

## **Data Supplement**

### **Canagliflozin Prevents Intrarenal Angiotensinogen Augmentation and Mitigates Kidney Injury and Hypertension in Mouse Model of Type 2 Diabetes Mellitus**

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Fig. S1

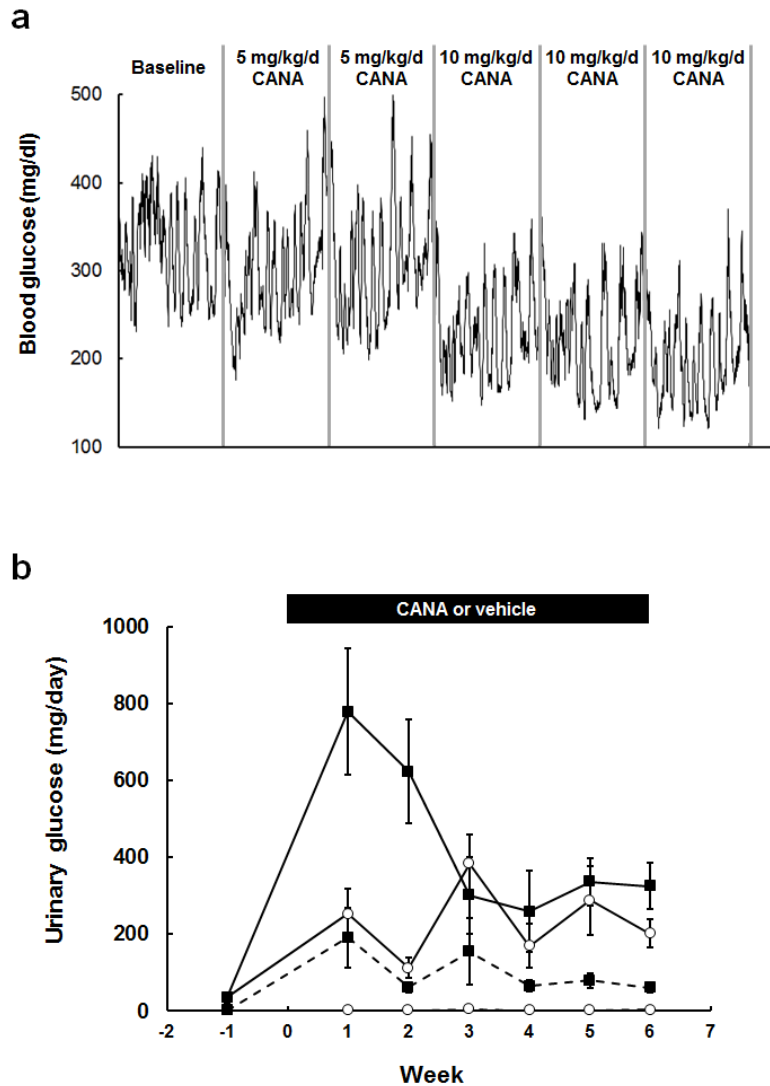


Fig. S1

**a.** Effects of low and high doses of CANA on hyperglycemia in a DM mouse. Blood glucose levels measured by a telemetry system were plotted every minute. The mouse was treated every day from 10:00 am - 10:30 am. After the baseline measurements, the mouse was treated with 5 mg/kg/day CANA for two days followed by 10 mg/kg/day CANA for three days.

**b.** Urinary glucose excretion in DM mice. ND+Veh: N=12, ND+CANA: N=11, DM+Veh: N=10-12, DM+CANA: N=14-16. Dotted lines show results in ND mice. Solid lines indicate results in DM mice. Open circles and closed squares are vehicle- and CANA-treated groups, respectively. Mean  $\pm$  SE. Asterisk ( $p < 0.05$ ) indicates significant difference among groups.

