Renal Tubular Acidosis and Immune Checkpoint Inhibitor Therapy: An Immune-Related Adverse Event of PD-1 Inhibitor—A Report of 3 Cases

Sandra M. Herrmann, Mariam P. Alexander, Michael F. Romero, and Ladan Zand

The main cause of acute kidney injury in patients receiving immune checkpoint inhibitors (ICIs) is acute interstitial nephritis. However, as their use continues to increase, other kidney manifestations are being described. We report 3 cases of patients treated with ICIs who developed predominantly electrolyte disorders secondary to renal tubular acidosis as an immune-related adverse event and discuss the potential mechanism. Nongap acidosis in combination with hypokalemia should raise suspicion for distal renal tubular acidosis in patients treated with ICIs.

BACKGROUND

Immune checkpoint inhibitors (ICIs) have opened a new era in the treatment of advanced malignancies, resulting in remarkable clinical responses and improving survival in multiple types of cancers. The number of indications for the use of these agents has been increasing at an unprecedented rate. This has resulted in an increase in incidence of immune-related adverse events as a result of the immunologic effects of these therapies. The immune-related adverse events can affect essentially any organ but most commonly involve the gastrointestinal tract, skin, lungs, and endocrine system.[1] Kidney immune-related adverse events incidence is low at 2% to 5%.[2] Acute tubulointerstitial nephritis (AIN) is the most common kidney immune-related adverse event, but cases of glomerulonephritis have been described.[3] Renal tubular acidosis (RTA) has been reported, but is uncommon. In this report, we present 3 cases of distal RTA, all secondary to programmed cell death protein 1 (PD-1) blockade, and review the potential mechanism for the development of distal RTA.

CASE REPORTS

Case 1

A man in his early 60s was referred to the Nephrology Clinic for evaluation of elevated creatinine level and electrolyte level abnormalities, including hypokalemia and metabolic acidosis. His history was significant for lung adenocarcinoma and a brain oligometastatic lesion, for which he underwent gamma knife radiation. He was then treated with 4 cycles of carboplatin with an area under the curve of 5, pemetrexed at a 500-mg/m^2 dose, and pembrolizumab at a 200-mg dose, with plans for a left lower lobe lobectomy.

Before undergoing lung resection, serum creatinine level was normal at 0.9 mg/dL, with no evidence of electrolyte abnormalities. During the patient's hospital stay for lobectomy, creatinine levels varied between 1.1 and 1.2 mg/dL. He returned for follow-up 6 weeks after dismissal and was found to have prominent electrolyte level abnormalities (Table 1). The patient indicated that he was taking ibuprofen, 400 mg, daily as needed for pain, as well as omeprazole, 40 mg, daily (initiated a few weeks prior). He was placed on treatment with oral potassium chloride, 40 mEq, twice daily. He was advised to discontinue ibuprofen therapy, omeprazole was switched to ranitidine, and he was referred to the Nephrology Clinic.

At the time of nephrology consultation (3 months after his last dose of pembrolizumab), the laboratory workup showed persistent elevation of creatinine level (1.4 mg/dL) and persistent nongap metabolic acidosis (bicarbonate, 16 mmol/L) with hypokalemia (potassium, 3.2 mmol/L). Urine microscopy was bland, with urinary protein-creatinine ratio of 0.4 g/g and urinary retinal-binding protein to creatinine ratio of 75,652 µg/g (normal, <172 µg/g). Additional urine studies are reported in Table 1. Distal RTA was diagnosed and the patient with was started on treatment with oral sodium bicarbonate, 1,300 mg, twice daily and continued on potassium supplementation.

An ultrasound-guided kidney biopsy was performed, which showed chronic active tubulointerstitial nephritis with moderate arteriosclerosis (Fig 1). The patient was subsequently started on treatment with oral prednisone, 40 mg, daily, with a slow taper over a period of 3 months. The oral potassium supplementation dose was also decreased, but he continues to receive potassium chloride, 20 mEq, daily in addition to sodium bicarbonate, 650 mg, twice daily. Table 1 shows the laboratory workup 2 months after discontinuing steroid therapy (9 months from the last dose of pembrolizumab). At this time, he has not resumed ICI therapy.

