

# Primary hyperoxalurias: diagnosis and treatment

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**Abstract** Primary hyperoxalurias (PH) comprise a group of three distinct metabolic diseases caused by derangement of glyoxylate metabolism in the liver. Recent years have seen advances in several aspects of PH research. This paper reviews current knowledge of the genetic and biochemical basis of PH, the specific epidemiology and clinical presentation of each type, and therapeutic approaches in different disease stages. Potential future specific therapies are discussed.

**Keywords** Primary hyperoxaluria · Oxalate · Kidney stones · Nephrocalcinosis · Hereditary kidney disease

## Introduction

The first report of multiple oxalate renal calculi and nephrocalcinosis was by Lepoutre in 1925 [1]. Archer et al. [2] first used the term primary hyperoxaluria to describe two cases of extensive kidney stones that began in early childhood. Since then, the definition of primary hyperoxaluria has been extended to encompass a group of recessive inborn errors of glyoxylate metabolism characterized by overproduction of oxalate mainly in the liver and massive excretion of oxalate in the urine. Calcium oxalate is insoluble in body fluids and tends to precipitate primarily in the kidney, forming urinary calculi or diffuse nephrocalcinosis. Calcium oxalate crystals can obstruct the renal tubules, disrupt cellular function, and

damage nearby cells by promoting inflammation and inducing oxidative stress [3–5]. As glomerular filtration rate (GFR) declines, the oxalate load produced by the liver can no longer be cleared by the kidneys, plasma oxalate increases, and calcium oxalate crystals are deposited in virtually all tissues in a devastating process termed systemic oxalosis. Patients can present at any age, ranging from infantile oxalosis with early-onset end-stage renal disease (ESRD) to nephrolithiasis first presenting in the seventh decade of life.

The most common, and most severe, form is primary hyperoxaluria type 1 (PH1; OMIM #259900) caused by absence, deficiency, or mistargeting of liver-specific enzyme alanine:glyoxylate aminotransferase (AGT). Primary hyperoxaluria types 2 (PH2; OMIM #260000) and 3 (PH3; OMIM #613616) are rarer and are caused by dysfunction of glyoxylate reductase–hydroxypyruvate reductase (GRHPR) and 4-hydroxy-2-oxoglutarate aldolase (HOGA1), respectively. Secondary hyperoxaluria, due to intestinal overabsorption or poisoning, can also occur and should be excluded prior to establishing the diagnosis of PH but is beyond the scope of this review.

## Clinical manifestations

The most common clinical feature of PH is nephrolithiasis (present in up to 90 % of patients at diagnosis), which can be identified by ultrasound (US) performed for evaluation of hematuria, abdominal pain, and/or recurrent urinary tract infections [6, 7]. Diffuse nephrocalcinosis is noted in about half of such patients. A significant proportion (20–59 % [7, 8]) present with end-stage renal disease (ESRD), and some are only diagnosed after renal allograft failure. About 10 % are identified before symptoms occur by screening family members of a symptomatic patient. In two registries, diagnosis was typically made early, with median age at symptom onset and

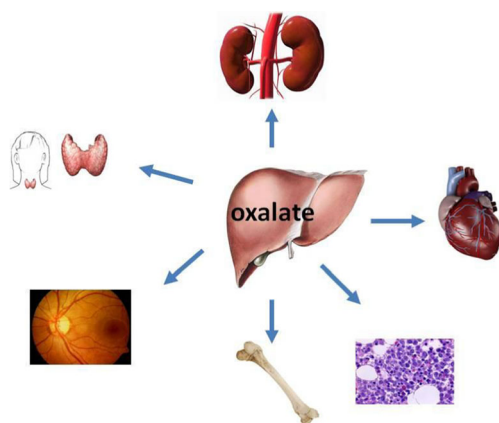
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diagnosis 5.5–6 and 7.3–10.0 years, respectively [6, 7]. Between 18 % and 38 % of PH patients are diagnosed as adults [6, 8]. Time from initial symptoms and definitive diagnosis of PH may reflect a lack of familiarity with this condition by the treating physicians.

