

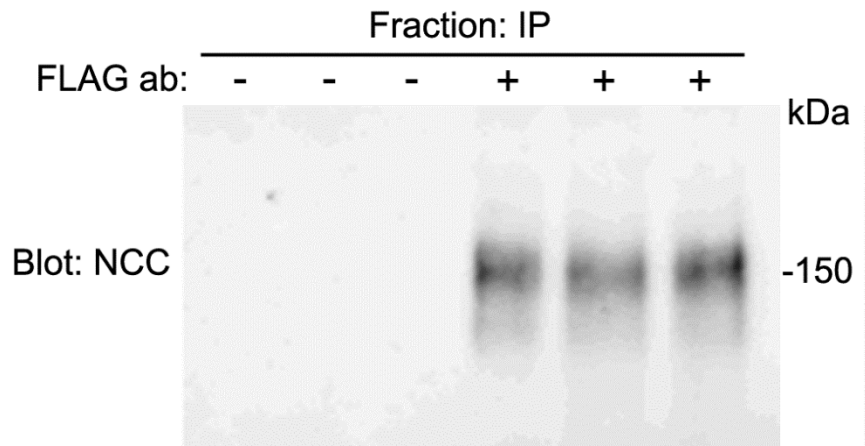
Supplementary Information

The thiazide sensitive sodium chloride co-transporter NCC is modulated by site-specific ubiquitylation

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Supplemental Figure 1. Specificity of the NCC immunoprecipitation (IP) using an anti-FLAG antibody. IP was performed routinely using antibody-deficient samples as negative controls. In IP without antibody, no signal corresponding to NCC can be detected with an anti-NCC antibody. Each lane represents a different sample.



Supplemental Figure 2. Specificity of the antibody targeting phosphorylated Threonine-58 NCC (pT58). Various MDCKI-rNCC cell lines with arginine/aspartic acid mutations at T58 (each lane represents a different cell line) were probed with either total NCC or a pT58-NCC specific antibody⁹. Total NCC was observed in all cell lines and whole kidney (WK) but not non-transfected control cells (NT). In contrast, signal using the pT58-NCC antibody was only observed in WK samples. Actin was used as a loading control.

