies have found no difference. The objective of this study was to determine whether cyclosporine impacts on prednisolone exposure as compared to tacrolimus.

Methods

Adult male kidney transplant recipients treated with prednisolone and either cyclosporine or tacrolimus were recruited for pharmacokinetic blood sampling at the out-patient clinic at the Princess Alexandra Hospital, Brisbane, Australia. Prednisolone plasma concentrations were determined using ultra high performance liquid chromatography. Dose adjusted area under the plasma concentration-time curve (AUC) of free and total prednisolone was estimated using a previously developed limited sampling strategy and non-compartmental analysis.

Results

A total of 55 patients were eligible for analysis; 38% received cyclosporine and 62% received tacrolimus co-therapy. No significant difference in mean dose adjusted total prednisolone AUC from 0 to 6 hours post-dose or mean dose adjusted free prednisolone AUC from 0 to 12 hours was observed between the cyclosporine and tacrolimus groups (449 versus 428 nmol·h/L/mg, p=0.43 and 32 versus 30 nmol·h/L/mg, p= 0.51, respectively).

Conclusion

Cyclosporine does not change the dose adjusted exposure of prednisolone compared to tacrolimus. Adult kidney transplant recipients can therefore continue on their usual prednisolone dose when changing therapy between cyclosporine and tacrolimus.

1 Introduction

Cyclosporine is known to interact with a range of different drugs, with notable examples including statins [1, 2], digoxin [3, 4], ezetimibe [5], caspofungin [6] and aliskiren [7]. The unifying suspected mechanism for these interactions is cyclosporine inhibition of the efflux transporter P-glycoprotein (p-gp) and/or other efflux or influx transporters such as Organic anion transporter peptide 1B1 (OATP1B1) [1, 2]. Inhibition of prednisolone apparent clearance (CL/F) by cyclosporine, in the broad sense, was first hypothesised in 1984. This was based on the observation that kidney transplant patients on cyclosporine were more likely to display cushingoid appearance than patients on azathioprine. The authors tested this hypothesis and indeed found that the CL/F of total prednisolone was significantly higher in 27 kidney transplant recipients on azathioprine compared with 10 recipients on cyclosporine [8]. However, six subsequent studies, also using azathioprine as comparator, arrived at conflicting conclusions [9-14]. It therefore remains an open question whether or not cyclosporine inhibits the CL/F of prednisolone in solid organ transplant recipients. Given that prednisolone remains a cornerstone in immunosuppression therapy in solid organ transplantation, it is important to determine whether switching patients from or to cyclosporine affects prednisolone treatment.

We present a prospective pharmacokinetic study specifically designed to compare differences in dose adjusted exposure to free and total prednisolone in adult male kidney transplant recipients treated with either cyclosporine or tacrolimus.

2 Methods

2.1 Participants

A prospective, observational cohort study was conducted between August 2011 and May 2012. Adult (≥18 years) male kidney transplant recipients on a daily oral maintenance dose of prednisolone between 5 and 12.5 mg were recruited into the study. To be eligible, subjects were required to have had undergone living or deceased donor kidney transplantation at least 12 months prior to enrolment and be receiving ongoing outpatient follow-up at the Princess Alexandra Hospital, Brisbane, Australia. In addition, subjects had to be clinically stable and receiving concurrent immunosuppressant therapy with either cyclosporine or tacrolimus. Exclusion criteria included 1) a history of alcohol abuse or significant liver disease, 2) glucocorticoid therapy for any other reason apart from the prevention of rejection and 3) treatment with pulse steroids in the previous 2 months. On the day of blood sampling the patients' medical history was recorded. Prednisolone and calcineurin inhibitor dose changes within the previous 7 days were not allowed. Medications were screened for p-gp and cytochrome P450 3A4 (CYP3A4) inhibitors/inducers. The study protocol

adhered to the declaration of Helsinki and was approved by the Princess Alexandra Hospital and University of Queensland Ethics Committees. All participants provided written informed consent.