### Primary hyperoxaluria type 1

PH1 is the most prevalent and severe form of primary hyperoxaluria. More than 50 % of patients progress to ESRD [9], with 28 % requiring renal replacement therapy (RRT) before the age of 15 [10]. Children with PH1 starting RRT are significantly younger than non-PH patients: median age 5.3 vs. 11.4 years [11]. Pyridoxine-responsive patients have a better prognosis. Infantile oxalosis is reported to account for 40 % of patients with PH requiring RRT [11], with inferior survival rates compared with non-PH patients: 86 %, 79 %, and 76 % at 1, 3, and 5 years from RRT initiation compared with 97 %, 94 %, and 92 % in non-PH patients [11]. This can be partly ascribed to systemic oxalosis, a process in which the decline in GFR results in plasma oxalate concentrations above the supersaturation threshold level. This inevitably leads to calcium oxalate deposition throughout the body, with devastating consequences (Fig. 1). Bone disease is most pronounced in the areas of rapid growth. Initially, transverse translucent symmetrical bands with thick, sclerotic margins can be seen at the end of the long bones. Advanced disease is characterized by cystic rarefaction of bones, loss of normal trabecular structure, and metaphyseal widening, causing multiple pathological fractures [12]. Calcium oxalate deposition was also noted in the periodontium, with bone loss and external tooth resorption [13]. Other manifestations include refractory anemia secondary to bone marrow replacement, hypothyroidism due to damage to the thyroid gland, myocardial deposition with conduction defects or cardiomyopathy, retinopathy, vasculopathy, and cerebrovascular infarction [14–19]. Interestingly,



**Fig. 1** Organs affected in primary hyperoxaluria (PH). Liver-produced oxalate induces kidney failure, and systemic oxalosis follows, damaging heart, blood vessels, bone, bone marrow, retina, and thyroid gland

the liver, which is responsible for oxalate overproduction in patients with PH, is spared. Kidney transplantation without removal of the native liver results in rapid graft loss due to deposition of oxalate in the transplanted kidney [20].

Variable phenotypes have been reported, even within the same family, ranging from asymptomatic urinary oxalate excretion and nephrolithiasis in adulthood to early-onset ESRD and systemic oxalosis [21, 22]. Frishberg [23] described the extreme of intrafamilial phenotypic variability among patients with PH1 bearing the same genotype: The first baby in that family was born >30 years ago with ESRD. A US performed shortly before birth showed massive nephrocalcinosis, and the neonate succumbed to the disease at the age of 3 days. Four additional members of this family were diagnosed with PH1 between 2 and 7 months of age, developed ESRD associated with severe oxalosis, and died before the age of 3 years. Interestingly, their 20-year-old brother was diagnosed with PH1 upon screening of the entire family, and he remained absolutely asymptomatic with no evidence of nephrocalcinosis or nephrolithiasis. Takayama [24] reported a milder form of PH1 in Japanese patients, possibly reflecting differences in genetic background or epigenetic factors.

### Primary hyperoxaluria type 2

PH2, described by Williams et al. [25], is rarer than PH1 and seems to have better long-term outcome [26]. Progression to ESRD occurs later than in PH1 [27, 28]. Hyperoxaluria-related graft failure after isolated renal transplantation has been described [29]. There is considerable clinical overlap between PH1 and PH2, making it impossible to differentiate clinically between them.

### Primary hyperoxaluria type 3

PH3 is typically characterized by early-onset nephrolithiasis with normal renal function. First or recurrent stone formation during the second or third decade of life has also been observed. No patient so far has been reported with diffuse nephrocalcinosis or progression to ESRD [30]. Some patients achieve complete clinical remission despite persistent hyperoxaluria [31]. Furthermore, several siblings of PH3 patients, identified as having the same homozygous mutation based on family screening, remain asymptomatic into their third decade of life despite having persistent hyperoxaluria [30, 32]. The yet undefined pathogenetic mechanism of asymptomatic hyperoxaluria is intriguing, as it may pave the pathway to developing therapeutic strategies for patients with PH. One possible mechanism may be related to the fact that PH3 represents derangement in the catabolism of hydroxyproline, which is extremely high in the urine of healthy infants, reflecting high collagen turnover, but decreases gradually and stabilizes at approximately 10 years of age [33].

Similarly, 4-hydroxy-glutamate (4OHGlu) excretion, a metabolite of hydroxyproline, steadily declines with patient age, and there is a rough correlation between 4OHGlu and 4-hydroxyproline. A decline in 4OHGlu levels with age is also apparent in PH3 patients, presumably the result of a similar mechanism to that in controls [34].

## Epidemiology

The exact prevalence of hyperoxaluria is unknown. Data is available only for PH1. European surveys suggest a prevalence of 0.7–2.9 per million population and a yearly incidence of 0.12–0.15 per million population or 1/120,000 live births [6, 9, 34, 35]. Higher prevalence is found in Mediterranean consanguineous populations [36]: PH1 accounts for 10 % of pediatric ESRD in Kuwait [37] and 13 % in Tunisia [38] compared with 0.5–2 % in Europe, North America, and Japan [11, 39, 40]. An adult dialysis unit from the Jenin District, Palestine, reported PH1 accounting for 10.7 % of prevalent patients [41].