Fig. S2

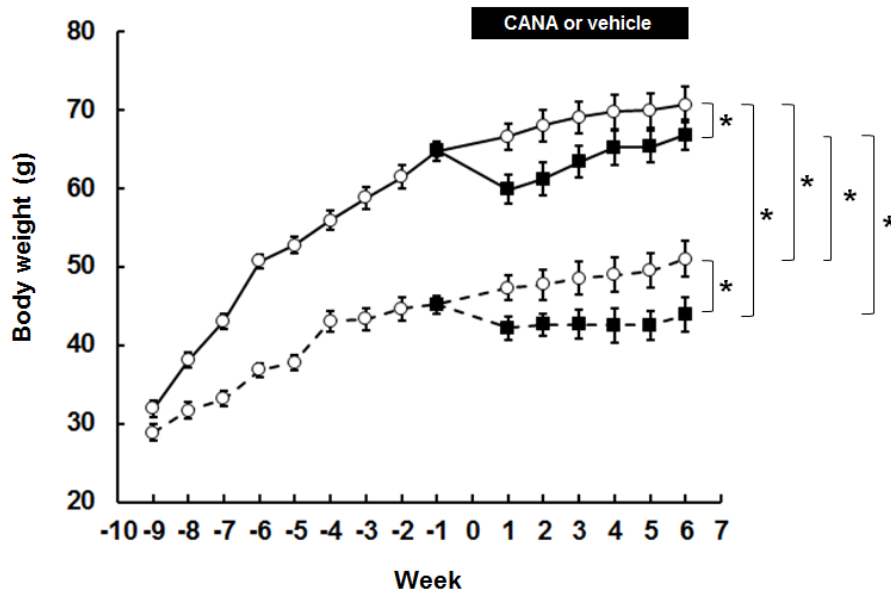


Fig. S2

Effects of CANA on body weight gain in T2DM mice. ND+Veh: N=9, ND+CANA: N=8, DM+Veh: N=7, DM+CANA: N=7. Mean  $\pm$  SE. Dotted lines show results in ND mice. Solid lines indicate results in DM mice. Open circles and closed squares are vehicle- and CANA-treated groups, respectively. Asterisk ( $p < 0.05$ ) indicates significant difference among groups.