2.2 Drug analysis

Each subject's prednisolone exposure was measured on a single occasion in the outpatient setting. Prior to arrival at the hospital, subjects were instructed to consume a light, non-fatty breakfast. All blood samples were taken via a peripherally inserted intravenous cannula. Blood samples were collected in EDTA tubes immediately prior to, and 1, 2, 4, and 6 hours after morning medications were taken. Plasma was prepared within 30 minutes by 10 minutes centrifugation at approximately 1750g. Plasma was stored at -20 °C. until analysis. Total and free prednisolone concentrations were determined in plasma using ultra high performance liquid chromatography (UHPLC) with tandem mass spectrometric detection [15]. For free prednisolone measurement, plasma ultrafiltrate was prepared by temperature-controlled ultrafiltration. Limit of quantitation for prednisolone was 5 nmol/L and linearity extended to 2000 nmol/L. The intra-assay CV was <5% (4.9%, 3.9% and 2.3% for 23 nmol/L, 125 nmol/L and 533 nmol/L respectively) and the inter-assay CV <10% for all analytes.

2.3 Estimation of exposure

It was decided *a priori* to estimate prednisolone exposure employing limited sampling strategies (LSS) previously developed in a cohort of kidney transplant recipients at the Princess Alexandra Hospital [16]. Total prednisolone AUC (area under the concentration-time curve) from 0 to 6 hours was estimated using equation 1:

$$\boldsymbol{Dose\ Adjusted\ AUC}_{\boldsymbol{0-6\ hours}}^{\boldsymbol{Total\ prednisolone}} = \frac{(\mathbf{11.155+2.906\times C_2+2.105\times C_4})}{\boldsymbol{Prednisolone\ Dose}}$$

,where C is the total prednisolone concentration sampled after 2 and 4 hours. Free prednisolone AUC from 0 to 12 hours was estimated using equation 2:

$$\boldsymbol{Dose\ Adjusted\ AUC}_{\boldsymbol{0-12\ hours}}^{\boldsymbol{Free\ prednisolone}} = \frac{(\mathbf{1.09+0.79\times C_1+1.95\times C_2+4.49\times C_4})}{\boldsymbol{Prednisolone\ Dose}}$$

,where C is the free prednisolone concentration sampled after 1, 2 and 4 hours. For completeness in a secondary analysis, AUC from 0 to t_{max} ($AUC_{0\text{-}tmax}$) and from 0 to infinity ($AUC_{0\text{-}inf}$), the maximum measured concentration (C_{max}) and the elimination rate constant (k_e) of total and free prednisolone were also estimated based on non-compartmental analysis using the linear trapezoidal rule (with any extrapolation done assuming first-order pharmacokinetics). All exposure parameters were adjusted for prednisolone dose. All calculations were carried out using STATA 12.1

(StataCorp, College Station, Texas, USA).

2.4 Statistical analysis

A pre-study power calculation determined that 53 subjects were sufficient to detect an 8 nmol·h/L/mg difference in free prednisolone dose-adjusted exposure, with a power of 80% and an alpha of 5%. Based on a previous study [16], the standard deviation (SD) was assumed to be 10 nmol·h/L/mg. Descriptive statistics used were median with 10^{th} and 90^{th} percentile for continuous variables and percentages for categorical variables. Equality tests of patient characteristics were done using Wilcoxon rank-sum test. Parameter distribution plots and a Shapiro-Wilk test were used to check the pharmacokinetic parameters for departure from approximate normal distribution (results not shown). A students T-test or Wilcoxon rank-sum test was then used as appropriate, depending on the P-value of the Shapiro-Wilk test with a cut-off value of 0.05. Regression analysis was performed to identify significant explanatory covariates on prednisolone pharmacokinetics. All analyses were carried out using STATA 12.1 (StataCorp, College Station, Texas, USA).

3 RESULTS

3.1 Participants

Fifty-nine patients were enrolled in the study. Two patients did not attend for blood sampling and one patient inadvertently took his morning medication prior to blood sampling. One patient was treated with azathioprine, and neither tacrolimus nor cyclosporine. Thus, data from 55 patients were eligible for analysis. Table 1 provides the baseline demographic and clinical characteristics of this cohort. Twenty-one (38%) of the patients were treated with cyclosporine and 34 (62%) were treated with tacrolimus. The groups were comparable with regard to age, body weight, glomerular filtration rate (GFR), haematocrit and albumin. Time post-transplantation trended towards being greater in the cyclosporine group explaining the slightly lower prednisolone dose in this group (a result of the common practice to slowly taper prednisolone dose over time). Total bilirubin was normal in both groups but slightly, and statistically significantly, greater in the cyclosporine group (Table 1). Use of known CYP3A4 and/or p-gp inhibitors diltiazem and/or lercanidipine was more frequent in the cyclosporine compared with the tacrolimus cohort (76% versus 44%) (Table 1).

