Supplemental Data

Mass Spectrometry Imaging Reveals Elevated Glomerular ATP/AMP in Diabetes/Obesity and Identifies Sphingomyelin as a Possible Mediator

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Supplemental Figure Legends

Figure S1
ATP and ADP measurement in the renal cortex of WT and Akita mice
(A and B) ATP and ADP levels in the renal cortex of WT and Akita mice. (C) ATP/ADP ratio in the renal cortex of WT and Akita mice. $n = 3$ per group. $^{*}P < 0.05$. Values are the means ± *SE*. RLU; relative light unit.

Figure S2
Representative MALDI-MSI images of analytes distributed along with structures of mouse kidney
(A) Representative WT kidney section coated with 2,5-Dihydroxybenzoic acid (DHB) matrix. Scale bar; 1mm. (B) Representative H&E-stained WT kidney section. Scale bar; 1mm. (C) Representative overall average spectrum of WT kidney obtained by MSI analysis. Blue triangles show selected peaks (analytes) that have uniform distribution in the kidney section. Red triangles show selected peaks that are mainly localized in the renal cortex. Green triangles show selected peaks that are mainly localized in the renal medulla. (D) Representative MALDI-MSI images of analytes that are uniformly distributed in the kidney sections. The numbers on the picture indicate mass-to-charge (*m/z*) values. (E) Representative MALDI-MSI images of analytes that are mainly distributed in the renal cortex. (F) Representative MALDI-MSI images of analytes that are mainly distributed in the renal medulla. (G-J) MALDI-MSI images of analytes distributed in; (G) corticomedullary junction side in the renal medulla (*m/z* 117.8), (H) renal pelvis (*m/z* 279.9), (I) corticomedullary junction (*m/z* 741.5) and (J) renal cortex (*m/z* 756.7). (K) Overlay MALDI-MSI image of G to J. Spatial resolution for MALDI-MSI images; 90 μm.

Figure S3
Morphometric changes and urinary albumin/creatinin ratio in Akita and HFD-fed mice
(A and B) Representative images of PAS-stained kidney sections (400× magnification). Scale bars; 50 μm. (C and D) Glomerular hypertrophy and mesangial matrix accumulation was observed in the Akita group compared with the WT group. (E and F) After 1 week of HFD feeding exhibited no changes in glomerular size and mesangial matrix. Fifteen randomly selected glomeruli per mouse were examined ($n = 4$ per group). (G and H) Urinary albumin/creatinine ratio (UACR). The UACR of Akita group was markedly increased as compared with that of the WT group (G). The UACR was slightly increased in the HFD group but there was no statistical significance between the WT and HFD groups (H). $n = 6$ per group. Values are the means ± *SE*. *** $P < 0.001$ and $^{*}P < 0.05$.

Figure S4
Metabolic characteristic of 20-week aged STD and HFD mice
(A-C) Body weight (**A**), blood glucose (**B**) and relative kidney weight (**C**) at 20-week age. $n = 5$ per group. $^{***}P < 0.001$ and $^{**}P < 0.01$. Values are the means ± *SE*.

