

## Case Report

## Is adenine phosphoribosyltransferase deficiency a still underdiagnosed cause of urolithiasis and chronic renal failure? A report of two cases in a family with an uncommon novel mutation

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### Abstract

We describe two patients that had a history of recurrent renal stones and chronic renal insufficiency. The first case was a 51-year-old man with an adenine phosphoribosyltransferase (APRT) deficiency who was diagnosed only after he had been referred for severe renal failure requiring hemodialysis. This led to a screening of the entire family, which identified six carriers and an additional affected relative (a 41-year-old man and the second case reported herein). Genetic analysis of the *APRT* gene revealed an atypical mutation previously described only once in a compound heterozygote.

**Keywords:** adenine phosphoribosyltransferase (APRT) deficiency; 2,8-dihydroxyadenine (2,8-DHA); renal failure; urolithiasis

ranges from 10 to 25% of normal but is not functional *in vivo* and is found in Japanese patients and once in a Polish patient [2]. The disease is manifested in homozygotes or compound heterozygotes.

We identified two cases in one family, both having recurrent urolithiasis. The first patient was referred to our unit for severe renal failure requiring haemodialysis. The second patient was a 41-year-old man, who was first cousin to patient 1. Although patient 2 had a history of delivered ureteral stones and mild renal insufficiency since his youth, he was diagnosed with APRT deficiency only following a family screening. In both patients we also observed an unusual *APRT* gene mutation, consisting of a homozygous 254 bp deletion–8 bp insertion of the *APRT* gene. To the best of our knowledge, these are the first reported cases in Italy.

### Introduction

Adenine phosphoribosyltransferase (APRT) deficiency, an enzyme disorder of purine metabolism, results in an inability to recycle adenine that is instead converted into 8-hydroxyadenine and 2,8-dihydroxyadenine (2,8-DHA). These compounds are actively secreted into the urine and their insolubility at any urinary pH leads to crystalluria and stone formation.

APRT deficiency is inherited as an autosomal recessive trait; the gene is located on chromosome 16q24 and was cloned in 1987 [1].

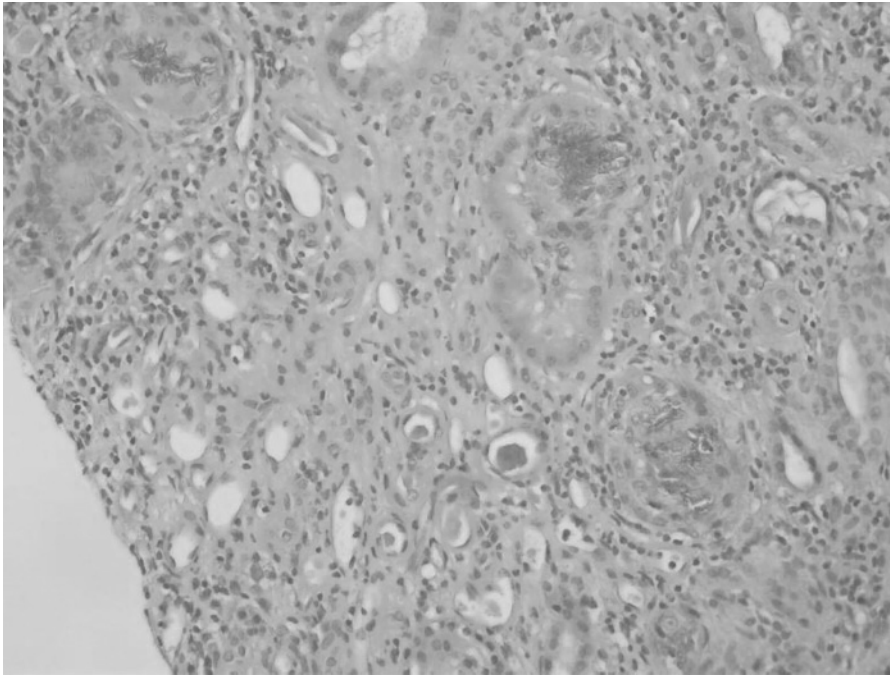
Two types of APRT deficiency have been identified and these are based on residual enzyme activity. In type I, a complete absence of enzymatic activity is found and is the characteristic of Caucasians. In type II, enzyme activity *in vitro*

### Case reports

#### Patient 1

This was a 51-year-old Caucasian man who had delivered renal stones (composed of uric acid and calcium oxalate) in 1995 and 2002; he was also suffering from a mild renal insufficiency (1.7 mg/dl serum creatinine, 150  $\mu$ mol/l). In April 2005, while receiving nephrological consultation at a different hospital, he presented with a moderate renal insufficiency (3.5 mg/dl serum creatinine, 310  $\mu$ mol/l); serum and urinary uric acid as well as blood pressure was normal, and a microscopic lithiasis was found at renal scan. He was treated with citrate, allopurinol and hydrochlorothiazide as well as a low calorie, low protein and low sodium diet. He lost 10 kg weight over 5–6 months, and diuretic therapy was stopped following a lipotimic episode. In January 2006, he was admitted to our hospital because of protracted nausea, vomiting and general fatigue over the previous 3 weeks. He showed signs of dehydration with orthostatic hypotension, severe hyponatraemia (114 mmol/l

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**Fig. 1.** Renal biopsy specimens showing severe tubulo-interstitial nephritis with birefringent needle-shaped crystalline precipitate in tubular lumen (patient 1).

