Clin Kidney J (2015) 8: 131–136
doi: 10.1093/ckj/sfu105
Advance Access publication 20 October 2014

Clinical Nephrology: Crystalluria

Editorial Comment

Crystalline-induced kidney disease: a case for urine microscopy

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Keywords: calcium oxalate; light chains; methotrexate; uric acid; urine microscopy

Introduction

Urine microscopy is an integral part of the clinical evaluation of patients presenting with kidney disease [1, 2]. Along with history and physical examination, directed serum tests, dipstick urinalysis, and at times, renal imaging, urine sediment examination provides an informative view into the kidney. Identification and quantification of the cells, casts and crystals present in the spun urine sediment allow the clinician to synthesize all of the data and construct a rational diagnosis. This is particularly true for patients with crystalline-induced kidney disease [3]. We present illustrative cases of five different and clinically important causes of crystalline-induced kidney injury demonstrating the diagnostic utility of urine microscopy in clinching the diagnosis and facilitating therapy of the underlying process.

Calcium oxalate nephropathy

A 52-year-old man was seen in the emergency department after being found home confused and lethargic, with intermittent emesis. In the emergency department, he was intubated because of his altered mental status and respiratory compromise. Labs upon admission demonstrated a sodium of 149 mEq/L, Cl of 108 mEq/L, bicarbonate of <5 mEq/L, urea of 30 mg/dL and creatinine of 2.7 mg/dL. His serum osmolality was 418 mOsm/kg. An arterial blood gas showed a pH of 7.01, pCO_2 of 16 mmHg and bicarbonate of 3.9 mEq/L. The patient was started on fomepizole for presumed toxic alcohol ingestion, and hemodialysis was initiated.

Urinalysis revealed a specific gravity (SG) of 1.009, pH 5.5, 1+ protein and moderate blood (6–10 RBC/HPF). Urine sediment examination revealed numerous monohydrated calcium oxalate crystals both free and forming 3–5 crystalline casts/LPF, which were birefringent with polarization (Figure 1A and B), raising concern for ethylene glycol (EG) intoxication. The EG level returned at 341 mg/dL, confirming the diagnosis garnered from urine microscopy. Kidney biopsy was undertaken when the patient remained dialysis dependent for 2 weeks and demonstrated significant calcium oxalate crystal deposition within the tubules (positive birefringence) along with tubular injury and a mild interstitial inflammatory infiltrate (Figure 1C and D). These findings were consistent with acute calcium oxalate nephropathy from EG ingestion. Steroids were given to treat the inflammatory component of calcium oxalate deposition with subsequent improvement of kidney function and discontinuation of dialysis.

Urine microscopy demonstrating free calcium oxalate crystals and crystalline casts provided the diagnosis of EG intoxication prior to the confirmation by a blood test. Ingestion of EG is classically complicated by increased osmolar gap and anion gap metabolic acidosis as well as multiple organ dysfunction from toxic metabolites such as glycolic acid and oxalic acid [4, 5]. Acute kidney injury (AKI) is primarily the result of calcium oxalate deposition within the renal tubules. This process leads to obstruction of urine flow in the tubules while the intratubular crystals promote an inflammatory response within the renal interstitium. These combine to promote acute kidney dysfunction. Targeting the inflammatory pathway earlier may potentially reduce the severity and shorten the course of kidney injury, perhaps avoiding the need for continued dialysis after removal of the toxic substance and its metabolites. Visualization of calcium oxalate crystalline casts at the time of presentation can facilitate an earlier diagnosis of oxalate nephropathy and treatment with steroids to quell the inflammatory response.