Case 2

A man in his 70s with a history of hypertension, hypothyroidism, polymyalgia rheumatic receiving a long-term

low dose of prednisone, and metastatic melanoma was referred to the Nephrology Clinic for evaluation of elevated creatinine level and electrolyte level abnormalities. Metastatic melanoma with BRAF mutation (V600E) was diagnosed and he was placed on nivolumab (240 mg intravenously) therapy 16 months before his current evaluation.

A year later (while still on nivolumab therapy), the patient presented to his oncologist with confusion, fatigue, and poor appetite. His blood work revealed the following values: creatinine, 4.98 mg/dL; serum urea nitrogen 80 mg/dL; sodium, 144 mmol/L; potassium, 6.1 mmol/L; chloride, 107 mmol/L; and bicarbonate, 19 mmol/L. The previous week, his creatinine level was 1.27 mg/dL with no evidence of electrolyte abnormalities. The patient was hospitalized and received intravenous normal saline solution. Given the concern for possible AIN in the setting of nivolumab and proton pump inhibitor (PPI) use (which he was taking before initiation of ICI therapy), treatments with both drugs were discontinued. Follow-up blood work showed that creatinine level had improved to 1.85 mg/dL but metabolic acidosis had worsened (bicarbonate, 15 mmol/L). The patient was receiving oral prednisone, 7.5 mg, daily long term. His dose of prednisone was increased to 30 mg daily and he was referred to the Nephrology Clinic. Urine microscopy was bland, with urine protein/osmolality of 0.86. Additional urine studies and an electrolyte panel from his nephrology visit are outlined in Table 1. A diagnosis of distal RTA was made and the patient was started on treatment with potassium citrate, 30 mEq, twice daily.

An ultrasound-guided kidney biopsy was performed, which showed acute and chronic tubulointerstitial nephritis with mild arteriosclerosis (Fig 1). The PPI was switched to ranitidine and he was continued on prednisone therapy with a planned slow taper. His most recent blood work after 12 weeks of slow prednisone taper is shown in Table 1. For the past 6 months, the patient has not received any additional ICI therapy.

Case 3

An man in his early 80s with a history of hypertension and grade 3 renal cell carcinoma status post left radical nephrectomy (6 years prior) and pulmonary emboli on apixaban therapy was found to have metastatic renal cell carcinoma involving the right kidney and the small bowel mesentery, for which he was started on treatment with nivolumab (240 mg intravenously; 75,652 μg/g).

Before initiating nivolumab treatment, his creatinine level was 1.4 to 1.5 mg/dL. After initiating nivolumab treatment, creatinine level ranged between 1.6 and 1.9 mg/dL. A year later, he was found to have brain metastasis, which was treated with gamma knife radiation. The following year (2 years after initiating ICI treatment), the patient was started on omeprazole treatment. Two months after starting PPI therapy, creatinine level was

Table 1. Initial and Follow-up Laboratory Workup

	Case 1	Case 2	Case 3
Initial blood tests			
Sodium, mmol/L	137	144	142
Potassium, mmol/L	2.8	4.2	3.9
Bicarbonate, mmol/L	17	15	11
Chloride, mmol/L	107	118	117
Creatinine, mg/dL	1.55	1.57	2.13
BUN, mg/dL	27	30	44
Anion gap	13	11	14
Serum albumin, g/dL	4.4	4.8	3.9
ABG (pH, P_{CO_2}), mm Hg	(VBG) 7.26, 39	7.25, 22	7.23, 20
Initial urine studies			
Urine pH	6.5	6.3	6.7
Urinary potassium-creatinine ratio, mEq/g	260	60 (urine potassium)	NA
Urine anion gap	10	48	NA
Urine ammonium	8	23	NA
Follow-up blood tests			
Sodium, mmol/L	139	142	144
Potassium, mmol/L	3.9	3.7	4.3
Bicarbonate, mmol/L	18	23	23
Chloride, mmol/L	106	111	114
Creatinine, mg/dL	1.1	1.2	1.65
SUN, mg/dL	10	25	27
Anion gap	15	12	11

