Metabolic acidosis is common in patients with chronic kidney disease (CKD) and may contribute to progression of CKD and all-cause mortality [1]. However, little is known about how CKD changes the response to an acute acid load and whether an altered response could contribute to adverse outcomes [2]. Therefore, our aim was to characterize the differences between the response to an acute oral acid load (to mimic the dietary acid load) in patients with CKD and healthy subjects.

To do so, we performed the short acid-loading test with ammonium chloride in 9 males with CKD Stage G4 and in 16 healthy male subjects (see Supplementary data for complete methods and baseline characteristics) [3]. The study was approved by the Medical Ethics Committee (MEC-2016-329) and registered at ClinicalTrials.gov (NCT03293446). In patients with CKD, all antihypertensive drugs (except β-blockers) were discontinued for 2 weeks to avoid drug interference. The test started after an overnight fast by giving a 10% oral solution of ammonium chloride (100 mg/kg body weight) over a period of 1 h together with a standardized meal (25 mmol sodium, 28 mmol potassium) and a water load (5 mL/kg followed by 2.5 mL/kg/h). The response to the acid load was observed for 6 h with repeated sampling of venous blood (at time 0, 3 and 6 h) and urine (hourly). Group comparisons were performed using repeated-measures two-way analysis of variance.

At baseline, blood and urine pH and plasma bicarbonate of the patients with CKD were significantly lower (Figure 1 and Supplementary data, Figure S1). After 3 h, ammonium chloride decreased blood pH and plasma bicarbonate and increased plasma potassium and plasma aldosterone similarly in both groups (Figure 1 and Supplementary data, Figure S1). At the end of the test, however, blood pH, plasma potassium and plasma aldosterone were returning to normal in the healthy subjects but not in the patients. Plasma renin did not change significantly during the test. The patients and healthy subjects adequately lowered urine pH (<5.3 in all participants). The healthy subjects and the patients also increased urine ammonium excretion after 1 h, but the increase in healthy subjects was significantly greater and persisted over time (Figure 1). This resulted in a significantly lower cumulative ammonium excretion in patients (Supplementary data, Figure S1). Accordingly, net acid excretion was also significantly lower in patients (no difference in titratable acid; Supplementary data, Figure S1). The same pattern as for urine ammonium excretion was also observed for urine sodium, chloride and potassium excretion. The urine excretion of creatinine, albumin and the low molecular weight proteins retinol-binding protein and renin also acutely increased after the acid load, with normalization thereafter (Figure 1 and Supplementary data, Figure S2). No differences between the groups were observed for the courses in creatinine and protein excretion, except for urinary renin. In patients with CKD, systolic blood pressure fell during the first 2 h and increased thereafter, whereas it remained stable in the healthy subjects (Supplementary data, Figure S2). Because patients were significantly older than healthy subjects, we also performed a subanalysis with older healthy subjects and found similar results (data not shown).

Here we characterized the response to an acute acid load on acid–base, electrolyte, creatinine and protein handling by the
kidney and addressed whether this response is altered in patients with CKD. We showed that urinary ammonium excretion is reduced in patients with CKD, increasing the duration of the acidosis. Of note, per-nephron ammonium excretion was likely higher in patients with CKD, although this was not sufficient to prevent the acidosis after acid loading. Persisting

**FIGURE 1:** Effects of an acute acid load with ammonium chloride on (A) venous blood pH and urine pH, (B) urine ammonium excretion, (C) plasma potassium ($K^+$), (D) plasma renin and aldosterone, (E) urine sodium excretion and cumulative excretion of sodium, chloride and potassium, (F) urine creatinine excretion and (G) urine renin excretion. Group comparison was performed using repeated measures two-way analysis of variance reporting the P-value for interaction. Cumulative excretions were compared with unpaired t-tests. Urine renin was not normally distributed and was therefore log-transformed for analysis.
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SUPPLEMENTARY DATA
Supplementary data are available at ndt online.

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CONFLICT OF INTEREST STATEMENT
None declared.

REFERENCES
1. Raphael KL. Metabolic acidosis in CKD: core curriculum 2019. Am J Kidney Dis 2019; 74: 263–275
2. Wesson DE, Bysse JM, Bushinsky DA. Mechanisms of metabolic acidosis-induced kidney injury in chronic kidney disease. J Am Soc Nephrol 2020; 31: 469–482
3. Wrong O, Davies HE. The excretion of acid in renal disease. Q J Med 1959; 28: 259–313
4. Palmer BF. Regulation of potassium homeostasis. Clin J Am Soc Nephrol 2015; 10: 1050–1060
5. Farooqui S, Sheriff S, Amlal H. Metabolic acidosis has dual effects on sodium handling by rat kidney. Am J Physiol Renal Physiol 2006; 291: F322–F331
6. Dubb J, Goldberg M, Agus ZS. Tubular effects of acute metabolic acidosis in the rat. J Lab Clin Med 1977; 90: 318–323
7. Gyurke ZS, Sulyok E, Guignard JP. Ammonium chloride metabolic acidosis and the activity of renin-angiotensin-aldosterone system in children. Eur J Pediatr 1991; 150: 547–549
8. Wagner CA. Effect of mineralocorticoids on acid-base balance. Nephron Physiol 2014; 128: 26–34
9. Tammaro G, Zacchia M, Zona E et al. Acute and chronic effects of metabolic acidosis on renal function and structure. J Nephrol 2018; 31: 551–559
10. Kovacs CP, Matsushita K, Sang Y et al. Serum potassium and adverse outcomes across the range of kidney function: a CKD Prognosis Consortium meta-analysis. Eur Heart J 2018; 39: 1535–1542
11. Remuzzi G, Cattaneo D, Perico N. The aggravating mechanisms of aldosterone on kidney fibrosis. J Am Soc Nephrol 2008; 19: 1459–1462
12. Abbate M, Zoja C, Remuzzi G. How does proteinuria cause progressive renal damage? J Am Soc Nephrol 2006; 17: 2974–2984
13. Rokouneir L, Heijmen BF, Nakano D et al. On the origin of urinary renin: a translational approach. Hypertension 2016; 67: 927–933

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