EFFECT OF SILDENAFIL CITRATE IN ISCHEMIC ACUTE KIDNEY INJURY IS DAMAGE EXTENSION DEPENDENT

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Introduction and Objectives

Many pathophysiologycal processes have been implicated in the renal ischemia-reperfusion injury, including generation of reactive oxygen species (ROS) and reactive nitrogen species (RNS) simultaneously with the induction of protection mechanisms such as the expression of heme oxygenase-1 (HO-1) enzyme. HO-1 is a protectant against diverse insults in assorted tissues. Sildenafil citrate (SIL), a phosphodiesterase type-5 inhibitor, catalyzes the breakdown of cGMP, one of the factors involved in smooth muscle relaxation.

This study evaluated the effect of SIL and its association with HO-1 enzyme in protecting kidney function in a time (damage) dependent ischemic acute kidney injury (AKI) animal model.

Methods

- Animals: adult, male, Wistar rats weighing 260-300 g were divided:
  - **SHAM** – control;
  - **Ischemia 30 min** - renal pedicles clamping for 30 min;
  - **Ischemia 30 + SIL** - SIL 0,25 mg/kg 60 min before 30 min renal ischemia;
  - **Ischemia 45 min** - renal pedicles clamping for 45 min;
  - **Ischemia 45 + SIL** - SIL 0,25 mg/kg 60 min before 45 min renal ischemia.
- Renal Function: serum creatinine, inulin clearance, sodium fractional excretion (FENa), renal blood flow (RBF).
- Oxidative Metabolites: urinary peroxides, thiobarbituric acid reactive substances (TBARS), nitric oxide (NO) and thiols in renal tissue.
- Quantitative PCR of HO-1 in kidney tissue.
- Renal Histological Analysis: fractional interstitial area (FIA) and tubuleinterstitial injury .
- Statistical Analysis: differences between groups were analyzed by one way analyses of variance ANOVA and Newman-Keuls multiple comparison test. Results are presented as mean±SEM and $p<0.05$ was considered statistically significant.

Results

Ischemic groups showed decreased renal function and oxidative stress. Preconditioning of SIL in animals submitted to 30 and 45 min renal ischemia ameliorated renal function and RBF (**Table 1**), as well as, oxidative metabolites generation (**Table 2**).

Table1: Renal Function

Groups	n	Serum creatinine (mg/dl)	Inuline clearance (ml/min 100g)	FENa (%)	RBF (ml/min)
SHAM	7	0,3±0,1	0,63±0,04	0,29±0,30	10,4±0,80
Ischemia 30	6	2,1±0,3[a]	0,32±0,02[a]	2,85±0,50	4,0±0,30[a]
Ischemia 30+SIL	6	0,8±0,2[abc]	0,50±0,04[ab]	0,68±0,11	7,5± 1,01[b]
Ischemia 45	5	2,9±0,2[a]	0,16±0,02[ab]	9,17±1,38[ab]	2,3±0,08[a]
Ischemia 45+SIL	5	2,4±0,2[a]	0,31±0,03[ac]	4,16±1,12[ac]	5,5±0,52[ac]

Data reported mean±SEM. [a]$p<0.05$ vs SHAM; [b]$p<0.05$ vs Ischemia 30; [c]$p<0.05$ vs Ischemia 45.

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Table 2: Oxidative Metabolites.

Groups	n	UP (nmol / g cr)	Thiol (nmol / mg protein)	Urinary TBARS (nmol / g cr)	Urinary NO (µmol / g cr)
SHAM	7	1,7±0,1	170,2±15,8	56,7±7,2	30,2±2,2
Ischemia 30	6	5,9±0,2[a]	117,1±10,1[a]	113,7±15,6[a]	76,6±6,6[a]
Ischemia 30+SIL	6	2,3±0,3[bc]	110,6±10,7[a]	47,5±2,1[b]	35,5±3,5[b]
Ischemia 45	5	9,7±1,8[ab]	129,3±16,6[a]	116,8±33,0[a]	76,2±14,5[a]
Ischemia 45+SIL	5	4,0±0,6[c]	68,0±11,6[ac]	44,9±7,2[bc]	46,9±5,3[c]

Data reported mean±SEM. [a]$p<0.05$ vs SHAM; [b]$p<0.05$ vs Ischemia 30; [c]$p<0.05$ vs Ischemia 45.