Myeloma light chain crystalline nephropathy

A 56-year-old man with IgG kappa multiple myeloma presented with AKI as manifested by a rise in serum creatinine from a baseline of 1.6–3.2 mg/dL. Despite aggressive anti-myeloma therapy, the malignancy was refractory and

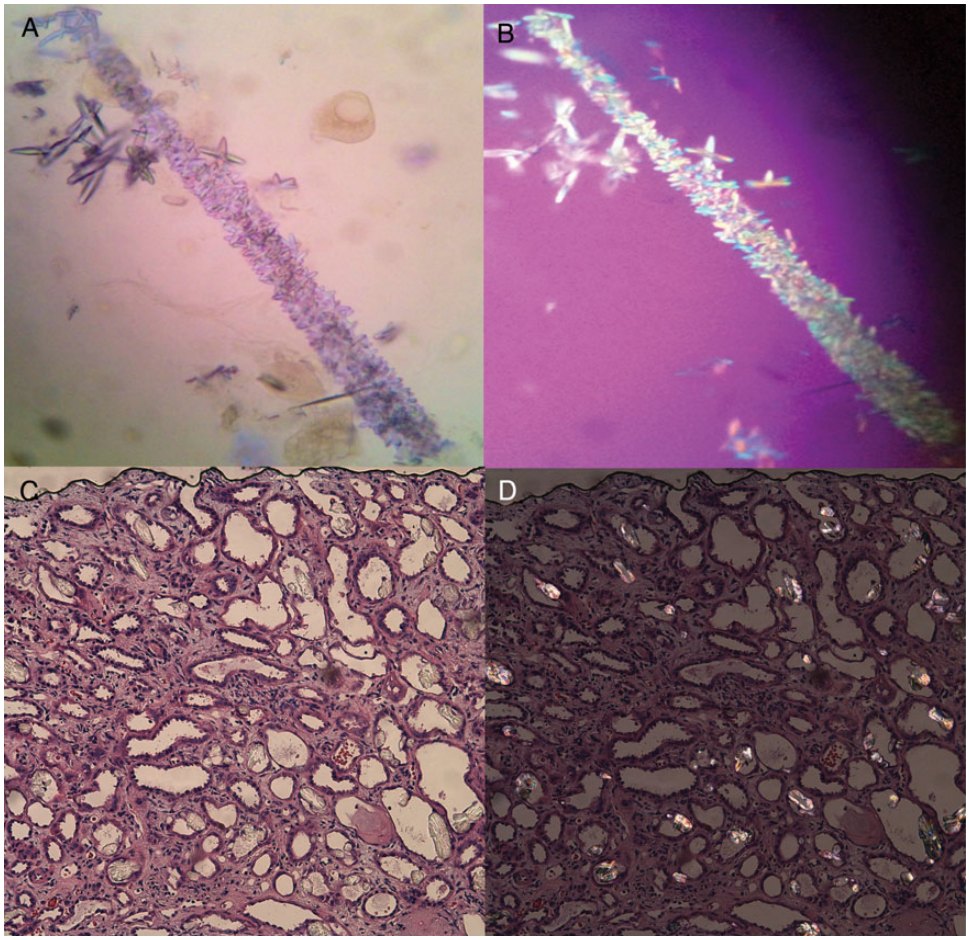

Fig. 1. Acute calcium oxalate nephropathy from ethylene glycol intoxication. (**A**) Urine sediment demonstrates calcium oxalate crystalline cast under light microscopy (×400) and with (**B**) polarization (×400). (**C**) Renal histology reveals calcium oxalate crystals within renal tubules under light microscopy (H&E, ×400) and with (**D**) polarization (×400).

serum-free light chains (LCs) increased. Blood pressure was normal and examination was unremarkable with no signs of hypervolemia or volume depletion. Laboratory data revealed normal electrolytes except for mild hyponatremia (Na 134 mEq/L), BUN of 49 mg/dL and serum creatinine of 3.2 mg/dL. Serum-free kappa LCs were elevated at 326 mg/dL, the kappa/lambda ratio was 200 and urine kappa LCs were elevated at 239 mg/dL.