3.2 Dose adjusted prednisolone exposure

Figure 1 shows total and free prednisolone concentration-time profiles obtained in the 55 patients included in the final analysis. Prednisolone pharmacokinetic parameters in patients receiving cyclosporine versus tacrolimus are compared in Table 2. Primary analysis: Based on LSS estimation, no significant difference in mean (SD) dose adjusted total prednisolone AUC from 0 to 6 hours post-dose ($dAUC_{0-6}$) or mean (SD) dose adjusted free prednisolone AUC from 0 to 12 hours ($dAUC_{0-12}$) was observed between the cyclosporine and tacrolimus groups [449 (86) versus 428 (103) nmol·h/L/mg, p=0.43 and 32 (11.0) versus 30 (9.5) nmol·h/L/mg, p= 0.51, respectively]. Secondary analysis: The elimination constant of free prednisolone k_e was significantly lower in the cyclosporine group [0.164 h^{-1} (0.023) versus 0.175 h^{-1} (0.028), p=0.04]. All other pharmacokinetic parameters were similar between the groups. Regression analysis indicated that body weight was an important covariate for prednisolone dAUC. However, even after adjusting for this, no significant difference in pharmacokinetic parameters was detected between the groups (data not shown). There was no significant effect of use of diltiazem and/or lercanidipine on prednisolone pharmacokinetic parameters (data not shown). A single patient reported use of carbamazepine.

4 DISCUSSION

This study is the first to compare the pharmacokinetics of prednisolone in adult male kidney transplant recipients on long term cyclosporine versus tacrolimus therapy. No detectable impact of cyclosporine on total and free prednisolone exposure was observed, with the implication being that no adjustment in prednisolone dose is necessary when switching a transplant patient between calcineurin inhibitors.

Five studies, with from 16 to 64 subjects, published between 1984 and 2004 compared prednisolone pharmacokinetics in patients treated with azathioprine or cyclosporine [8-10, 12, 14]. All studies found lower prednisolone clearance and/or increased prednisolone AUCs in the cyclosporine group. If it weren't for two contradicting studies published in 1987 and 1988 the interaction would have been firmly established. The study published in 1987 compared 25 kidney transplant recipients treated with cyclosporine with 25 patients on azathioprine[11]. No significant difference between the groups was detected for free (unbound) and total prednisolone metabolic clearance, renal clearance, volume of distribution and systemic availability. The authors questioned the validity of the previous studies, highlighting multiple methodological issues including unequal age and sex distribution, lack of information on the use of other potentially interacting medications, failure to account for prednisolone dose and insufficient duration of pharmacokinetic sampling and not measuring free prednisolone exposure. In 1988 one more study compared clearance of free prednisolone before and after kidney transplantation and also found no difference,

albeit in a small population of 6 patients[13].

These methodological issues have all been addressed by our study. For the first time, we chose to study exclusively male patients.

There is evidence to support an impact of sex, and even hormonal cycle variation, on prednisolone exposure in fertile women [17, 18]. The most recent of the previously published cyclosporine-prednisolone interaction studies reported higher dose adjusted prednisolone exposure (348 nmol·h/L/mg prednisolone) for 28 patients on cyclosporine compared to 15 patients on azathioprine (285 nmol·h/L/mg prednisolone). However, prednisolone AUC was also found to be higher in females compared to males (415 versus 297 nmol·h/L/mg prednisolone) [14], with the highest AUCs seen in cyclosporine treated women on estrogen supplements (median 595 nmol·h/L/mg prednisolone). This suggests that the unbalanced distribution of sex in this study (i.e. 15 women in the cyclosporine group (54%) versus 5 (33%) in the azathioprine group) might at least partially explain the observed difference in prednisolone exposure.

Antihypertensive therapy with lercanidipine or diltiazem was more common in the cyclosporine group (76%) than in the tacrolimus group (44%). This is because diltiazem is a popular cyclosporine adjuvant because of its dose (cost) saving effect, owing to its inhibition of CYP3A4, in addition to controlling the blood pressure (hypertension is a common adverse effect of cyclosporine). In the analysis it is important to control for use of diltiazem and lercanidipine because the exerted inhibition of CYP3A4 and p-gp might affect the pharmacokinetics of prednisolone, which in this case might introduce bias in favour of rejecting the null hypothesis of cyclosporine not interacting with prednisolone pharmacokinetics. However, no effect of lercanidipine and/or diltiazem use on prednisolone pharmacokinetics was detected.

