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High prevalence of and potential mechanisms for chronic kidney disease in patients with acute intermittent porphyria

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Acute intermittent porphyria (AIP) is a genetic disorder of the synthesis of heme caused by a deficiency in hydroxymethylbilane synthase (HMBS), leading to the overproduction of the porphyrin precursors δ-aminolevulinic acid and porphobilinogen. The aim of this study is to describe the clinical and biological characteristics, the renal pathology, and the cellular mechanisms of chronic kidney disease associated with AIP. A total of 415 patients with HMBS deficiency followed up in the French Porphyria Center were enrolled in 2003 in a population-based study. A follow-up study was conducted in 2013, assessing patients for clinical, biological, and histological parameters. *In vitro* models were used to determine whether porphyrin precursors promote tubular and endothelial cytotoxicity. Chronic kidney disease occurred in up to 59% of the symptomatic AIP patients, with a decline in the glomerular filtration rate of ~1 ml/min per 1.73 m^2 annually. Proteinuria was absent in the vast majority of the cases. The renal pathology was a chronic tubulointerstitial nephropathy, associated with a fibrous intimal hyperplasia and focal cortical atrophy. Our experimental data provide evidence that porphyrin precursors promote endoplasmic reticulum stress, apoptosis, and epithelial phenotypic changes in proximal tubular cells. In conclusion, the diagnosis of chronic kidney disease associated with AIP should be considered in cases of chronic tubulointerstitial nephropathy and/or focal cortical atrophy with severe proliferative arteriosclerosis.

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Acute intermittent porphyria (AIP) is an inherited autosomal dominant disorder of the synthesis of heme owing to a defect in hydroxymethylbilane synthase (HMBS).[1,2] The prevalence of symptomatic disease in Europe is 1/180,000,[3] but the prevalence of *HMBS* mutations in the general population is 1/1675,[4] suggesting that the prevalence of the disease, based on acute AIP symptoms, is underestimated, in part because the clinical manifestations are nonspecific and clinicians are often unaware of the underlying diagnosis. HMBS deficiency fosters the accumulation of the porphyrin precursors δ-aminolevulinic acid (ALA) and porphobilinogen (PBG; Supplementary Figure S1 online). In situations in which heme synthesis is stimulated in the liver, such as during the menstrual cycle, caloric restriction, infection, or the use of medications that induces P450 cytochromes synthesis, the serum concentrations of ALA and PBG greatly increase. This increase may prompt acute symptoms of the disease to occur. The acute clinical expression of AIP is the neurovisceral attack. An AIP acute attack is mainly characterized by severe abdominal pain, often accompanied by nausea, vomiting, tachycardia, and hypertension. The acute attacks may be complicated by neurologic findings: agitation, confusion, peripheral neuropathy, and coma.

During AIP attacks, ALA and PBG are massively excreted in urine, where their presence confirms the diagnosis.[5] The treatment of AIP relies on the correction/avoidance of precipitating factors, hydration, nutritional support, pain

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Table 1 | Clinical and laboratory characteristics of the '2013 cohort' ($n = 136$)

	AIP patients ($n = 74$)	Asymptomatic carriers ($n = 62$)	*P*-value
Age (years)	65 (56–72)	59 (51–76)	0.3
Time since AIP diagnosis (years)	28 (22–32)	—	—
Chronic AIP (>4 crisis/year)—*n* (%)	4 (5.5)	—	—
Female sex—*n* (%)	62 (87)	41 (68)	0.002
Hypertension—*n* (%)	46 (62)	25 (42)	0.006
Diabetes—*n* (%)	2 (3)	3 (5)	0.6
2003 eGFR (ml/min per 1.73 m^2)	60 (48–74)	78 (70–88)	<0.0001
2013 eGFR (ml/min per 1.73 m^2)	51 (41–67)	84 (70–93)	<0.0001
2003 CKD—*n* (%)[a]	36 (48)	5 (8)	<0.0001
2013 CKD—*n* (%)[a]	44 (59)	7 (11)	<0.0001
ESRD—*n* (%)[b]	5 (6.7)	0	<0.0001
Kidney transplantation	2 (2.7)	0	—
Urine protein/creatinine ratio (g/mmol creatinine)	0.04 (0–0.1)	0 (0–0.07)	0.3
Serum uric acid (μmol/l)	372 (316–452)	227 (327–625)	<0.0001
Urine ALA/creatinine (μmol/mmol creatinine)[c]	4 (2.7)	2.6 (1.4–2.6)	<0.0001
Urine PBG/creatinine (μmol/mmol creatinine)[d]	2.1 (4.9–10)	0.4 (0.8–3)	0.007