Figure S1, related to Figure 1

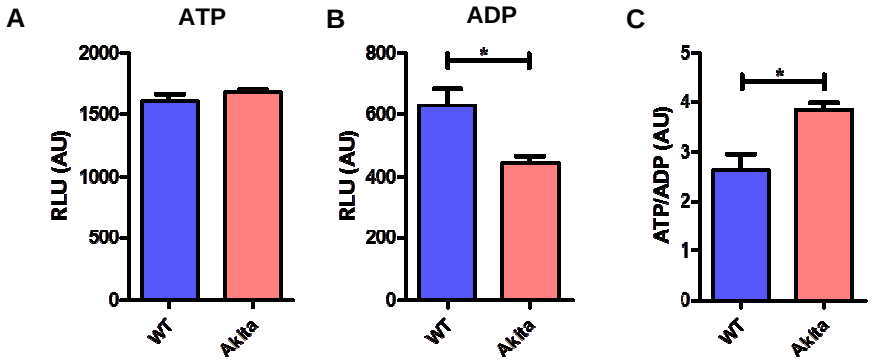

Figure S2, related to Figure 2

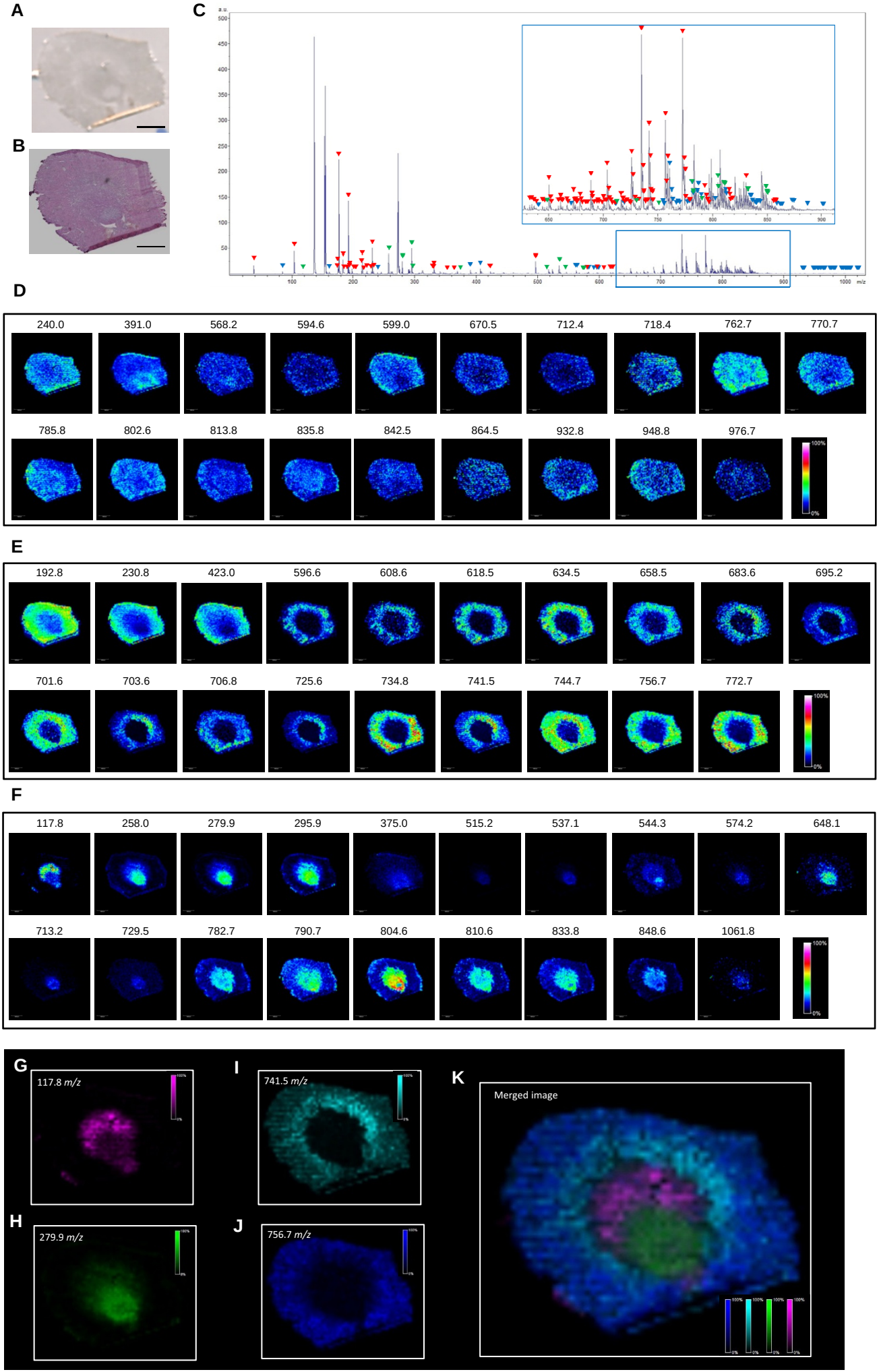