It is well established that it is the free fraction of drugs that are biologically active. Previous studies have therefore also been criticized for only measuring total prednisolone. Moreover, the choice of assay might be of importance. Specifically the use of immunoassays has been criticized for introducing a risk of misinterpretation caused by cross reactivity of prednisolone and cortisol [11, 19]. In the present study we estimated the pharmacokinetic parameters based on both free and total prednisolone concentrations determined exclusively using state of the art ultra-high performance liquid chromatography (UHPLC) with tandem mass spectrometric detection.

The previous studies discussed above all used azathioprine as comparator for cyclosporine. Our study is the first to investigate the cyclosporine prednisolone interaction by using tacrolimus as comparator. Despite tacrolimus being a CYP3A4 substrate, there is no indication in the literature suggesting significant p-gp/CYP3A4 inhibiting or

inductive properties [20, 21]. Theoretically, a study with a control group not using a calcineurin inhibitor would be ideal, but is not a protocol practiced by the Princess Alexandra Hospital transplant unit.

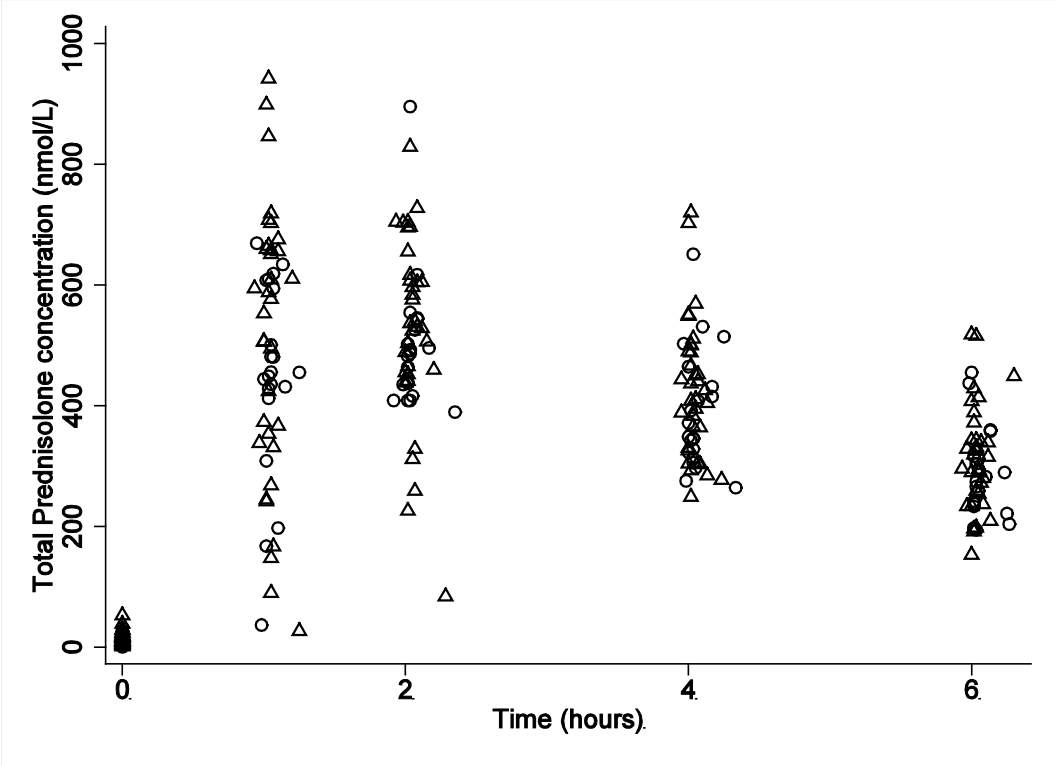
In a secondary analysis k_e of free prednisolone was significantly lower in patients on cyclosporine compared to the tacrolimus. While this did not translate into a significant increase in free prednisolone exposure the finding is interesting because it suggests that cyclosporine may in fact influence elimination of prednisolone, albeit in a subtle and non-clinically relevant way. P-gp plays an excretory role for prednisolone not only in the liver, but also in the kidney (up to 25% of prednisolone is excreted unchanged in the urine [22, 23]), and cyclosporine is extensively bound to liver and kidney tissue [24-26]. One possible explanation for the observed results is that the concentration of cyclosporine in the intestinal mucosa is too low and/or too transient to inhibit the absorption of prednisolone to a degree that impacts on exposure, but high and consistent enough in the liver and/or kidney to reduce the intrinsic free prednisolone clearance in these organs and thereby affecting the slope of the terminal part of the elimination curve. However, it must be borne in mind that this study was not designed to specifically demonstrate a difference in elimination constants and that the k_e estimates are based on limited data only.