$\text{Na}^+$ ) and metabolic acidosis ( $14 \text{ mmol/l HCO}_3^-$ ); we diagnosed advanced non-oliguric renal failure ( $12 \text{ mg/dl}$  serum creatinine,  $1060 \text{ }\mu\text{mol/l}$ ) and he began haemodialysis. Uricaemia was in the normal range or only slightly above the upper limit. He showed microscopic haematuria and light proteinuria, without crystalluria.

A percutaneous renal biopsy showed severe tubulointerstitial nephritis with birefringent needle-shaped crystalline precipitate in tubular lumen, tubular epithelium and interstitium. Extensive tubular atrophy, interstitial chronic inflammation and fibrosis, foreign body-type reaction, with no glomerular changes, were also observed (Figure 1). During histological examination, we were unable to determine whether crystalline deposits were of uric acid or 2,8-DHA.

Because the clinico-pathological features and laboratory findings were inconsistent with uric acid nephropathy, we performed a biochemical and molecular biological evaluation for suspected APRT deficiency. A pathological presence of adenine in serum and urine was identified and APRT enzyme activity was absent. Molecular biological analysis of the amplified *APRT* gene revealed a homozygous 254 bp deletion–8 bp insertion in exon 3. These results confirmed that our patient had APRT deficiency [3]. Although a diet poor in purine content and allopurinol was given, there was no improvement in renal function.

The patient's parents were native to two areas from the middle portion of Italy and were not related; both had many siblings, some of which had left the country. Some of these relatives were offered screening by biochemical assay for the presence of adenine in urine and serum, by erythrocyte APRT activity and by molecular biological analysis of the *APRT* gene [4]. As showed in Figure 2, a first cousin was homozygous and a sister and two first cousins were

healthy; all other relatives were carriers for the same *APRT* gene mutation. In heterozygotes, APRT enzymatic activity was reduced (Table 1), but serum and urinary adenine concentrations were non-detectable.

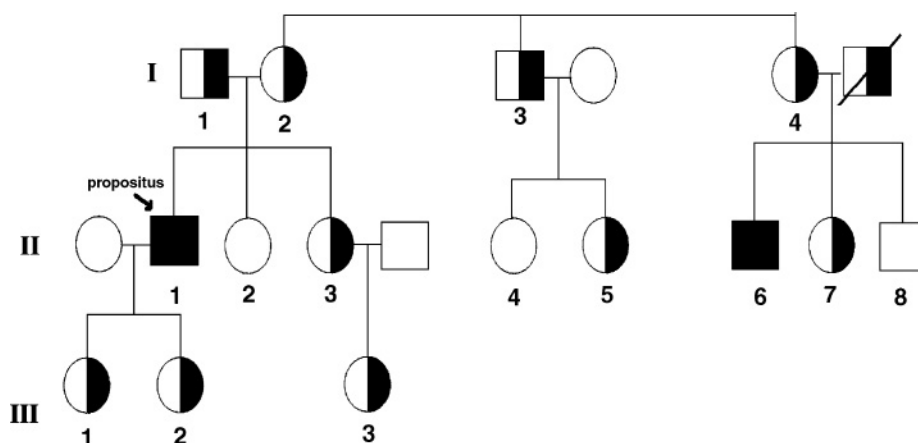
#### *Patient 2*

This subject was a 41-year-old man who was first cousin to patient 1. He had experienced mild renal insufficiency ( $1.4 \text{ mg/dl}$  serum creatinine,  $124 \text{ }\mu\text{mol/l}$ ) and had a hypoplastic kidney since his youth. He had also experienced recurrent episodes of renal colics and had delivered numerous stones until 20 years of age. At that time, the chemical composition of the stones was believed to be uric acid. Even though uricaemia had always been in the normal range, he was treated with allopurinol which stopped stone formation and no further worsening of renal function was observed. At the time of screening, allopurinol was stopped for a few days, and large amounts of adenine were detected in both serum and urine; APRT activity was totally absent and the same gene mutation as that found in the proband was identified.

His father also suffered from urolithiasis and developed severe renal failure of unknown cause that required renal replacement therapy, and he died prematurely of cancer.