Note: Conversion factors for units: creatinine in mg/dL to μmol/L, ×88.4; SUN in mg/dL to mmol/L, ×0.357.
Abbreviations: ABG, arterial blood gas; NA, not available; BUN, blood urea nitrogen; VBG, venous blood gas .

noted to be further elevated (2.62 mg/dL), at which point he was seen in the Nephrology Clinic.

At that time, serum bicarbonate level was 16 mmol/L (anion gap of 9) and potassium level was 5.3 mmol/L. Urine microscopy was bland with protein/osmolality of 0.43. The elevated creatinine level was attributed to the combination of PPI and ICI. The PPI was switched to ranitidine and ICI treatment was put on hold. The metabolic acidosis at that time was attributed to decreased kidney function. A week later, repeat laboratory workup showed further elevation of creatinine level to 3.26 mg/dL and as a result, the patient was started on treatment with prednisone, 75 mg, daily empirically for presumed AIN. Kidney biopsy was not pursued in the setting of a solitary kidney and need for long-term anticoagulation therapy. The following week, blood work showed persistent acidosis (Table 1). Urinary retinal-binding protein to creatinine ratio was elevated at 79,130 μg/g (normal, <172 μg/g). The patient was initiated on treatment with sodium bicarbonate, 1,300 mg, twice daily. Three months after starting steroid therapy, prednisone dosage has been progressively tapered to 10 mg daily and laboratory workup shows improvement in creatinine and serum bicarbonate levels (Table 1). Nivolumab treatment remains on hold (last dose 4 months prior).
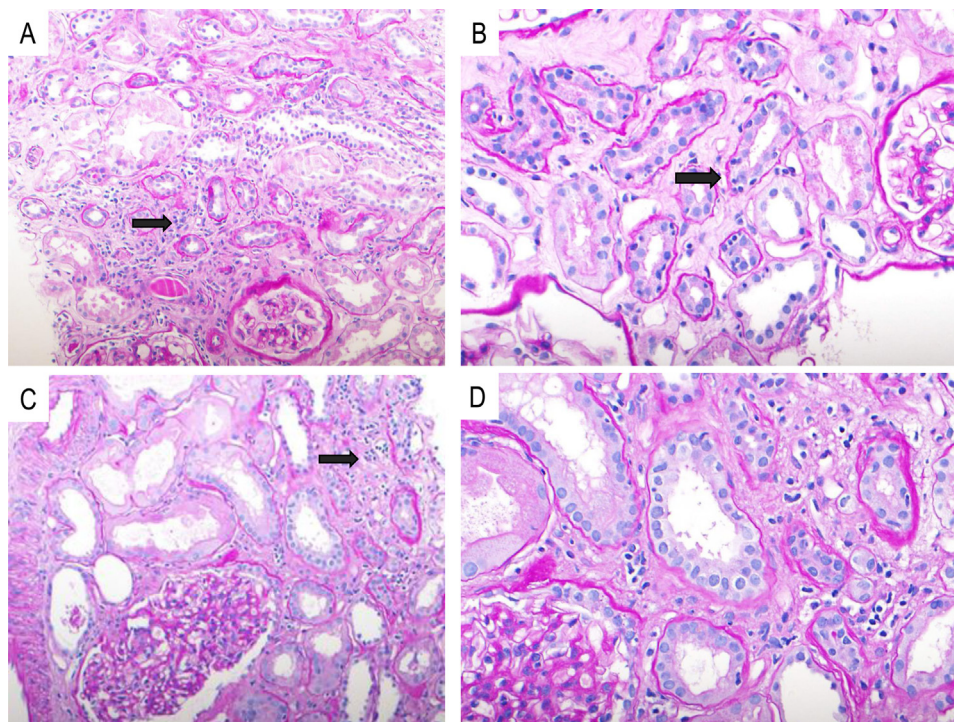
Figure 1. Periodic acid–Schiff (PAS) staining reveals interstitial inflammation and tubulitis. (A) Kidney biopsy representation of case 1 shows inflammation in the area of scarring (arrow) (PAS; original magnification, ×200). (B) Kidney biopsy representation of case 1 shows moderate tubulitis in partially atrophic tubules (black arrow) (PAS; original magnification, ×400). (C) Kidney biopsy representation of case 2 shows tubulitis and interstitial inflammation in areas of fibrosis (black arrow) (PAS; original magnification, ×200). (D): Kidney biopsy representation of case 2 shows tubulitis in nonatrophic and partially atrophic tubules (PAS; original magnification, ×400).

