Changes in renal oxygenation and function secondary to cyclosporine A: BOLD MRI

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INTRODUCTION

Despite advances in solid organ transplantation, recent studies have shown that the use of cyclosporine A (CsA) and other calcineurin inhibitors in solid organ transplant patients could be a primary cause of end stage renal disease (ESRD), even in non-kidney transplants [1]. Therefore, it is important to identify therapeutic strategies to prevent CsA nephrotoxicity. We have demonstrated that Nox2, an isoform of NADPH oxidase, plays a vital role in CsA induced nephrotoxicity [2]. The goal of this experiment was to study the effects of CsA on renal function, in mice with and without the Nox2 gene, using blood oxygen level dependent (BOLD) MRI [3].

MATERIALS AND METHODS

Institutional Animal Care and Use Committee approval was obtained. Wildtype (WT; n = 12) and Nox2 knockout C57BL/6(KO; n = 13) mice were obtained from (The Jackson Laboratory, Bar Harbor, Maine, USA) and separated into four different treatment groups (Table 1). Mice were maintained for one month in an aseptic room with a low salt diet and sterilized water. Mice were injected with either CsA (30mg/kg) or vehicle (cremophor oil (390mg/kg)) every weekday for one month. Cremophor oil is an inert substance used in the control group. Mice were anesthetized with 3% isoflurane, followed by an intraperitoneal injection of 80uL of a 3:1 mixture of ketamine hydrochloride (100mg/mL) and xylazine hydrochloride (20mg/mL) prior to imaging. Scans were performed on a 3.0T MR scanner (GE Healthcare, Milwaukee, WI, USA) with a specially designed mouse coil. BOLD MR images were obtained with the following parameters: TR/TE/flip/BW = 100ms/6.6-41.4ms /30°/±31.25kHz, FOV = 6.0 cm, and 256 x 256 matrix. After imaging, mice were sacrificed and serum samples harvested for BUN analysis. BOLD images were analyzed as previously reported by Sadowski et al [4].

RESULTS AND DISCUSSION

We observed a trend towards increased cortical and outer medullary R2* (decreased oxygen bioavailability) and increased BUN levels (decreased kidney function) in the WT + CsA group compared to WT mice that did not receive CsA and to Nox2 KO mice with or without CsA (Figure 1 and Figure 2 respectively). The differences were not statistically significant, likely due to the small number of mice in each group. This study is ongoing in order to increase the number of mice in each study group. WT treated CsA mice demonstrated changes in renal oxygenation and function possibly due to the administration of CsA. These changes were not seen if CsA was not administrated or if Nox2 was not present (Nox2 KO mice).

CONCLUSIONS

This study demonstrates changes in renal oxygenation and renal function in mice receiving CsA. Those mice without the Nox2 gene (Nox2 KO mice) did not demonstrate changes in renal oxygenation or function. This suggests that in the absence of the Nox2 gene, there are no changes in renal oxygenation or function due CsA. Further studies are needed to confirm these finding, as Nox2 therapeutic targets could potentially ameliorate the effects of CsA on kidney function.

REFERENCES

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Mouse gentoype	*Treatment*	
	Vehicle	**CsA**
Wildtype	WT + vehicle (n= 6)	WT + CsA (n= 6)
Nox2 KO	KO + vehicle(n= 8)	KO + CsA (n= 5)

Table 1. Table representing four treatment groups with numbers of mice used (n) in each group. WT = Wildtype; KO = Knockout; CsA = cyclosporine

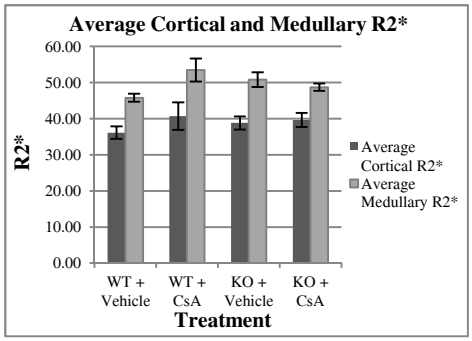

Figure 1. Average cortical and medullary R2* measurements for wildtype (WT) and knockout mice (KO) mice treated with and without cyclosporine.

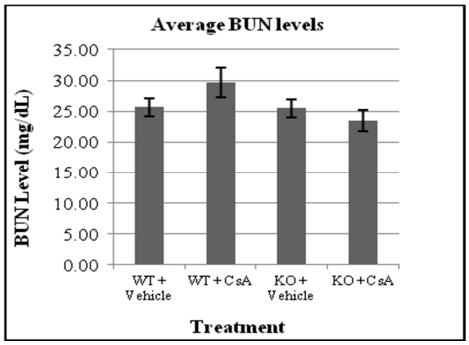

Figure 2. Average BUN levels for wildtype (WT) and knockout mice (KO) mice treated with and without cyclosporine.