SOUTHERN ILLINOIS UNIVERSITY EDWARDSVILLE

Role of Dapagliflozin in Attenuating Renal Tubule Damage: An Immunohistochemical Analysis of a Diabetic Rat Model

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SCHOOL OF PHARMACY

BACKGROUND

- Diabetic nephropathy is a prevalent complication in both type 1 and type 2 diabetes, which contributes to the progression of end-stage renal disease
- Previous research indicates that hyperglycemia leads to renal fibrosis by recruiting various cytokines
- Extracellular matrix deposits are a key feature of renal fibrosis. In this process, interstitial myofibroblasts produce alpha-smooth muscle actin (ASMA)
- SGLT2-Is have shown benefit in clinical trials by decreasing the progression of end-stage renal disease

METHODS

Immunohistochemical Staining

- Five experimental groups: healthy control, diabetic control, insulin treatment, dapagliflozin treatment, and combination dapagliflozin and insulin treatment
- Phalloidin tagged with Alexa 488 (1:600) – used to identify renal structures including glomeruli and tubules
- Alpha actin tagged with Alexa 647 (1:300) – used to identify ASMA
- DAPI (1:300) – used to identify nuclei

Data Collection

- Confocal microscope with Olympus Fluoview program
- Oil immersion technique utilized with 20x magnification lens
- Fiji software was used to quantify area affected by ASMA
- Free-hand drawing tool used to outline portions of image with high intensity of ASMA

DISCUSSION

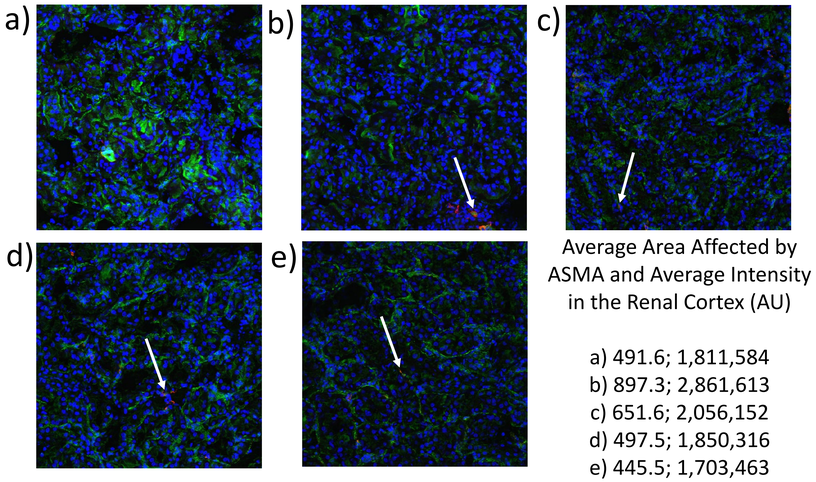
- Decreased oxygenation in the medulla with dapagliflozin treatment
- Increased glucose in the medulla with dapagliflozin treatment
- Alpha-smooth muscle actin non-specific
- Fiji software could lead to investigator error
- Discrepancies in image dimensions when analyzing
- Potential inadequate timeframe to see full effects of dapagliflozin treatment

CONCLUSION

- Dapagliflozin attenuates renal damage in the cortex, but not the medulla within this study
- Basic science research important for pre-clinical experiments as well as determination of new mechanisms not originally studied

RESULTS

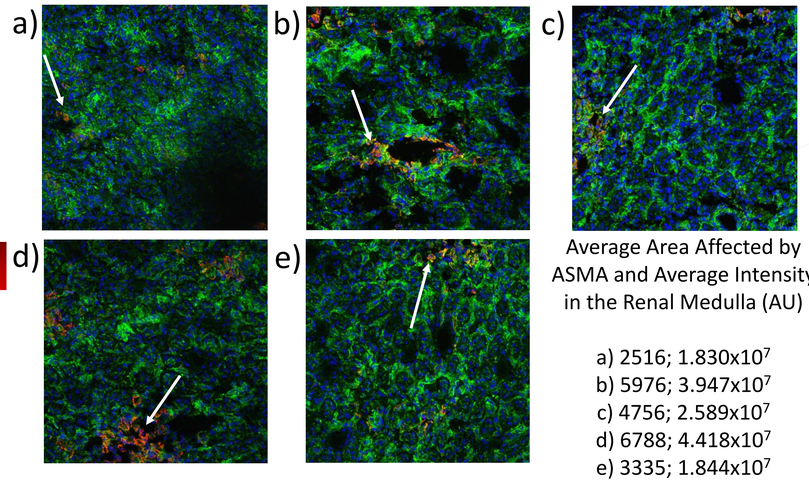
Renal Cortex



Average Area Affected by ASMA and Average Intensity in the Renal Cortex (AU)

a) 491.6; 1,811,584
b) 897.3; 2,861,613
c) 651.6; 2,056,152
d) 497.5; 1,850,316
e) 445.5; 1,703,463

Renal Medulla

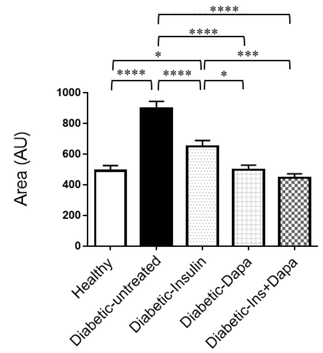


Average Area Affected by ASMA and Average Intensity in the Renal Medulla (AU)

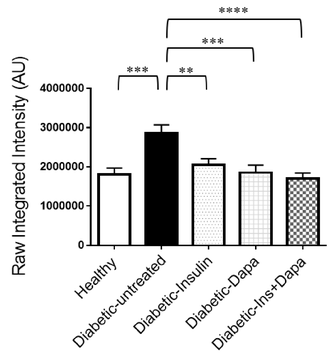
a) 2516; 1.830x10^7
b) 5976; 3.947x10^7
c) 4756; 2.589x10^7
d) 6788; 4.418x10^7
e) 3335; 1.844x10^7

a) Healthy Control; b) Diabetic Control; c) Diabetic-Insulin Treatment; d) Diabetic-Dapagliflozin Treatment; e) Diabetic-Combination Insulin and Dapagliflozin Treatment

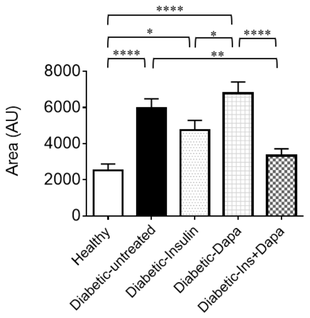
Cortex Area



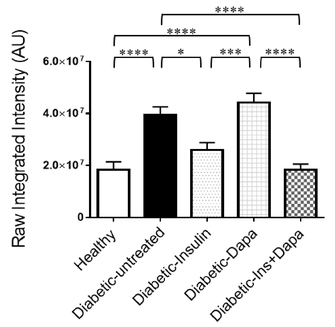
Cortex-Inte Density



Medulla Area



Medulla-Inte Density



P<0.05: *; P<0.01: **; P<0.001: ***; P<0.0001: ****

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