To investigate the potential mechanism for developing distal RTA in these patients, kidney biopsy frozen sections from patients 1 and 2 were further stained by indirect immunofluorescence for acid-base transporters in α-intercalated cells (α-ICs) including the α4 and B1 subunits of the vacuolar H^+-ATPase (vacuolar-type H^+-ATPase [V-ATPase]) and the anion-exchanger 1 (AE1; SLC4A1) as previously described.[4,5] Similar staining was also done on kidney tissue from a donor implantation needle biopsy as a control for comparison. Results are shown in Figure 2. To quantify the staining, data were normalized to the control (control being 100%). Because staining was not performed on the same day, they were each compared with the electrogenic sodium bicarbonate cotransporter 1 (NBCe1A) (green channel). For each set, the excitation, exposure, and adjustment of the channel brightness was constant for control, case 1, and case 2. Compared with the control kidney, staining for α4 and B1 subunits of the H^+-ATPase and AE1 was less prominent in patients 1 and 2 (Table 2). However, staining for cKit (another marker for the α-IC) was not reduced.

DISCUSSION

Kidney immune-related adverse events are increasingly associated with ICI therapy. Despite their low occurrence, as the use of ICI therapy increases, so does the incidence of nephrotoxicity. Acute tubulointerstitial nephritis is the most common nephrotoxicity associated with ICI therapy.[3] Distal RTA appears to be an emerging side effect that is not as well recognized and to date there are only 2 reported cases in the literature.[6,7] We report 3 additional cases of distal RTA secondary to the use of ICIs. We hypothesize that distal RTA occurrence is more common than realized and that mild cases may be missed because the metabolic acidosis may be attributed to the underlying decreased kidney function. This was true in our third patient. Acute kidney injury (AKI) commonly presents with metabolic acidosis and clinicians may not readily consider an alternative cause of acidosis when evaluating a patient with AKI. In patients 2 and 3, the clue was the worsening acidosis despite improvement in kidney function, the disproportionate degree of acidosis to kidney failure, and absence of a concomitant anion gap acidosis. The high urinary pH and low urinary ammonium level were consistent with impaired ammoniagenesis, as would be seen in distal RTA.