Fig. S3

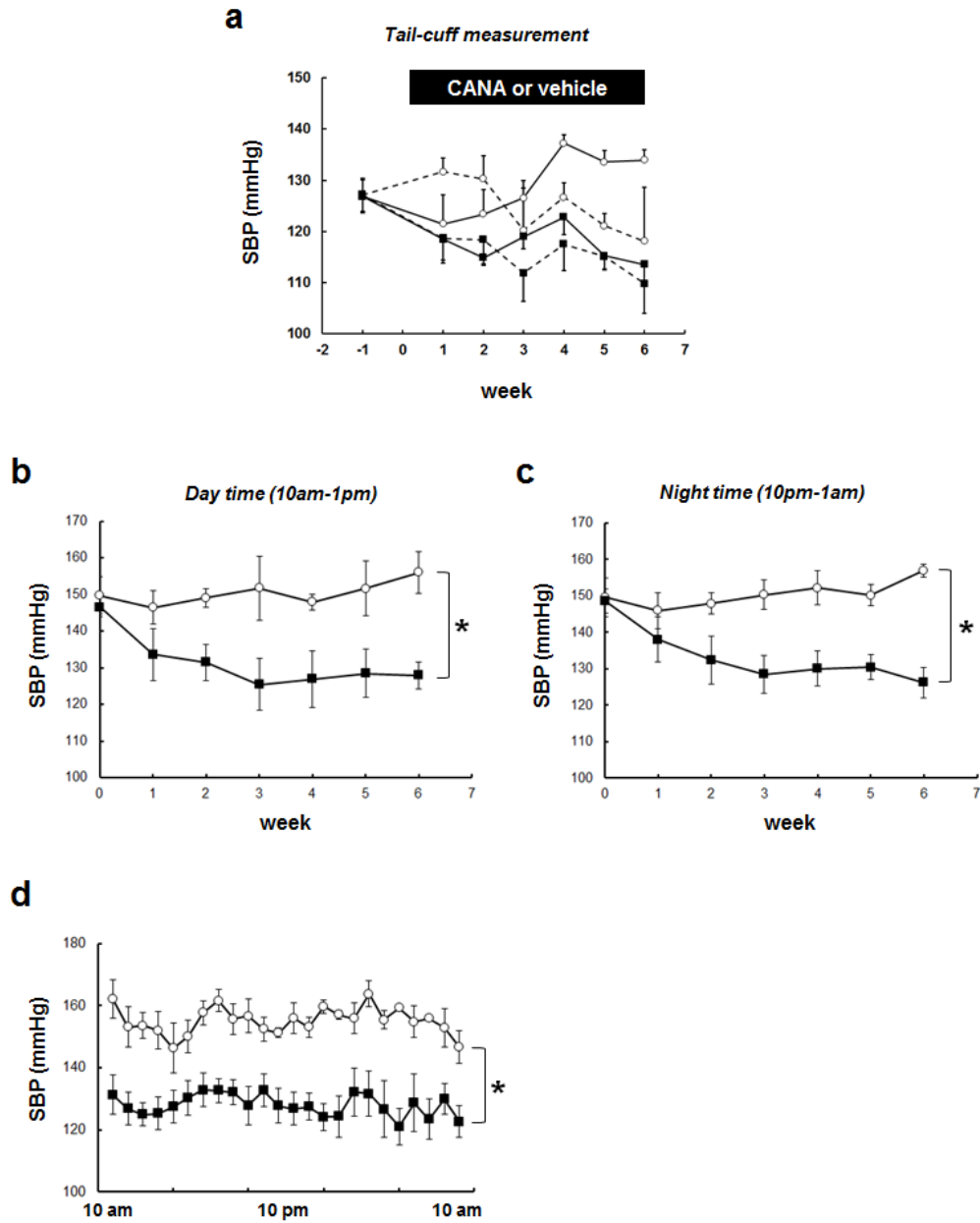
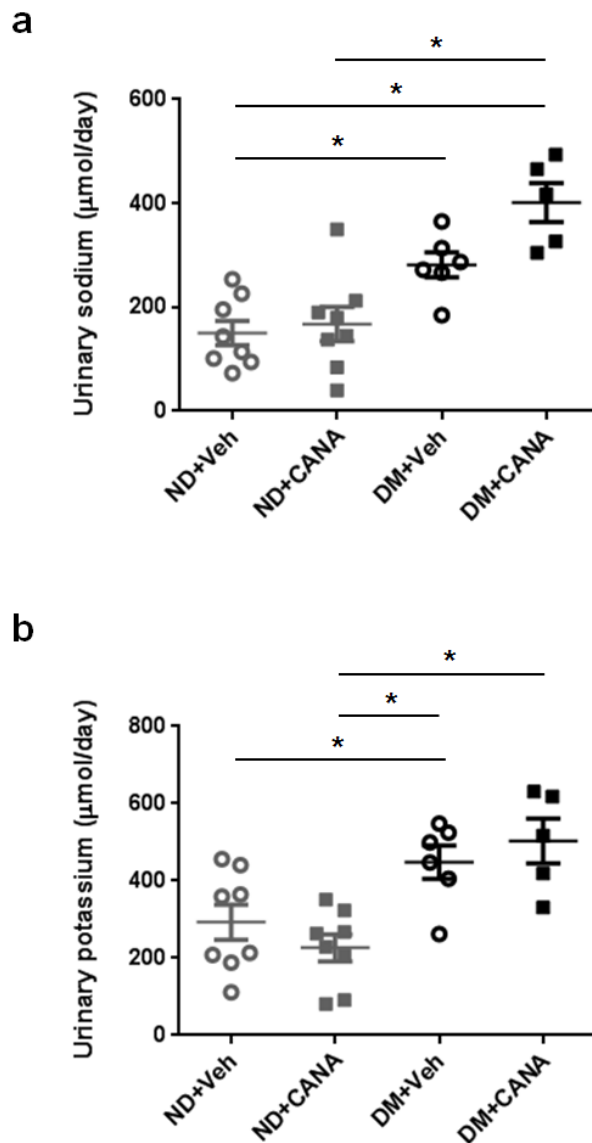


Fig. S3

**a.** Effect of CANA on SBP. SBP were measured by a tail-cuff system. ND+Veh: N=4-5, ND+CANA: N=5, DM+Veh: N=6-7, DM+CANA: N=5-7. Dotted lines show results in ND mice. Solid lines indicate results in DM mice. Open circles and closed squares are vehicle- and CANA-treated groups, respectively. Mean  $\pm$  SE. Panel **b** (day time) and **c** (night time) show changes in SBP monitored by a telemetry system during 6 week of treatment. Open circles and closed

squares are vehicle- and CANA-treated DM groups, respectively. Panel **d** indicates SBP during 24 hours on week 6. Mean  $\pm$  SE. N=4 in Vehicle group, N=5 in CANA group. Asterisk ( $p < 0.05$ )