5 CONCLUSION

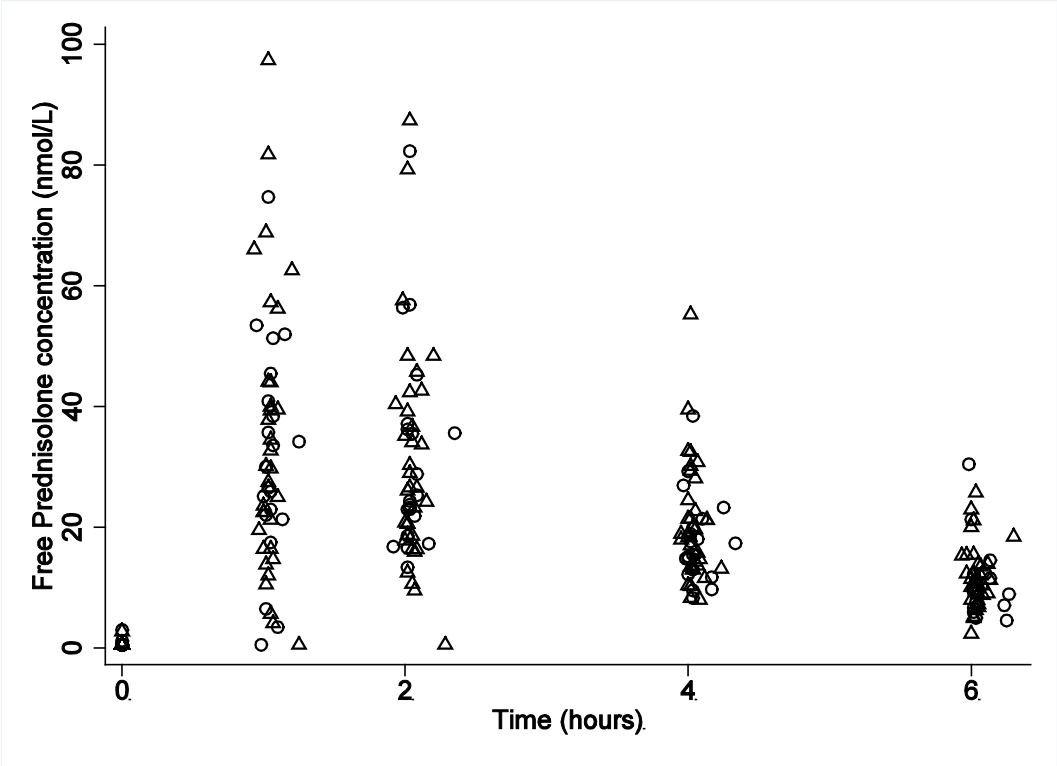
This study did not detect a difference in total and free prednisolone exposure in adult male patients on cyclosporine or tacrolimus therapy. Hence, there is no need for prednisolone dose adjustment when substituting cyclosporine for tacrolimus (or vice versa) in adult male transplantation recipients.