Abbreviations: AIP, acute intermittent porphyria; ALA, δ-aminolevulinic acid; CKD, chronic kidney disease; eGFR, estimated glomerular filtration rate; ESRD, end-stage renal disease; PBG, porphobilinogen.
Continuous variables are expressed as median and interquartile ranges, and nominal variables as number and proportion. The values are those obtained in 2013, except for those specified as obtained in 2003.
[a]CKD is defined as eGFR < 60 ml/min per 1.73 m^2.
[b]ESRD was defined as the need for chronic dialysis therapy or kidney transplantation.
[c]Normal value < 3 μmol/mmol creatinine.
[d]Normal value < 1 μmol/mmol creatinine.

relief, and, in severe cases, administration of heme arginate, which inhibits ALA synthase and reduces ALA and PBG synthesis.[6] Neurovisceral attacks usually resolve within 4 days.

The long-term complications of AIP include hepatic carcinoma (HCC) without cirrhosis,[7–9] and chronic kidney disease (CKD). The association of CKD with AIP has been described, and few reports have documented a chronic tubulointerstitial nephropathy with mild hypertension.[10–13]

No large-scale follow-up study of CKD associated with AIP has been conducted to date, and the biological mechanisms that promote kidney injury are not known. We conducted this population-based, observational study to provide a comprehensive survey of the clinical, biological, and pathological characteristics of CKD associated with AIP (hereafter referred to as PAKD, for porphyria associated kidney disease), and we used *in vitro* models to provide mechanistic insights into how porphyrin precursors might promote kidney injury.

RESULTS

CKD is highly prevalent among AIP patients

In 2003, the French Porphyria Center performed an observational study of a population of 415 patients (184 AIP patients and 231 asymptomatic carriers) who provided a serum creatinine measurement. The demographic characteristics of this population are shown in Supplementary Table S1 online. As expected, most of the patients were lean women, with a median age of 50 years, with a prevalence of liver cancer significantly higher (3%) compared with the asymptomatic carriers (0.4%). Ten years later, in 2013, these 415 patients were contacted by mail to provide responses to a detailed questionnaire and to undergo some specific biological measurements at the steady state of the disease (see Supplementary Materials and Methods online). The response rate to this survey was 33% (136/415), with 74 AIP patients and 62 asymptomatic carriers (the '2013 cohort'). The demographic and medical characteristics of this cohort are listed in Table 1; except for estimated glomerular filtration rate (eGFR) and age, there was no difference between the 2003 and 2013 cohorts at these two time points. The majority of the AIP patients were female, with no significant difference in age compared with the asymptomatic carriers. As expected, the ALA and PBG urinary concentrations at the steady state were significantly lower in asymptomatic carriers compared with the AIP patients.

CKD, defined as eGFR < 60 ml/min per 1.73 m^2, was diagnosed in 59% of the AIP patients, compared with 11% in the asymptomatic carriers (Table 1 and Figure 1a). The renal function observed in asymptomatic carriers was similar to the general population.[14,15] Proteinuria/creatininuria ratio was negligible. Hypertension was frequent in this population, as 62% of the AIP patients were hypertensive, compared with 42% of the asymptomatic carriers ($P = 0.006$). Among the 184 AIP patients in the 2003 cohort, 5 (2.7%) have been identified as having reached end-stage renal disease between 2003 and 2013, and 2 of them received a kidney transplant.

AIP contributes to CKD

Hypertension and 'AIP patients' status occurred more frequently in individuals with CKD (Table 2), and the unadjusted odds ratio for hypertension was 3.8 (95% confidence interval (CI) 1.8–8.1, $P = 0.0004$) and for AIP patients the odds ratio was 11.9 (95% CI 4.6–26, $P < 0.0001$). As AIP patients are frequently hypertensive,[10] and hypertension contributes to CKD, we determined whether AIP was independently associated with CKD (defined by an eGFR < 60