In our series, all 3 cases of distal RTA occurred in combination with AIN. This is similar to the other 2 reported cases in the literature.[6,7] In the first 2 patients, the diagnosis of AIN was biopsy proven. In the third patient, although a kidney biopsy was not pursued, the significant elevation in urinary retinal-binding protein to creatinine
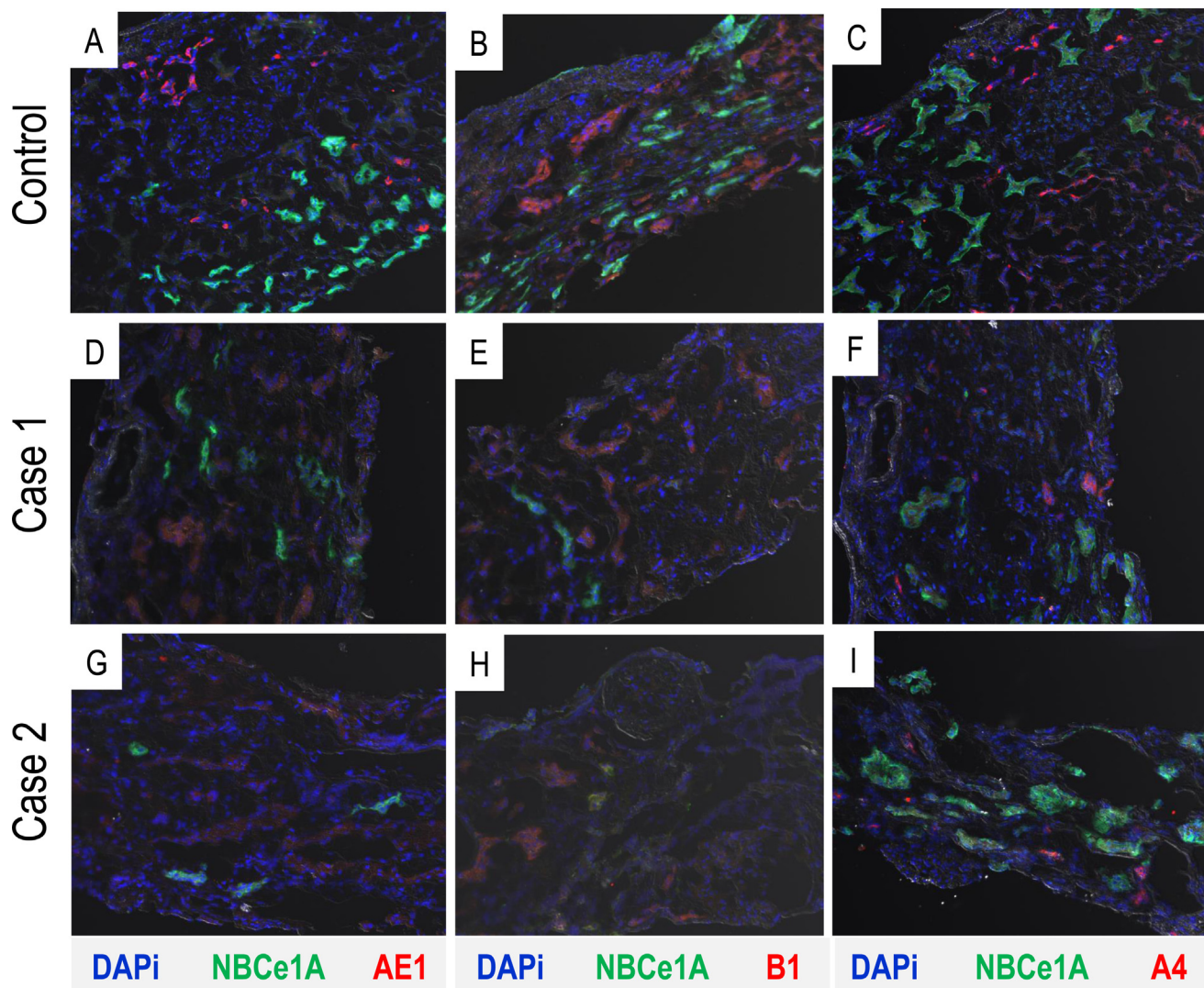
Figure 2. Immunofluorescent labeling with specific antibodies indicates cellular phenotypes. Tissues were stained with multiple antibodies to indicate nephron and cell phenotype. Loss of intercalated cells expressing anion exchanger type 1 (AE1) and B1 and α4 subunits of vacuolar-type H^+-ATPase (V-ATPase). (A-C) Healthy human kidney was stained for electrogenic sodium bicarbonate cotransporter 1 (NBCe1A; green, proximal tubule specific),[20] DAPI (4′,6-diamidino-2-phenylindole; blue, nuclei) along with AE1 (A, red) or B1 subunit of V-ATPase (B, red) or α4 subunit of V-ATPase (C, red). (D-F) Kidney tissue from case 1 was stained for the same markers (D, red) or B1 subunit of V-ATPase (E, red) or α4 subunit of V-ATPase (F, red). (G-I) Kidney tissue from case 2 was the same markers as control and patient 1. with AE1 (G, red) or B1 subunit of V-ATPase (H, red) or α4 subunit of V-ATPase (I, red).