Figure 1. Total (a) and free (b) prednisolone concentrations from 0 to 6 hours post-dose in 55 adult kidney transplant recipients on either tacrolimus (triangles) or cyclosporine (circles).

a)



b)



Tabel 1. Demographic characteristics of the 55 patients studied

Characteristic	Cyclosporine cohort[a]	Tacrolimus cohort[a]	P-value
Number of patients (n) (all male)	21 (20 Caucasians)	34 (32 Caucasians)	-
Age (years)	61 [45-64]	55 [36-64]	0.17
Body weight (kg)	82 [70-119]	85 [66-112]	0.87
Serum creatinine (µmol/L)	132 [104-191]	137 [101-205]	0.48
Glomerular filtration rate[b] (mL/min)	68 [39 - 97]	67 [35-92]	0.63
Haematocrit (fraction)	0.41 [0.37-0.44]	0.41 [0.34-0.46]	0.94
Albumin (g/L)	40 [36-43]	39 [36-43]	0.52
Total bilirubin (µmol/L)	17 [14-24]	14 [8-17]	<0.01
Time post-transplantation (months)	109 [30-189]	43 [16-217]	0.07
Prednisolone dose (mg)	5 [5-6]	5 [5-8]	0.02
Cyclosporine/tacrolimus total daily dose (mg)	175 [100-225]	3 [1.5-8]	-
Concomitant diltiazem and/or lercanidipine (n (%))	16 (76%)	15 (44%)	-
Concomitant mycophenolate mofetil (n(%))	17 (81%)	28 (82%)	-
Concomitant trimethoprim-sulfamethoxazole (n(%))	2 (10%)	6 (18%)	-

a- Results presented as median [10^{th}-90^{th} percentile], unless specified otherwise
b- Calculated using the Cockcroft-Gault equation

Table 2. Comparison of prednisolone pharmacokinetic parameters adjusted for dose in patients receiving cyclosporine versus tacrolimus

Parameter	Cyclosporine cohort[a]	Tacrolimus cohort[a]	P-value
Primary analysis (limited sampling strategy)			
Total prednisolone $dAUC_{0\text{-}6\ h}$ (nmol·h/L/mg)	449 [86]	428 [103]	0.43[c]
Free prednisolone $dAUC_{0\text{-}12\ h}$ (nmol·h/L/mg)	32 [11.0]	30 [9.5]	0.51[c]
Secondary analysis (trapezoidal method[b])			
Total prednisolone dC_{max} (nmol/L/mg)	105 [18]	105 [23]	1.00[c]
Free prednisolone dC_{max} (nmol/L/mg)	7.4 [2.8]	6.7 [2.5]	0.43[d]
Total prednisolone $dAUC_{0\text{-}inf}$ (nmol·h/L/mg)	976 [211]	949 [219]	0.65[d]
Free prednisolone $dAUC_{0\text{-}inf}$ (nmol·h/L/mg)	44.4 [14.9]	41.7 [10.9]	0.45[c]
Total prednisolone k_e (h^{-1})	0.186 [0.040]	0.181 [0.036]	0.50[d]
Free prednisolone k_e (h^{-1})	0.164 [0.023]	0.175 [0.028]	0.04[d]

a – Results presented as mean [SD]
b –Sum of trapezes and extrapolating from 24 hours to infinity using linear extension of last 3 observations and assuming $C_0 = C_{24}$
c – Student's T-test
d – Wilcoxon rank-sum test
dAUC – dose adjusted area under the plasma concentration time curve, dC_{max} – dose adjusted maximal observed plasma concentration, k_e - elimination constant derived using C_4, C_6 and C_{24} (=C_0)

Acknowledgments:

The authors acknowledge financial support from a National Health and Medical Research Council (NHMRC) Project Grant #511109. No author has any conflict of interest to declare.