PH2 and PH3 are presumably rarer, with a few dozen cases reported for each type [30, 31, 42–44]. PH3 became the second most prevalent form of PH [45]. Analysis of the US National Institutes of Health National Heart, Lung, and Blood Institute (NHLBI) Exome Sequencing, and the 1000 Genomes Project databases revealed higher-than-expected carrier frequency (CF) and incidence rate (IR), with PH1 and PH3 having similar CFs (~1:174 vs ~1:204) and IRs (~1:119,714 vs ~1:165,029). PH2 was ~94 % less prevalent (CF ~1:807, IR ~1:2,602,898). The much higher prevalence of PH1 compared with PH3 in clinical registries may reflect underdiagnosis of the latter because of its much milder phenotype or lower penetrance [46]. The increasing availability of genetic testing may provide more accurate information in the future.

## Genetics

PH1 is caused by mutations in the *AGXT* gene, a single-copy gene encoding the liver-specific enzyme AGT. *AGXT* is composed of 11 exons spanning 10 kbp and located on chromosome 2q36-q37 [47]. At least 178 pathogenic mutations have been described, involving all exons [48]. The majority are point mutations (missense, nonsense and splice-site mutations), and approximately a quarter are minor deletions or insertions [49]. The so-called minor allele haplotype is composed of a number of polymorphic variants (allelic frequency 5–10 %), all of which include a substitution of leucine to proline at codon 11 (c.32C>T, p.Pro11Leu). The minor allele has a reduced enzymatic capacity [50]. Furthermore, the P11L substitution introduces a weak mitochondrial targeting signal

that in conjunction with a pathogenic variant, particularly c.508G>A (p.Gly170Arg), causes hyperoxaluria by peroxisome-to-mitochondrion mistargeting of AGT [51, 52]. Multiple mutations can be found even within a distinct ethnic group [53]. No clear genotype–phenotype correlation has been found [23, 54], except for pyridoxine responsiveness, which seems to be particularly beneficial in patients homozygous for the p.Gly170Arg or Phe152Ile mutations [55, 56].

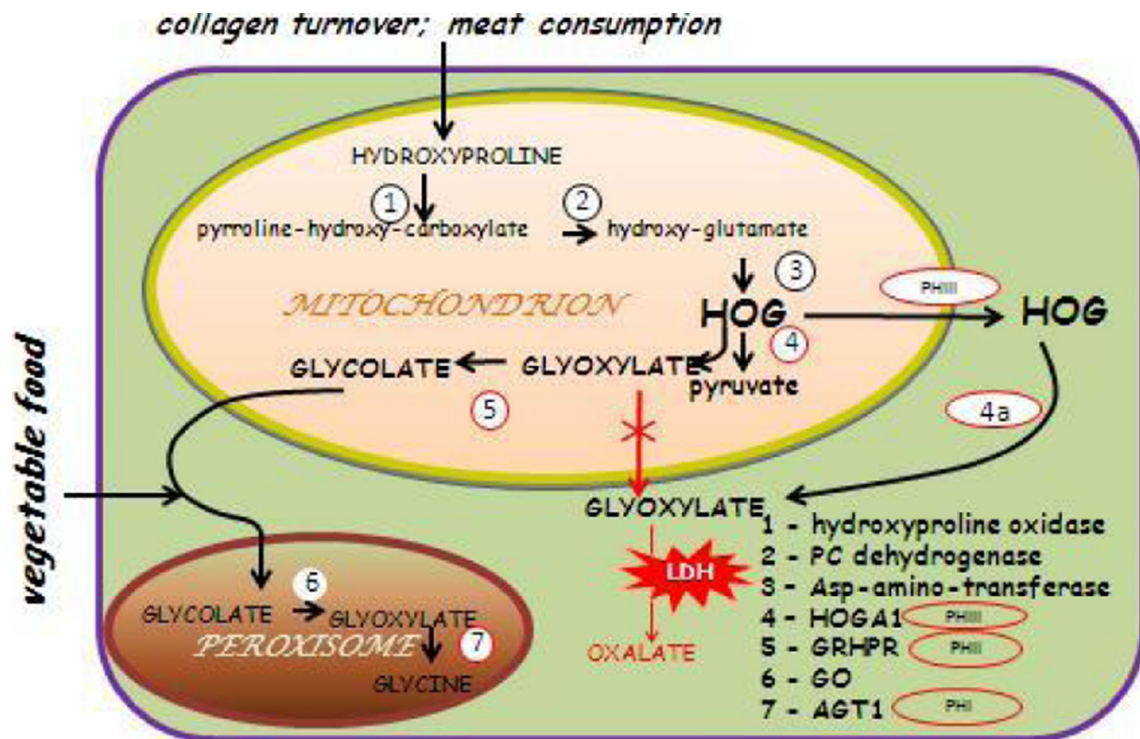
In PH2, the gene encoding GRHPR is located in the centromeric region of chromosome 9 and contains nine exons spanning 9 kbp [57, 58]. At least 30 mutations in the *GRHPR* gene have been described [43, 59, 60]. A possible founder effect, of northern European origin, has been found for the two most common mutations [61].

