Background and Aims: Sympathetic nervous system has a key role in the regulation of renal function. We previously showed that, in mouse, the β3-adrenoreceptor (β3-AR) localizes in most of the nephron segments and its selective agonism promotes a potent antidiuretic effect. Here, we evaluate the expression of β3-AR in kidney intercalated cells (ICs) which are associated with the regulation of acid-base homeostasis in distal segments of the kidney tubule. The aim of this study is to investigate whether the β3-AR might play a role in the acid-base homeostasis operated by the kidney.

Method: The β3-AR expression in mouse kidney ICs was investigated by confocal microscopy. Wild type (wt) and β3-AR knock-out (ko) mice were used to study the role of β3-AR in the renal acid-base homeostasis. Mouse urine were collected using metabolic cages and pH was measured. Immunofluorescence and Western blotting experiments were performed to evaluate the localization and the abundance of renal H+-ATPase in wt and β3-AR ko mice. Kidney cells expressing human β3-AR (M1-β3-AR) and endogenous H+-ATPase were used to investigate the possible effects of β3-AR stimulation on the activity/localization of H+-ATPase. Live imaging experiments using the pH-sensitive dye BCECF were carried out to determine intracellular pH and assess H+-ATPase activity. Immunofluorescence experiments were performed to reveal the effects of β3-AR activation on H+-ATPase localization.

Results: Co-localization study of β3-AR with either H+-ATPase or Cl⁻/HCO₃⁻ exchanger pendrin in mouse kidney showed that β3-AR is expressed in H⁺-secreting type A, in HCO₃⁻-secreting type B, and in non-A non-B ICs. The urine pH of β3-AR ko mice was significantly higher compared with wt mice. In line with these results, localization and expression analysis showed that renal H⁺-ATPase significantly decreased in β3-AR ko mice compared to wt mice, supporting the idea that H⁺ secretion is partially blunted in these animals. Of note, exposure of M1-β3-AR cells to a selective β3-AR agonist induced a 2.5 fold increase of H⁺-ATPase activity compared to resting cells and this effect was prevented by a selective β3-AR antagonist or by the H⁺-ATPase inhibitor bafilomycin. Moreover, β3-AR agonism enhanced H⁺-ATPase apical expression in M1-β3-AR cells. In addition, the PKA inhibitor H89 abolished the stimulatory effect of β3-AR agonism, demonstrating the involvement of the cAMP/PKA pathway.

Conclusion: The present data suggest that modulation of H⁺-ATPase activity in renal ICs by endogenous β3-AR agonists may play an additional physiological role in hormonal control of renal acid-base homeostasis.