References

1. Shitara Y, Itoh T, Sato H, et al. Inhibition of transporter-mediated hepatic uptake as a mechanism for drug-drug interaction between cerivastatin and cyclosporin A. J Pharmacol Exp Ther. 2003;304(2):610-6.
2. Hedenmalm K, Alvan G, Ohagen P, et al. Muscle toxicity with statins. Pharmacoepidemiology and drug safety. 2010;19(3):223-31.
3. Dorian P, Cardella C, Strauss M, et al. Cyclosporine nephrotoxicity and cyclosporine--digoxin interaction prior to heart transplantation. Transplant Proc. 1987;19(1 Pt 2):1825-7.
4. Robieux I, Dorian P, Klein J, et al. The effects of cardiac transplantation and cyclosporine therapy on digoxin pharmacokinetics. J Clin Pharmacol. 1992;32(4):338-43.
5. Bergman AJ, Burke J, Larson P, et al. Interaction of single-dose ezetimibe and steady-state cyclosporine in renal transplant patients. J Clin Pharmacol. 2006;46(3):328-36.
6. Sable CA, Nguyen BY, Chodakewitz JA, et al. Safety and tolerability of caspofungin acetate in the treatment of fungal infections. Transpl Infect Dis. 2002;4(1):25-30.
7. Rebello S, Compain S, Feng A, et al. Effect of cyclosporine on the pharmacokinetics of aliskiren in healthy subjects. J Clin Pharmacol. 2011;51(11):1549-60.
8. Ost L. Effects of cyclosporin on prednisolone metabolism. Lancet. 1984;1(8374):451.
9. Langhoff E, Madsen S, Flachs H, et al. Inhibition of prednisolone metabolism by cyclosporine in kidney-transplanted patients. Transplantation. 1985;39(1):107-9.
10. Langhoff E, Madsen S, Olgaard K, et al. Clinical results and cyclosporine effect on prednisolone metabolism of cadaver kidney transplanted patients. Proc Eur Dial Transplant Assoc Eur Ren Assoc. 1985;21:963-8.
11. Frey FJ, Schnetzer A, Horber FF, et al. Evidence that cyclosporine does not affect the metabolism of prednisolone after renal transplantation. Transplantation. 1987;43(4):494-8.
12. Ost L. Impairment of prednisolone metabolism by cyclosporine treatment in renal graft recipients. Transplantation. 1987;44(4):533-5.
13. Rocci ML, Jr., Tietze KJ, Lee J, et al. The effect of cyclosporine on the pharmacokinetics of prednisolone in renal transplant patients. Transplantation. 1988;45(3):656-60.
14. Potter JM, McWhinney BC, Sampson L, et al. Area-under-the-curve monitoring of prednisolone for dose optimization in a stable renal transplant population. Therapeutic drug monitoring. 2004;26(4):408-14.
15. McWhinney BC, Briscoe SE, Ungerer JP, et al. Measurement of cortisol, cortisone, prednisolone, dexamethasone and 11-deoxycortisol with ultra high performance liquid chromatography-tandem mass spectrometry: Application for plasma, plasma ultrafiltrate, urine and saliva in a routine laboratory. J Chromatogr B Analyt Technol Biomed Life Sci. 2010;878(28):2863-9.
16. Barraclough KA, Isbel NM, McWhinney BC, et al. Evaluation of limited sampling strategies for total and free prednisolone in adult kidney transplant recipients. Eur J Clin Pharmacol. 2011;67:1243-52.
17. Boekenoogen SJ, Szefler SJ, Jusko WJ. Prednisolone disposition and protein binding in oral contraceptive users. J Clin Endocrinol Metab. 1983;56(4):702.
18. Magee MH, Blum RA, Lates CD, et al. Prednisolone pharmacokinetics and pharmacodynamics in relation to sex and race. J Clin Pharmacol. 2001;41(11):1180-94.
19. Bergmann TK, Barraclough KA, Lee KJ, et al. Clinical pharmacokinetics and pharmacodynamics of prednisolone and prednisone in solid organ transplantation. Clin Pharmacokinet. 2012;51(11):711-41.

20. de Jonge H, de Loor H, Verbeke K, et al. In vivo CYP3A activity is significantly lower in cyclosporine-treated as compared with tacrolimus-treated renal allograft recipients. Clin Pharmacol Ther. 2011;90(3):414-22.
21. Saitoh H, Saikachi Y, Kobayashi M, et al. Limited interaction between tacrolimus and P-glycoprotein in the rat small intestine. Eur J Pharm Sci. 2006;28(1-2):34-42.
22. Jusko WJ, Rose JQ. Monitoring prednisone and prednisolone. Ther Drug Monit. 1980;2(2):169-76.
23. Vogt M, Derendorf H, Kramer J, et al. Biowaiver monographs for immediate release solid oral dosage forms: prednisolone. J Pharm Sci. 2007;96(1):27-37.
24. Kawai R, Mathew D, Tanaka C, et al. Physiologically based pharmacokinetics of cyclosporine A: extension to tissue distribution kinetics in rats and scale-up to human. J Pharmacol Exp Ther. 1998;287(2):457-68.
25. Lensmeyer GL, Wiebe DA, Carlson IH, et al. Concentrations of cyclosporin A and its metabolites in human tissues postmortem. J Anal Toxicol. 1991;15(3):110-5.
26. McMillan MA. Clinical pharmacokinetics of cyclosporin. Pharmacol Ther. 1989;42(1):135-56.