Urinalysis revealed an SG of 1.010, pH 5.5, 1+ protein and 1+ blood. Urine sediment analysis demonstrated aggregates of various shaped and sized crystals and one to three crystalline casts per LPF (Figure 2A and B), which had 25% birefringence with polarization. The kidney biopsy also demonstrated LC crystalline casts. Many tubular profiles showed LC crystals in the cytoplasm of the tubular epithelial cells and eosinophilic refractile LC crystals of varying shapes in the tubular lumens (Figure 2C). Electron microscopy revealed multifaceted crystals in the tubular lumens and within the cytoplasm of tubular epithelial cells (Figure 2D). These findings were consistent with monoclonal LC-induced crystalline nephropathy as the cause of kidney injury. Despite attempts at other forms of chemotherapy, kidney function continued to decline ultimately requiring initiation of hemodialysis.

This case is unique in that free monoclonal LC crystals and LC crystalline casts were viewed in the urine sediment of a patient with IgG kappa multiple myeloma and underlying kidney disease. More sophisticated urine testing has confirmed that LCs can be seen in the urine. Immunofluorescence (IF) staining (anti-sera to LC immunoglobulins) of the urine sediment was used to identify monoclonal LCs in patients with kidney disease and various monoclonal gammopathies [6]. Urinary casts and other various shaped particles were demonstrated in the urine of 20 out of 27 patients with monoclonal disease and 0 out of 25 patients with non-monoclonal kidney disease. Monoclonal LCs are thought to be directly nephrotoxic to tubular cells, both when present within the cell cytoplasm and when they aggregate in tubular lumens [7]. These data confirm that the urine sediment contains identifiable LCs both free and within casts, while our case suggests that urine microscopy can identify LC crystals in the urine sediment, which can provide a window into the process within the renal parenchyma. It is possible that urine sediment exam in patients with an underlying monoclonal gammopathy may provide information about kidney injury and serve as a biomarker of early kidney disease.

Methotrexate crystalline-induced AKI

A 32-year-old man with a primary central nervous system lymphoma was admitted for elective chemotherapy. On