Figure S3, related to Figure 3 and 4

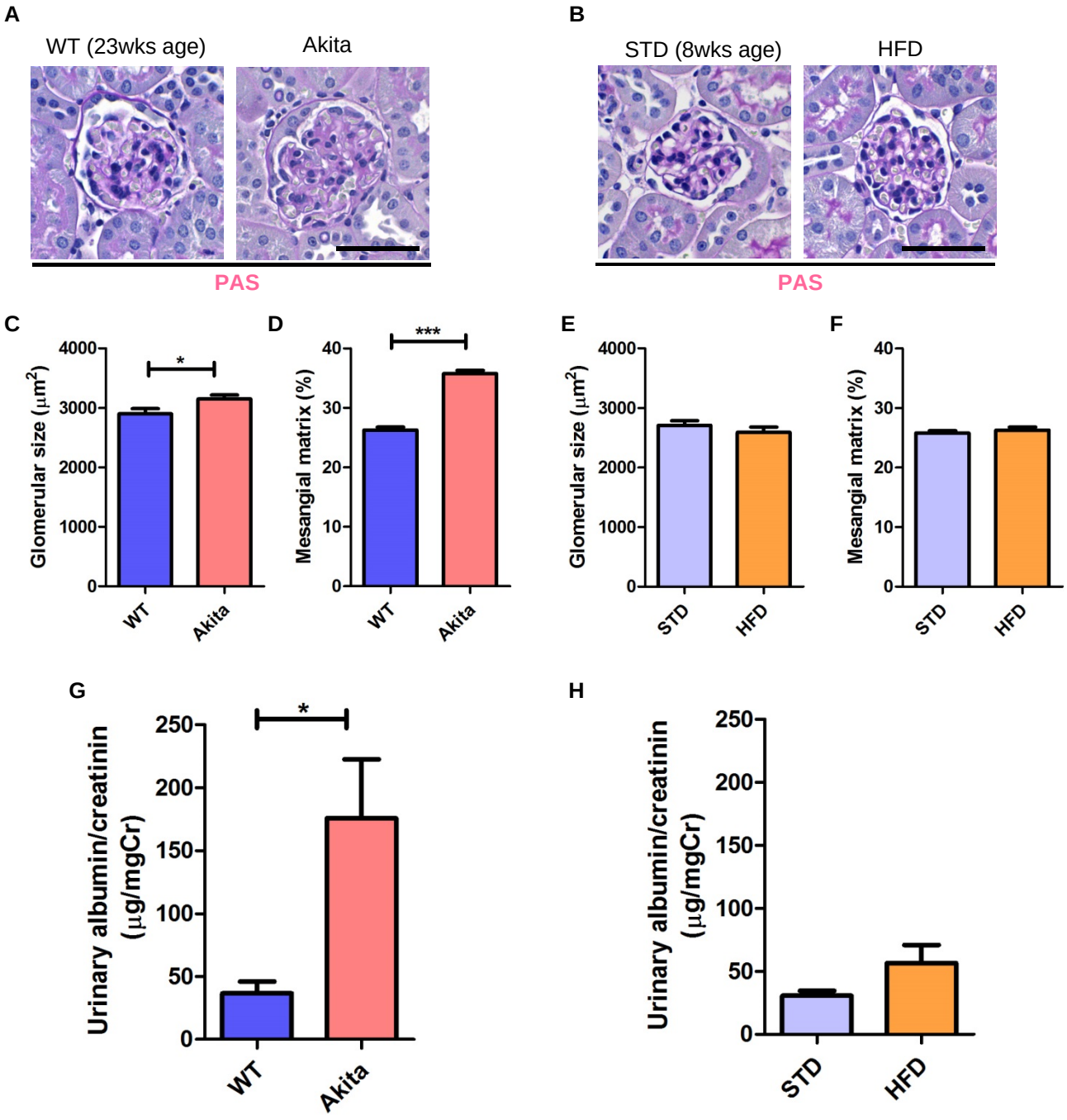

Figure S4, related to Figure 4