ratio was highly suggestive of tubular injury, as could be seen with AIN. The clinical features of distal RTA coincided with an elevation in serum creatinine levels in cases 2 and 3, but features of distal RTA were present before the elevation in serum creatinine level in case 1. Thus, it is important that physicians caring for such patients are aware of distal RTA as a potential side effect associated with ICI therapy because it may be the first sign that the patient may develop AIN. In addition to the concomitant occurrence of AIN and distal RTA in these patients, all 3 cases were also receiving omeprazole in addition to the ICI. In cases 1 and 3, the AKI and distal RTA occurred shortly after starting PPI therapy. Recent studies have shown that PPI use increases the occurrence of AKI in patients who are

on ICI therapy.[3,8] Whether the addition of PPI to ICI results in further activation of the immune system or the occurrence of distal RTA in association with PPI was coincidental is at this point unclear.

The occurrence of distal RTA in combination with AIN raises the possibility that the underlying mechanism for distal RTA may be immune mediated. Distal RTA has been reported in various autoimmune disorders, including Sjögren syndrome, primary biliary cirrhosis, autoimmune hepatitis, systemic lupus erythematosus, and rheumatoid arthritis.[9,10] Thus to have distal RTA cases in association with ICI therapy is not surprising.

The inherited forms of distal RTA are due to mutations in either the anion exchanger (AE1; SLC4A1) or mutations

Table 2. α-Intercalated Cell Markers Are Decreased in Cases 1 and 2

Acid-Base Transporter	Control	Case 1	Case 2
AE1	+++++	++	++
NBCe1	+++++	+++	++
B1-V-ATPase	+++++	++	++
NBCe1	+++++	+	++
A4-V-ATPase	+++++	+++	+++
NBCe1	+++++	++	++++

Note: Date from Fig 2 relatively quantified from constant illumination and acquisition times. Scoring system: 0% to 20%, 0; 20% to 40%, +; 40% to 60%, ++; 60% to 80%, +++; 80% to 99%, ++++; and 100%, +++++ (control). Abbreviations: AE1, anion exchanger type 1; B1, A4, subunits of vacuolar-type H⁺-ATPase; NBCe1, electrogenic sodium bicarbonate cotransporter 1; V-ATPase, vacuolar-type H⁺-ATPase.

in the B1 subunit or α4 subunit of the V-ATPase in collecting duct α-IC.[11] Previous reports that have studied acquired distal RTA in the setting of autoimmunity have shown decreased expression of V-ATPase pump and AE in the renal biopsy specimen.[12,13] Similarly, autoantibodies isolated from patients with Sjögren syndrome can interact directly with V-ATPase pumps in the α-IC.[4] In patients 1 and 2, staining for the α4 or B1 subunit of V-ATPase pump and staining for the AE1 were all reduced compared with the control (Fig 2; Table 2), similar to patients with Sjögren syndrome, suggesting that the process may be immune mediated. Interestingly, we also observed a reduction in NBCe1A stain (Fig 2, green stain). It is difficult to determine the cause of this reduction in staining. One possibility is that this reduction may be due to kidney damage in the setting of AIN. The other possibility is that NBCe1A is downregulated due to decreased ammoniagenesis that can occur in the setting of distal RTA.[14]