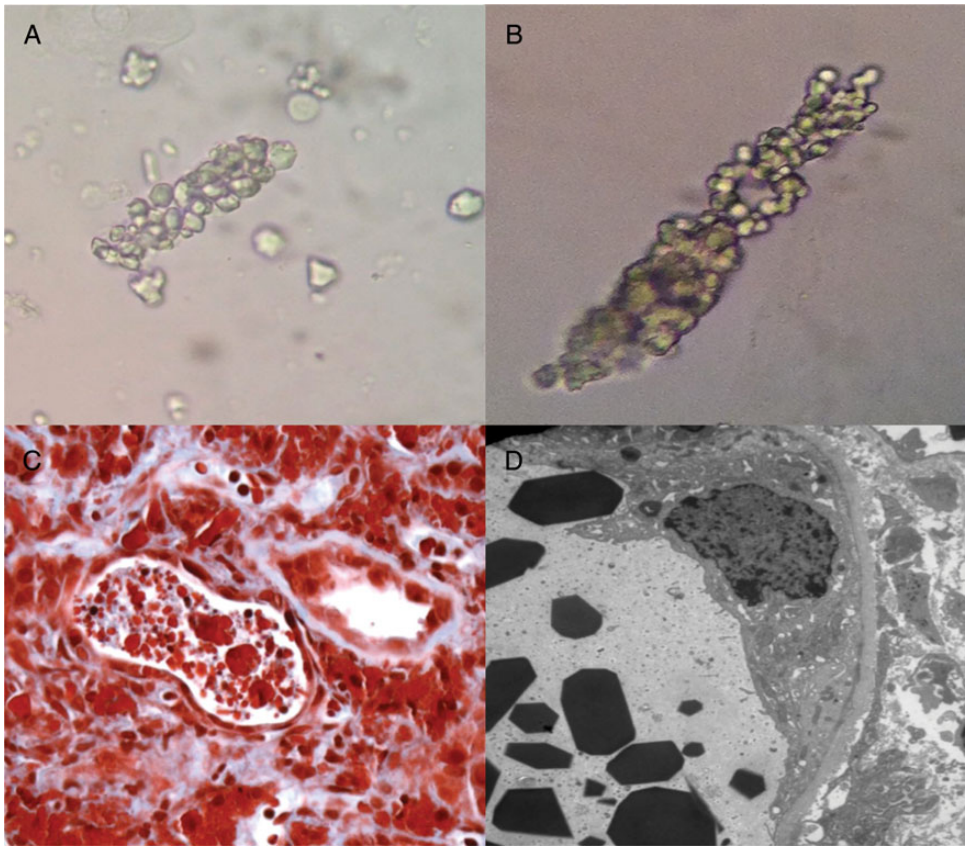

Fig. 2. Myeloma LC crystalline-induced kidney injury. (**A** and **B**) Urine sediment reveals monoclonal LC casts under light microscopy (×400). (**C**) Renal histology demonstrates monoclonal LC crystals within renal tubules under light microscopy (H&E, ×400) and with (**D**) electron microscopy.

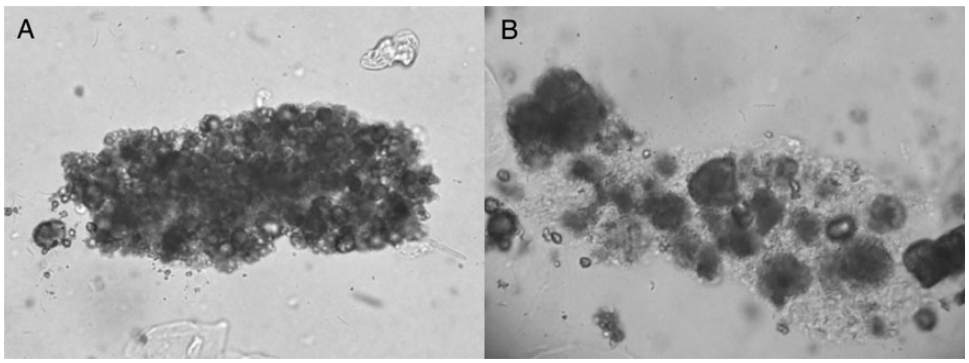

Fig. 3. Methotrexate crystalline-induced kidney injury. (**A**) Urine sediment shows free and clumped methotrexate crystals under light microscopy (×400) and a (**B**) methotrexate crystalline cast (× 400).

admission, high-dose methotrexate (5.5 g/m^2 × 1 dose) was administered after 2 L of isotonic sodium bicarbonate solution were infused. Leucovorin rescue was given 24 h following high-dose methotrexate dosing while isotonic bicarbonate intravenous infusion was continued to achieve an alkaline urine pH. Within 48 h, serum creatinine rapidly rose from 0.7 to 1.9 mg/dL consistent with AKI. On examination, blood pressure and pulse were normal without orthostatic changes, and the patient was not febrile. Urine output was consistently >100 mL/h. Lungs, heart and abdominal exam were normal. The methotrexate level at 48 h was 58 mcmol/L (0–5 mcmol/L). Electrolytes were normal except for a serum bicarbonate of 35 mEq/L. Ultrasound of the kidneys demonstrated normal sized kidneys with moderately increased echogenicity.

Urinalysis revealed that SG 1.012, pH 7.0, while blood, glucose, protein and leukocyte esterase were negative. Urine microscopy of the urine sediment revealed numerous brownish/gold crystals, which were free as well in clumps and two to three crystalline casts per LPF (Figure 3A and B). Methotrexate-induced AKI was diagnosed and intravenous fluids were continued. Kidney biopsy was not performed. One week later kidney function improved with the serum creatinine concentration reaching 1.2 mg/dL.

