Supplementary Materials

Short-term changes induced by intermittent thermal dehydration, rehydration with 10% fructose and the effect of conivaptan.

Methods

Six groups of rats (n=4) were thermally dehydrated (37°C, 1 h/day) and rehydrated afterward with fructose 10% for 2 h. Three groups received conivaptan (C, 3mg/kg BW) and three received vehicle (water) by gastric gavage. A set of F+V and F+C groups were studied after 1, 3 and 5 episodes of intermittent thermal dehydration. At each point, a group of normal, non-dehydrated rats was included. At the end of each time point, urine was collected during 18 h in metabolic cages, after rehydration. At the end of the collection, rats were euthanized with isoflurane and blood collected from the abdominal aorta. Kidneys were resected, separated in cortex and medulla and stored in liquid nitrogen until further processing.

Plasma and urine creatinine, plasma copeptin, the urine marker of renal damage N-acetyl-β–glucosaminidase activity (NAG), lipid peroxidation and KHK expression in renal cortex was evaluated as described in Methods section in the main paper.

Results

We detected a slight but significant increment in plasma copeptin likely indicating that vasopressin response is activated during heat exposure (Fig S1).

Urinary excretion of NAG started to increase after 3 days, and renal cortex lipid peroxidation and KHK expression also had a significant increase after 5 and 3 days respectively of intermittent heat dehydration (Fig S2). Despite, there were no changes in CrCl during the studied time points (not shown). Blockade of V1a and V2 vasopressin receptors with conivaptan was able to prevent this early changes. These findings suggest that even a few episodes of mild dehydration induced by heat and rehydration with fructose initiate the activation of pathways that damage kidney.

Thus, these studies are consistent with the observation that renal injury is slow and likely cumulative, and provides supportive data for our study.

Supplementary Figure

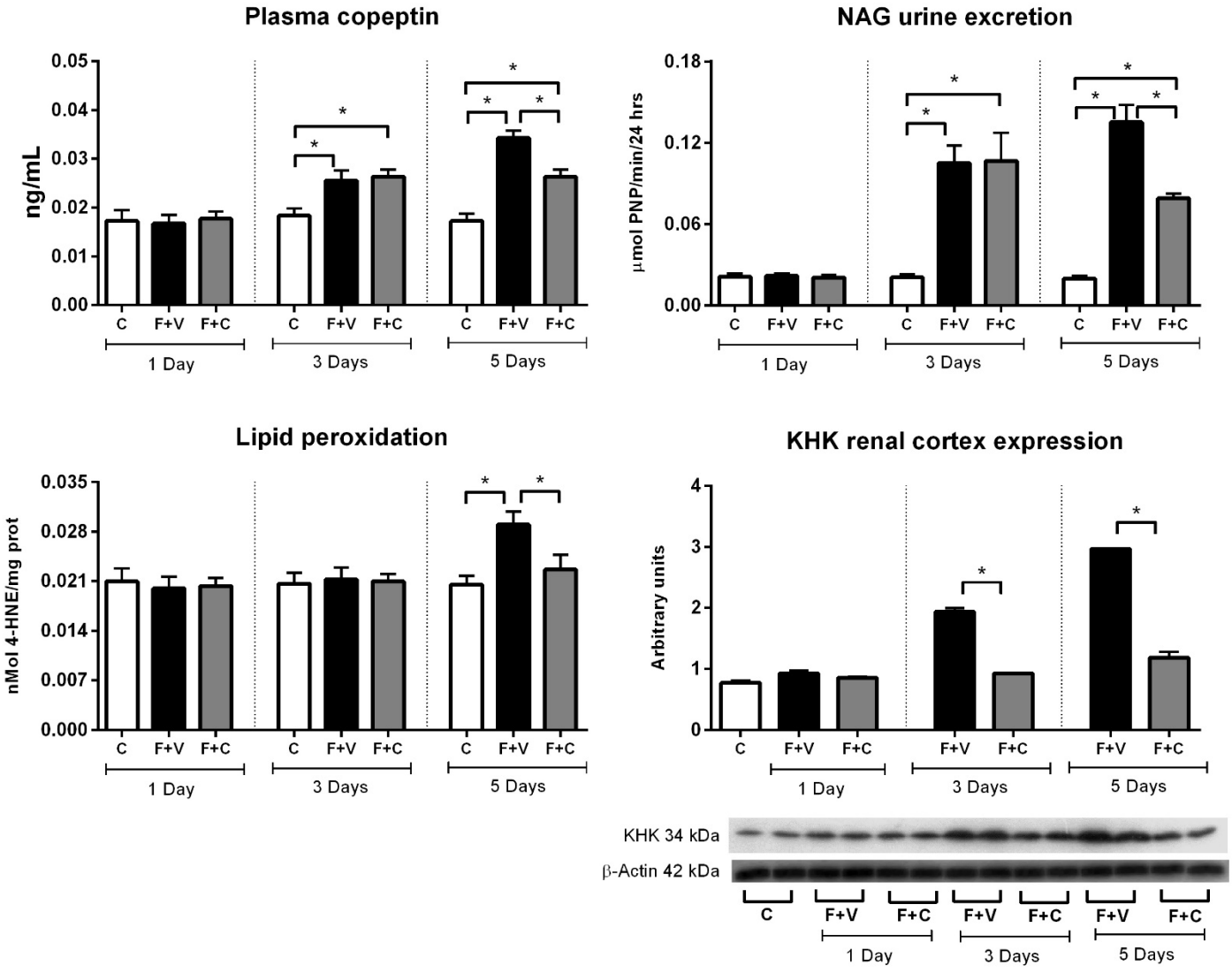

Figure S1. Short-term changes induced by intermittent thermal dehydration and rehydration with 10% fructose. Plasma copeptin concentration, NAG urine excretion as well as renal cortex lipid peroxidation and KHK expression after 1, 3 and 5 episodes of heat dehydration and rehydration with 10% fructose for 2 h