**Fig. S4**



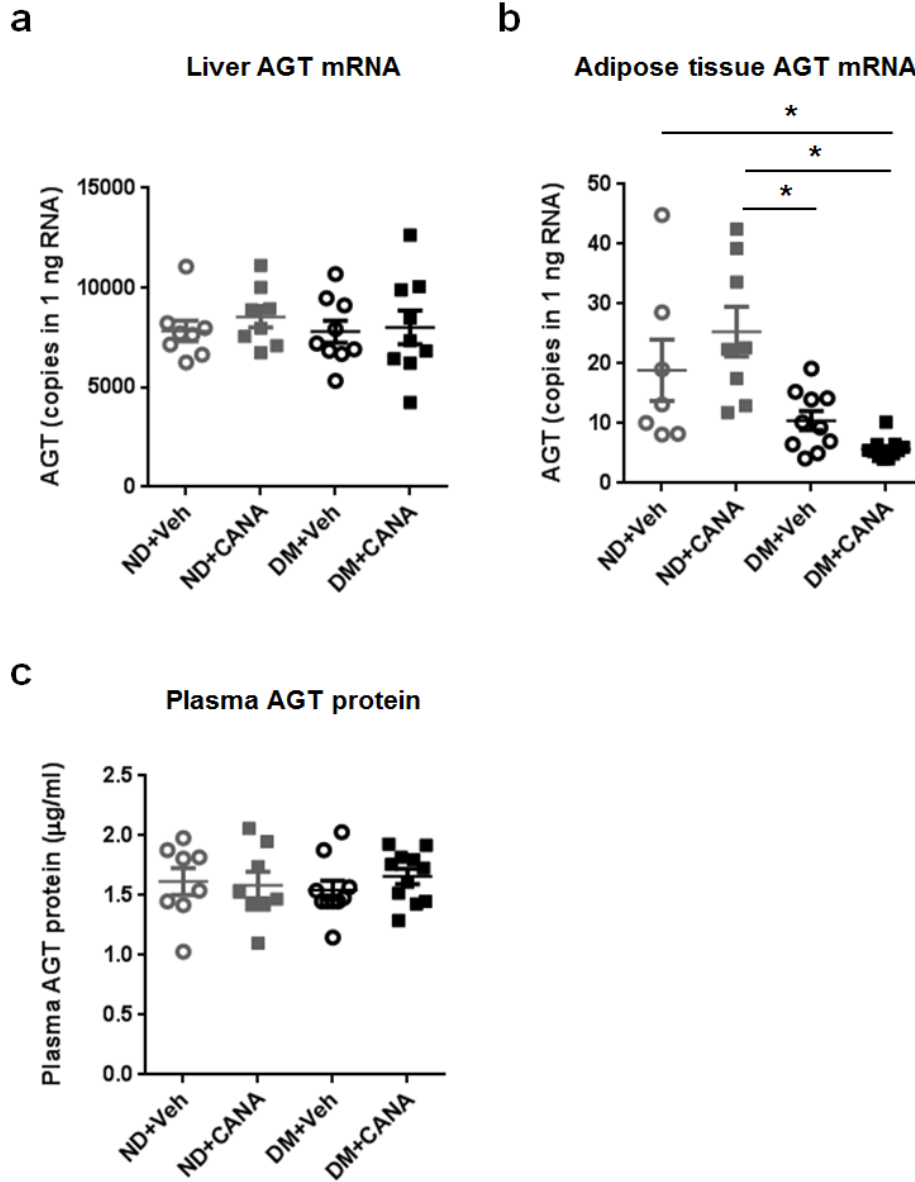
indicates significant difference between groups.

**Fig. S4**

Effects of CANA on urinary sodium and potassium excretion in T2DM mice. ND+Veh: N=8, ND+CANA: N=8, DM+Veh: N=6, DM+CANA: N=5. **a.** Changes in urinary  $\text{Na}^+$  levels on week 6.

**b.** Changes in urinary K<sup>+</sup> levels on week 6. Mean ± SE. Asterisk (p < 0.05) indicates significant difference among groups.