Even in the absence of a kidney biopsy, urine sediment examination clinched the diagnosis of methotrexate-

induced kidney injury based on the finding of crystalline casts composed of the antimetabolite. Cast formation in this setting clearly indicates that methotrexate crystals were causing renal tubular injury and the associated acute decline in kidney function. This was a relatively unexpected finding as the patient had maintained an alkaline urine and brisk urine output. High-dose methotrexate causes kidney injury through multiple possible mechanisms; however, intratubular crystal precipitation is the most likely [8–10]. Methotrexate and its metabolites precipitate within the renal tubular lumens in the setting of low urine pH, decreased urinary flow rates from volume depletion and with high urinary methotrexate concentrations [11]. Our patient's urine sediment showed not only free methotrexate crystals, but also had crystal-containing casts, which are essentially the equivalent of intratubular crystals seen in kidney tissue.

Acute uric acid nephropathy

A 43-year-old healthy woman with recent onset of fatigue, weakness, easy bruising and decreased urine production was noted to have AKI with serum creatinine 4.5 mg/dL (previous serum creatinine was 0.7 mg/dL). Blood pressure was normal and examination was notable for conjunctival pallor and lower extremity petechiae. Laboratory data revealed a serum potassium of 5.9 mEq/L, serum uric acid 15 mEq/L, serum calcium 8.1 mg/dL, serum phosphorus 8.1 mg/dL, BUN 39 mg/dL and serum creatinine 4.5 mg/dL. White blood cell count was 108×10^9/L, hemoglobin 7.9 g/dL and platelets 35×10^9/L. Renal ultrasound demonstrated normal sized kidneys with increased echogenicity.

Urinalysis revealed an SG of 1.015, pH 5.5, 1+ protein, trace blood and trace leukocyte esterase. Urine sediment examination demonstrated numerous uric acid crystals and three to five crystalline casts per LPF (Figure 4), which were birefringent with polarization. Kidney biopsy was not obtained as the patient had evidence of acute uric acid nephropathy based on the finding of uric acid crystalline casts in the urine sediment. As the patient remained oliguric despite intravenous fluid resuscitation in the setting of stage 3 AKI and tumor lysis syndrome, continuous renal replacement therapy (CRRT) was initiated and rasburicase was administered. Acute myeloid leukemia (AML, M1) was subsequently diagnosed. CRRT was discontinued after 6 days and the patient recovered kidney function after 2 weeks with serum creatinine reaching 1.1 mg/dL.

This case shows the value of urine microscopy in rapidly diagnosing the cause of AKI in this patient with spontaneous tumor lysis syndrome in the setting of previously unknown AML. In an acid urine, uric acid crystals tend to precipitate within renal tubular lumens, where they can obstruct urinary flow and incite an inflammatory reaction. Both of these effects can lead to development of AKI [12]. Uric acid crystalline casts seen in the urine sediment supported acute uric acid nephropathy rather than another cause of AKI such as acute calcium/phosphate deposition, intrarenal infiltration or leukostasis by leukemic myeloid blasts, or ischemic/nephrotoxic acute tubular necrosis. These findings confirm that urine microscopy can provide important information about the underlying kidney lesion in the setting of crystalline-induced kidney disease. These data suggested early in the course that CRRT and rasburicase would be useful despite the absence of a formal diagnosis of AML at presentation.