SIL treatment induced HO-1 expression in kidney tissue (**Figure 1**). Renal tubules in ischemic kidney showed loss of brush border, vacuolation of cells and acute tubular necrosis mostly in 45 min ischemia (**Figure 2A**). In addition, an increase in interstitial area was observed (**Figure 2B**).

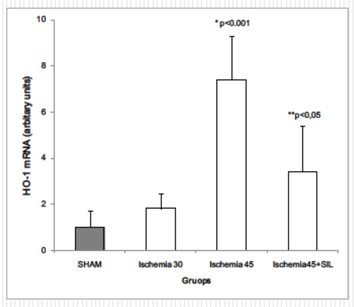


Figure 1: Quantitative PCR of HO-1 in kidney tissue.

Data reported mean±SEM. *$p<0.05$ vs SHAM; **$p<0.05$ vs Ischemia 45.

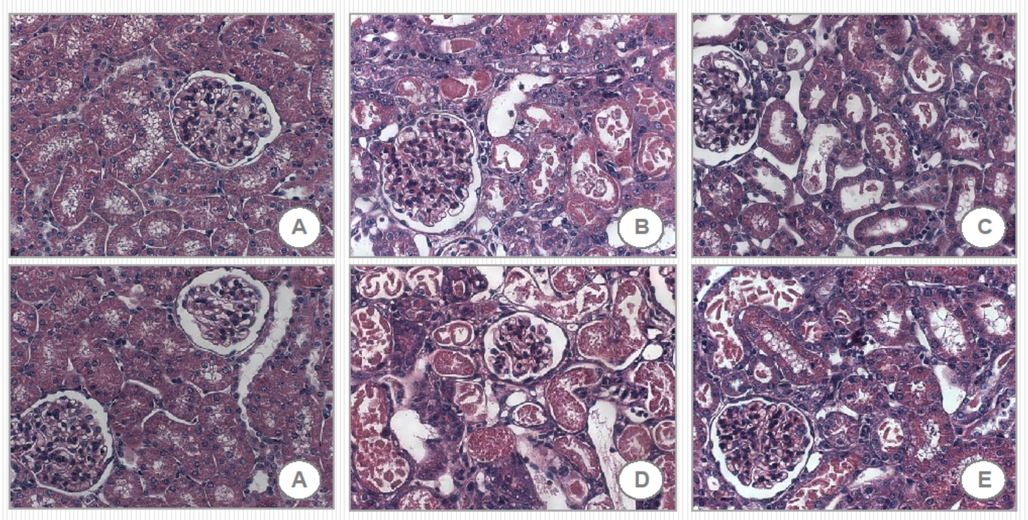
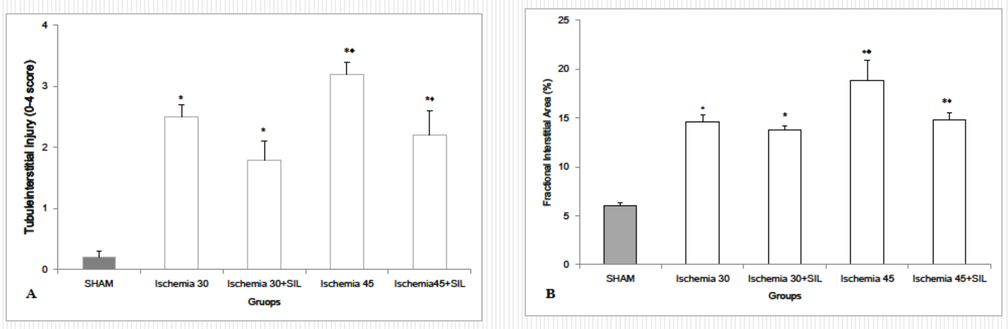


Figure 2: Renal histological analysis. (**A**) SHAM, (**B**) Ischemia 30, (**C**) Ischemia 30+SIL, (**D**) Ischemia 45, (**E**) Ischemia 45+SIL.



Data reported mean±SEM. *$p<0.05$ vs SHAM; ♠$p<0.05$ vs Ischemia 30; ♦$p<0.05$ vs Ischemia 45.

Conclusions

Study concludes that the functional and histological damage induced by time of ischemia determines SIL protective effect in AKI, which mechanisms involve HO-1 expression.

References

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