The mechanism by which distal RTA develops in patients who are receiving ICIs is not yet clear. One potential mechanism may be nonspecific tubular injury secondary to the inflammatory cells. However, if this were to be the only mechanism causing distal RTA, we would have expected to see a higher incidence of distal RTA in patients who develop AIN secondary to ICI therapy or other causes of tubular injury. Given the decreased staining of the V-ATPase, we postulate that PD-1 inhibitors may exert their effect by modulating adenosine, which is one of the extracellular activators of type α-ICs.[15] Renal epithelial cells release adenosine triphosphate (ATP) and adenosine diphosphate (ADP) in response to stimuli. The nucleotides are in turn hydrolyzed to produce adenosine in the lumen through ectonucleotides that are located along the renal tubule.[16] The adenosine in turn activates the purinergic type 1 receptors (A2A and A2B) to induce V-ATPase–dependent H⁺ secretion partly by increasing V-ATPase accumulation in the apical membrane of the medullary intercalated cells.[15] Adenosine exerts this effect through the purinergic receptors in a cyclic adenosine monophosphate (cAMP)/protein kinase A–dependent pathway.[15]

There is growing evidence that tumor cells are efficient in converting ATP to adenosine and modulate the immune system through purinergic signaling. Consequently, ongoing preclinical and clinical trials are evaluating the role of purinergic receptor (A2A and A2B) antagonists in certain malignancies (NCT02403193). PD-1 inhibitors have been shown to synergize the inhibition of adenosine signaling through the purinergic receptor.[15] The combination of A2A receptor antagonist and PD-1 inhibitors is currently being studied (NCT03099161, NCT02655822). It is therefore tempting to hypothesize that the decreased V-ATPase expression that was noted in our patients could be mediated by the PD-1 inhibition of the purinergic receptors. This concept will need to be explored in the future.

It is noteworthy that the 3 cases in our series and the 2 previously reported cases of distal RTA were secondary to the use of either pembrolizumab or nivolumab. Both are PD-1 inhibitors rather than programmed death-ligand 1 (PD-L1) inhibitors. There is evidence that PD-1 inhibitors are more likely to cause immune-related adverse events compared with PD-L1 inhibitors.[17,18] This differential effect could be explained because blocking PD-1 would also prevent the interaction of PD-1 receptor with PD-L2, which in turn may further inhibit the immune system.[19] All 3 patients were treated with steroids. Two patients showed complete resolution of the acidosis and the other showed improvement in the acidosis after stopping the PD-1 inhibitor and treatment with steroids. The specific class effect in addition to distal RTA improvement with steroids are both consistent with the hypothesis that the development of distal RTA is immune mediated.

The importance of the immune system in the control and abatement of malignant cells is now appreciated. The use of immune checkpoint inhibition takes advantage of blocking our innate biological stops (checkpoints) as a strategy directed against cancer cells. However, this comes at a price. In this case report, we have presented 3 additional patients who developed distal RTA in combination with AIN after receiving PD-1 inhibitors.

Our case report has limitations. Although we propose that an immune-mediated process was the cause of distal RTA, we cannot rule out the possibility that nonspecific tubular injury from inflammatory cells may have played a role. Comparing staining of V-ATPase in α-IC in patients with AIN secondary to ICI treatment with and without distal RTA in the future would be useful. All patients were successfully treated with corticosteroid therapy with slow taper (due to a long drug half-life) as well as discontinuation of treatment with the PD-1 inhibitor and other nephrotoxic drugs. Given these findings, we recommend that all patients receiving PD-1 inhibitors should be monitored closely, with blood work for early detection of renal impairment or electrolyte/acid-base abnormalities. Additional urine studies may also be needed if distal RTA is suspected. The choice of reintroducing ICI treatment

should be decided on further discussion with the oncologist, the nephrologist, and most importantly, the patient.