Sulfadiazine crystalline-induced AKI

A 46-year-old woman with a history of deceased donor kidney transplant 4 years previoulsly developed headaches and altered sensorium and was diagnosed with neurotoxoplasmosis. The patient was treated with intravenous sulfadiazine, pyrimethamine and folinic acid and reduction in immunosuppressive regimen (steroids discontinued, cyclosporine dose reduced and mycophenolate mofetil reduced). Over the next week to 10 days, the patient deteriorated with worsening sensorium, mild

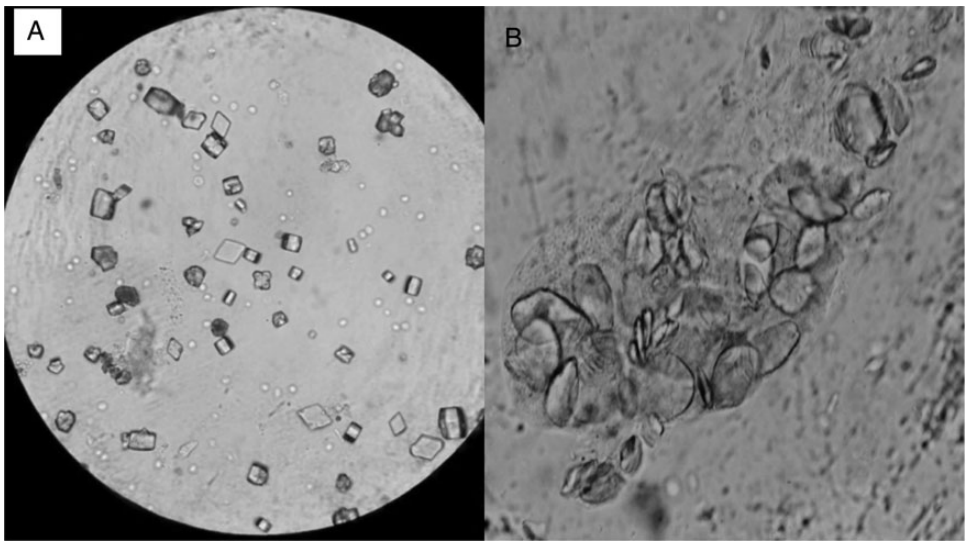

Fig. 4. Uric acid crystalline nephropathy. (**A**) Urine sediment reveals free uric acid crystals under light microscopy (×100) and a (**B**) uric acid crystalline cast (×600).

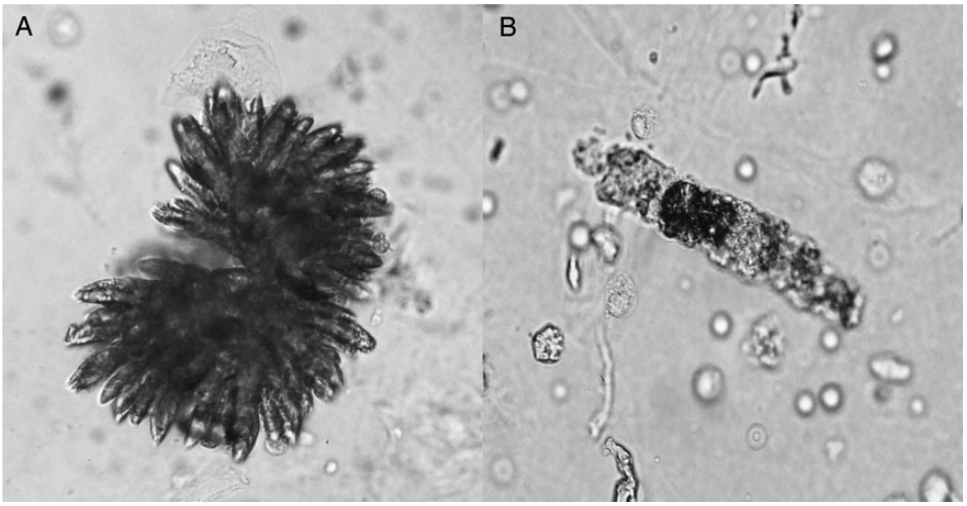

Fig. 5. Sulfadiazine crystalline-induced kidney injury. (**A**) Urine sediment demonstrates free sulfadiazine crystals under light microscopy (×100) and a (**B**) sulfadiazine crystalline cast (×200).

hypotension and a rising serum creatinine concentration (1.3–1.8 mg/dL). Blood pressure was restored with intravenous fluids. Examination was unremarkable except for fluctuating mental status. There was no rash. Renal ultrasound of the allograft revealed increased echogenicity and high resistive indices.