**Fig. S5**



**Fig. S5**

Effects of CANA on AGT levels in liver, adipose tissue and plasma in T2DM mice. ND+Veh: N=8, ND+CANA: N=8, DM+Veh: N=9, DM+CANA: N=10. AGT mRNA levels in liver (**a**) and adipose tissue (**b**). **c.** Plasma AGT levels. Mean ± SE. Asterisk (p < 0.05) indicates significant difference among groups.

Fig. S6

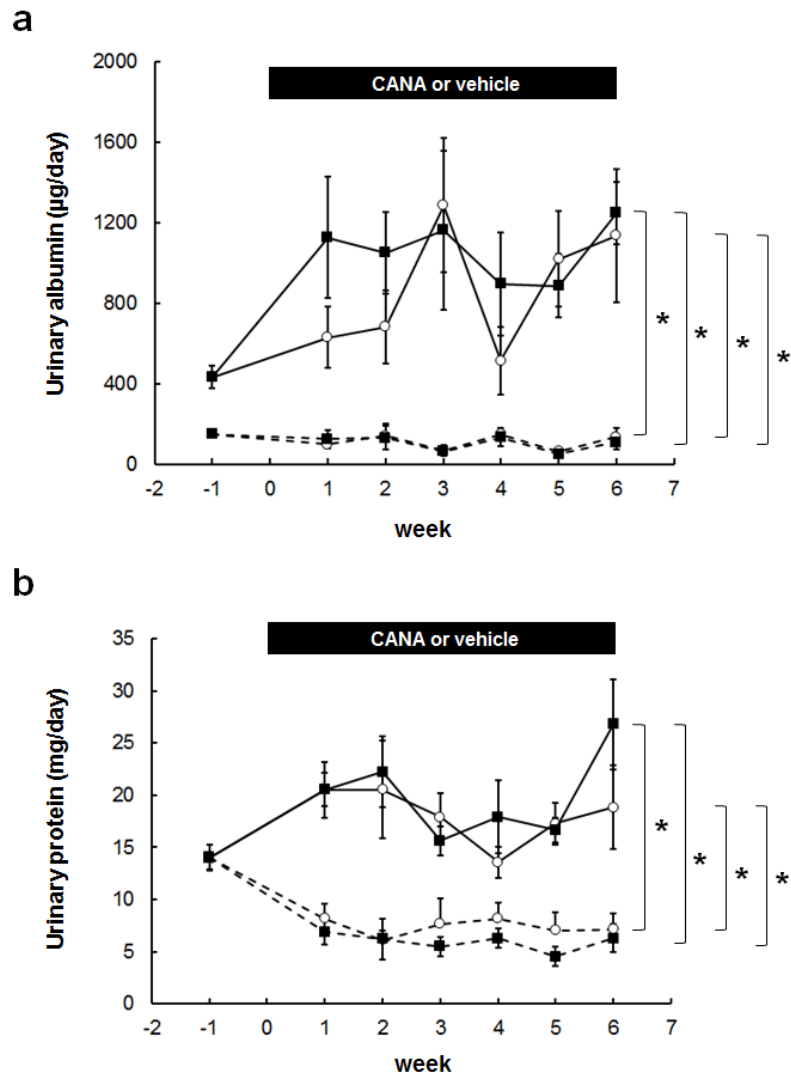


Fig. S6

**a. b. c. and d.** Urinary albumin and protein levels, respectively. Panels **a.** and **b.** show urinary albumin and protein levels normalized based on 24-hour urine volume. ND+Veh: N=4-8, ND+CANA: N=5-8, DM+Veh: N=5-7, DM+CANA: N=6. Dotted lines show results in ND mice. Solid lines indicate results in DM mice. Open circles and closed squares are vehicle- and CANA-treated groups, respectively. Mean  $\pm$  SE. Asterisk ( $p < 0.05$ ) indicates significant difference among groups.