## **Discussion**

The two types of APRT deficiency have a wide clinical spectrum of phenotypes that are identical and involve only the kidney. The disease ranges from minimal symptoms, such as brownish crystals on the nappy, to more severe pathologies which include stone formation in the urinary



**Fig. 2.** Isolation scheme of the *APRT* mutated alleles in members of the screened family. Filled squares = *APRT* deficiency homozygotes (II-1 = patient 1; II-6 = patient 2); half-filled symbols = *APRT* deficiency heterozygotes; open symbols = the wild-type phenotypes.

**Table 1.** *APRT* enzyme activity in the patients affected by *APRT* deficiency and in their screened relatives

	<i>APRT</i> activity (normal range: 20–40 U/l red blood cells)
Propositus (II-1)	0
Father (I-1)	9.16
Mother (I-2)	8.47
Sister (II-2)	32.44
Sister (II-3)	8.80
Daughter (III-1)	6.93
Daughter (III-2)	10.35
Niece (III-3)	4.20
Uncle (I-3)	7.9
First cousin (II-4)	38.89
First cousin (II-5)	12.52
Aunt (I-4)	11.2
First cousin (II-6)	0
First cousin (II-7)	18.65
First cousin (II-8)	36.78

tract or crystal precipitation in the renal parenchyma causing acute and chronic damage [5,6,7]. Progressive chronic renal failure, due to tubulointerstitial nephritis, is less common and has been reported in patients having a history of urolithiasis as well as in the absence of renal stone formation or chronic obstructive uropathy; it can also recur in renal allografts. Approximately 15% of homozygotes are symptomless, and in these patients the disease has been diagnosed during routine urine analysis or during family screening. The onset of symptoms can occur at any age, from birth to the 7th decade, and in one-third of cases they appear only in adulthood [5,6,8]. An association between hypoplastic kidney and *APRT* deficiency has also been described and a genetic correlation is suspected [9].

*APRT* deficiency, described for the first time in 1974 [10], is considered as a rare inborn error of metabolism but its exact prevalence is still unknown [8]. According to some authors, heterozygote status is relatively common, occurring in 0.4–1.2% of the population, and the expected homozygosis should therefore range from 1:250 000 to 1:30 000 of live births [6], which contrasts with an Australian study showing a much higher incidence (1:250

of disease [11]. Only about 300 cases have been described worldwide, with most occurring in Japan and Iceland. In the majority of these cases, there was a 20- to 50-year span from the initial symptoms to recognition of the stone composition; in some cases, the enzyme defect had been identified just after renal transplantation or allograft rejection [12–15]. These findings strongly support the argument that this remains an underdiagnosed condition. Although a lack of symptoms in some patients is one of the reasons, this disorder is still under-recognized as a cause of urolithiasis and renal damage, even among urologists and nephrologists. In addition, a misdiagnosis of uric acid stones frequently occurs. In these cases, as in our patient 2, a generic treatment with allopurinol is very often performed without the exact diagnosis. However, it is important to emphasize that even if the two stone types are radiolucent and have the same behaviour upon routine chemical testing, they have very different macroscopic features, and UV as well as infrared spectrophotometer assays would be useful for correct identification. Finally, patients with *APRT* deficiency have serum and urinary uric acid levels in the normal range, while the 2,8-DHA crystals are usually abundant in urine of untreated subjects.

In type I *APRT* deficiency, 17 different mutations have been described and two of these are most frequently found in Caucasian populations. The mutation found in members of the family in this report is unusual and was previously described in only one allele of a compound heterozygote [16] but never in homozygotes.

The clinical history of our patients illustrate that

- *APRT* deficiency is still grossly misdiagnosed or underdiagnosed and it will be many years before we recognize the exact nature of this disorder;
- it is easily treatable and that allopurinol provides an effective treatment because it prevents clinical symptoms and renal damage or may improve renal dysfunction in most cases; nevertheless, an early and a well-timed diagnosis and treatment are mandatory to prevent the more deleterious consequences. In patient 1, the previously described therapy may have accelerated his worsening renal function. When *APRT* deficiency was recognized and a

proper treatment was begun, the patient showed no recovery in renal function, and this was probably because of already advanced renal damage. In this patient, the absence of urinary 2,8-DHA crystals upon microscopic analysis was probably due to a lowered 2,8-DHA clearance; however, the clinician's inability to identify them cannot be ruled out.

We also argue that an accurate diagnosis is needed for this disorder, and that it is identified only when clinicians are looking for it. A skilful routine microscopic analysis of urine is essential. We also believe that purine metabolites should be measured in serum and urine of patients that have suspected uric acid stones but also have normal uricaemia and/or urinary uric acid. In such cases, determination of APRT activity and molecular analysis of the *APRT* gene would allow definitive diagnosis.

*Conflict of interest statement.* None declared.

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