Urinalysis revealed an SG of 1.013, pH 5.5, trace protein, trace blood and 1+ leukocyte esterase. Urine sediment examination demonstrated five to eight white blood cells per HPF, and numerous sulfadiazine crystals and two to three crystalline casts per LPF (Figure 5A and B), which were birefringent with polarization. Kidney biopsy was not obtained as the patient had evidence of crystal-related injury based on the finding of sulfadiazine crystalline casts in the urine sediment. Urinary alkalinization and increased urinary flow rates were achieved with isotonic sodium bicarbonate intravenous fluids. Kidney function improved and returned to baseline over the next several days after the serum creatinine peaked at 2.1 mg/dL.

The utility of urine microscopy is evident in this case as the cause of AKI was quickly diagnosed and therapy was commenced for sulfadiazine crystalline-induced kidney injury. As with uric acid and methotrexate crystals, sulfadiazine crystals tend to precipitate within renal tubular lumens in an acidic urine. In this circumstance, AKI can develop [9]. Rapid intervention with bicarbonate-containing intravenous fluids helped solubilize the sulfadiazine crystals within the tubules and improve kidney function. In addition, identification of sulfadiazine crystalline casts seen in the urine sediment supported this drug crystal as the cause of AKI rather than other possibilities such as rejection, ischemic or nephrotoxic acute tubular necrosis, or sepsis.

Discussion

Urine microscopy is a crucial part of the evaluation of kidney disease. It is particularly helpful in the assessment of patients with AKI, hematuria and proteinuria [1]. Thorough examination of the spun urinary sediment provides information that cannot be otherwise obtained by dipstick urinalysis or automated/laboratory technician performed urine examination. Expert differentiation of urinary cell morphology, accurate identification of cellular and non-cellular casts and recognition and diagnosis of various endogenous and drug-related crystals can lead to rapid diagnosis of the kidney-related process.

Various crystals, whether endogenous to the body or introduced as drugs, may deposit within the renal tubules and interstitium. This is in part based on the filtering function of the kidney whereupon the organ receives nearly 25% of the cardiac output and concentrates various molecules in the tubules, in this case crystals [3]. In addition, the solubility characteristics of the crystal, and the patient's volume status, urine flow rate and urine pH contribute to crystal precipitation within the kidney.

Intratubular crystal precipitation results in both distal renal tubular obstruction and associated tubulointerstitial inflammation [13]. This latter mechanism of kidney injury likely plays an important yet under-recognized role in kidney injury based on the inflammatory pathway promoted by these intratubular crystals [14, 15]. Crystals activate innate immunity via the NACHT, LRR and PYD domains-containing protein (NLRP) 3 inflammasome. NLRP3 subsequently activates caspase-1, which cleaves two interleukin (IL) precursors to active IL-1B and IL-18 [16]. The production of these cytokines ultimately leads to inflammation within the renal tubulointerstitium, which promotes acute tubular injury and clinical AKI. Recognizing this process early with urine microscopy offers the hope for treatment with an anti-inflammatory agent such as corticosteroids.

In conclusion, it is apparent that examination of the urine sediment is an accurate diagnostic test for patients with underlying crystalline-induced nephropathy. As seen in the cases discussed in this paper, urine microscopy provided evidence that endogenous or drug-induced crystals were the proximate cause of kidney injury. By definition, the crystalline casts identified in the urine sediment supported intratubular crystal deposition and associated kidney injury. In essence, the clinician is able to visualize the 'renal histology' in the urine sediment, making the urine sediment the proverbial 'liquid biopsy'.

Conflict of interest statement. None declared.

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Received for publication: 21.8.14; Accepted in revised form: 18.9.14