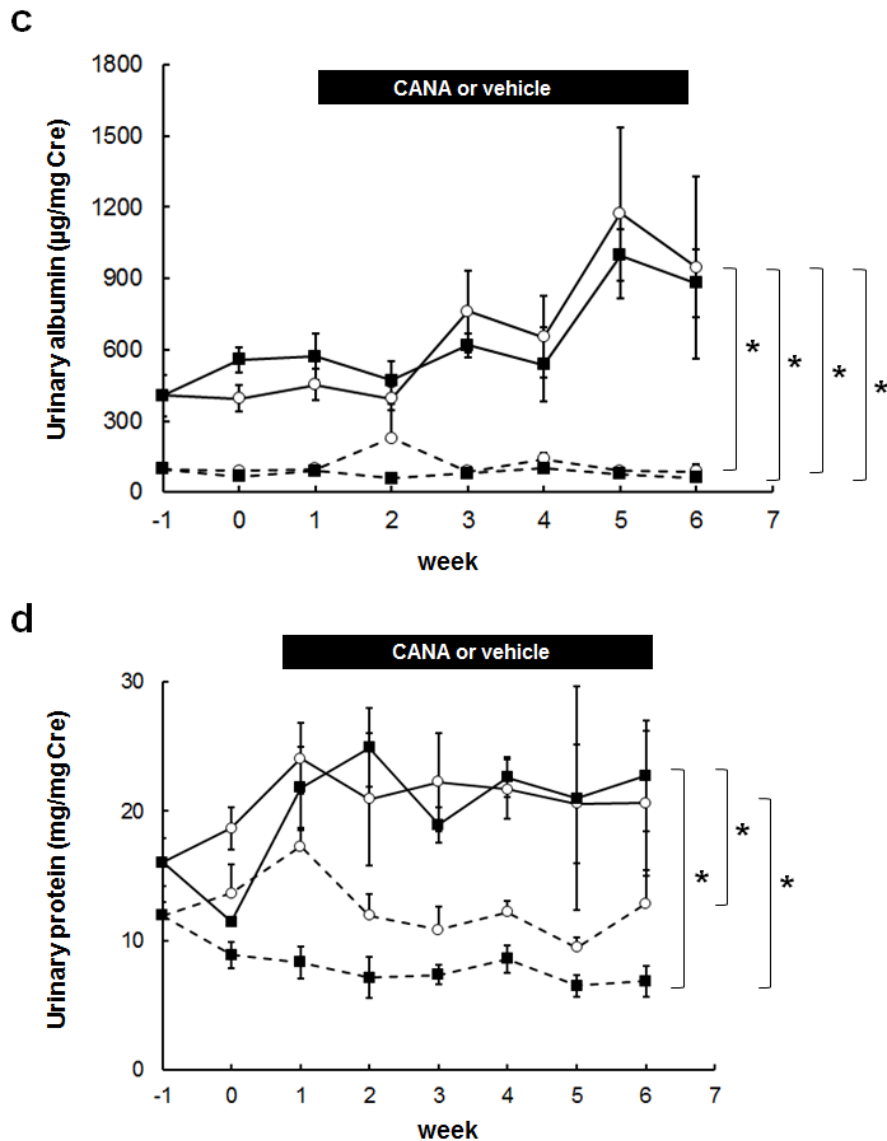


Fig. S6

**a. b. c. and d.** Urinary albumin and protein levels, respectively. Panels **c.** and **d.** show urinary albumin and protein levels normalized based on urinary creatinine concentrations. ND+Veh: N=4-8, ND+CANA: N=5-8, DM+Veh: N=5-7, DM+CANA: N=6. Dotted lines show results in ND mice. Solid lines indicate results in DM mice. Open circles and closed squares are vehicle- and CANA-treated groups, respectively. Mean  $\pm$  SE. Asterisk ( $p < 0.05$ ) indicates significant difference among groups.



**Table S1**

Gene copies in renal cortex (copies in 1 ng RNA)

	<b>ND+Veh</b>	<b>ND+CANA</b>	<b>DM+Veh</b>	<b>DM+CANA</b>
<b>N</b>	8	8	9	9
<b>Renin</b>	258 ± 41	371 ± 33	430 ± 55	621 ± 31*
<b>ACE</b>	313 ± 27	274 ± 36	261 ± 22	309 ± 22
<b>AT1a</b>	869 ± 37	927 ± 22	803 ± 57	825 ± 38
<b>SGLT2</b>	5314 ± 291	5079 ± 116	6472 ± 260*	5762 ± 342

\*: p&lt;0.05 vs ND+Veh

Table S1

Number of renin, ACE, AT1a and SGLT2 mRNA copies in renal cortex. ND+Veh: N=8, ND+CANA: N=8, DM+Veh: N=9, DM+CANA: N=9. Mean ± SE. Asterisk (p < 0.05) indicates significant difference compared to ND+Veh group.