REFERENCES

1. Postow MA, Sidlow R, Hellmann MD. Immune-related adverse events associated with immune checkpoint blockade. *N Engl J Med*. 2018;378:158-168.
2. Herrmann SM, Perazella MA. Immune checkpoint inhibitors and immune-related adverse renal events. *Kidney Int Rep*. 2020;5(8):1139-1148.
3. Cortazar FB, Kibbelaar ZA, Glezerman IG, et al. Clinical features and outcomes of immune checkpoint inhibitor-associated AKI: a multicenter study. *J Am Soc Nephrol*. 2020;31:435-446.
4. Devuyst O, Lemaire M, Mohebbi N, Wagner CA. Autoantibodies against intercalated cells in Sjogren's syndrome. *Kidney Int*. 2009;76(2):229.
5. Landry GM, Furrow E, Holmes HL, et al. Cloning, function, and localization of human, canine, and Drosophila ZIP10 (SLC39A10), a Zn(2+) transporter. *Am J Physiol Renal Physiol*. 2019;316:F263-F273.
6. El Bitar S, Weerasinghe C, El-Charabaty E, Odaimi M. Renal tubular acidosis an adverse effect of PD-1 inhibitor immunotherapy. *Case Rep Oncol Med*. 2018;2018:8408015.
7. Charmetant X, Teuma C, Lake J, Dijoud F, Frochot V, Deeb A. A new expression of immune checkpoint inhibitors' renal toxicity: when distal tubular acidosis precedes creatinine elevation. *Clin Kidney J*. 2020;31(2):435-446.
8. Seethapathy H, Zhao S, Chute DF, et al. The incidence, causes, and risk factors of acute kidney injury in patients receiving immune checkpoint inhibitors. *Clin J Am Soc Nephrol*. 2019;14(12):1692-1700.
9. Rodriguez Soriano J. Renal tubular acidosis: the clinical entity. *J Am Soc Nephrol*. 2002;13:2160-2170.
10. DeFranco PE, Haragsim L, Schmitz PG, Bastani B. Absence of vacuolar H(+)-ATPase pump in the collecting duct of a patient with hypokalemic distal renal tubular acidosis and Sjogren's syndrome. *J Am Soc Nephrol*. 1995;6:295-301.
11. Pereira PC, Miranda DM, Oliveira EA, Silva AC. Molecular pathophysiology of renal tubular acidosis. *Curr Genom*. 2009;10:51-59.
12. van den Wildenberg MJ, Hoorn EJ, Mohebbi N, et al. Distal renal tubular acidosis with multiorgan autoimmunity: a case report. *Am J Kidney Dis*. 2015;65:607-610.
13. Caruana RJ, Buckalew VM Jr. The syndrome of distal (type 1) renal tubular acidosis. Clinical and laboratory findings in 58 cases. *Medicine (Baltimore)*. 1988;67:84-99.
14. Burki R, Mohebbi N, Bettoni C, Wang X, Serra AL, Wagner CA. Impaired expression of key molecules of ammoniagenesis underlies renal acidosis in a rat model of chronic kidney disease. *Nephrol Dial Transplant*. 2015;30:770-781.
15. Battistone MA, Nair AV, Barton CR, et al. Extracellular adenosine stimulates vacuolar ATPase-dependent proton secretion in medullary intercalated cells. *J Am Soc Nephrol*. 2018;29:545-556.
16. Menzies RI, Tam FW, Unwin RJ, Bailey MA. Purinergic signaling in kidney disease. *Kidney Int*. 2017;91:315-323.
17. Khunger M, Rakshit S, Pasupuleti V, et al. Incidence of pneumonitis with use of programmed death 1 and programmed death-ligand 1 inhibitors in non-small cell lung cancer: a systematic review and meta-analysis of trials. *Chest*. 2017;152: 271-281.
18. Pillai RN, Behera M, Owonikoko TK, et al. Comparison of the toxicity profile of PD-1 versus PD-L1 inhibitors in non-small cell lung cancer: a systematic analysis of the literature. *Cancer*. 2018;124:271-277.
19. De Sousa Linhares A, Battin C, Jutz S, et al. Therapeutic PD-L1 antibodies are more effective than PD-1 antibodies in blocking PD-1/PD-L1 signaling. *Sci Rep*. 2019;9:11472.
20. Dinour D, Chang MH, Satoh J, et al. A novel missense mutation in the sodium bicarbonate cotransporter (NBCe1/SLC4A4) causes proximal tubular acidosis and glaucoma through ion transport defects. *J Biol Chem*. 2004;279:52238-52246.