In 2010, Belostotsky [30] identified mutations in the *HOGAI* gene (formerly *DHDPSL*, chromosome 10q24.2) as causing PH3. About 20 pathogenic mutations in *HOGAI* have been described, including a 3-bp deletion (c.944\_946delAGG) prevalent among individuals of Ashkenazi Jewish descent. Beck et al. [31] described a potential European founder mutation (c.700+5G>T) as well as a triallelic PH mutation: homozygous *HOGAI* and a heterozygous *AGXT* mutation. Heterozygous *HOGAI* mutations were implicated as a putative risk factor for idiopathic calcium oxalate urolithiasis [62].

## Biochemistry

Endogenous oxalate (HOOC-COOH) is a metabolic end product synthesized mainly in the liver and excreted almost exclusively in the urine. There is no evidence of oxalate being used or catabolized in mammalian tissue [63]. Most oxalate excreted in the urine of both normal individuals and PH patients is endogenous in origin, and only a small amount is derived from the diet. The main precursor of oxalate in humans is glyoxylate. Glyoxylate is produced either by peroxisomal oxidation of glycolate from vegetable matter or by catabolism of hydroxyproline derived from turnover of collagen, a major constituent of extracellular matrix and meat products, taking place in the mitochondria. Interestingly, there is a clear correlation between diet and intracellular compartmentalization of glyoxylate detoxification in different mammalian species: AGT in all herbivores studied is located either exclusively in the peroxisome, or in both the peroxisome and the mitochondria. Conversely, in carnivores, AGT is either mitochondrial or both mitochondrial and peroxisomal. Most omnivores have both mitochondrial and peroxisomal AGT. Humans are unique in having purely peroxisomal AGT despite having an omnivorous diet [64].

Glycolate is converted by glycolate oxidase (GO) to glyoxylate. Glyoxylate undergoes transamination to glycine by AGT, with alanine as the amino donor and pyridoxal 5'-phosphate (active vitamin B6 derivative) as a cofactor (Fig. 2).



**Fig. 2** Cellular glyoxylate metabolism

Hydroxyproline is catabolized to glyoxylate and pyruvate in the mitochondria. This glyoxylate is reduced to glycolate by glyoxylate reductase–hydroxypyruvate reductase (GRHPR). Glycolate then enters the peroxisome and is catabolized to glycine, as described above.

AGT deficiency causes glyoxylate accumulation in PH1 [65]. Glyoxylate is mainly metabolized to oxalate by cytosolic lactate dehydrogenase (LDH) and partly reduced by cytosolic GRHPR to glycolate. This results in massive hyperoxaluria accompanied by glycolic aciduria, which is the hallmark of the disease. In about a third of cases, functional AGT is produced but is mistargeted to the mitochondria [66]. Lack of AGT in the peroxisome results in an identical PH1 phenotype.

GRHPR deficiency in PH2 [25] results in glyoxylate accumulation and hyperoxaluria. Hydroxypyruvate, which is normally reduced by GRHPR to D-glycerate, is converted by LDH to L-glycerate in its absence. The urine of PH2 patients is therefore characterized by hyperoxaluria combined with L-glycemic aciduria.

Deficiency in HOGA1, the final enzyme in mitochondrial hydroxyproline catabolism, is responsible for PH3 [30, 67]. The mechanism by which HOGA1 deficiency causes hyperoxaluria has not been elucidated. A putative explanation is that under normal conditions, glyoxylate is converted to glycolate in the mitochondria, rather than in the cytosol where it can be catabolized to oxalate (Fig. 2). Glycolate exits the mitochondria and enters the peroxisome where it is metabolized to glycine. HOGA1 deficiency results in accumulation of

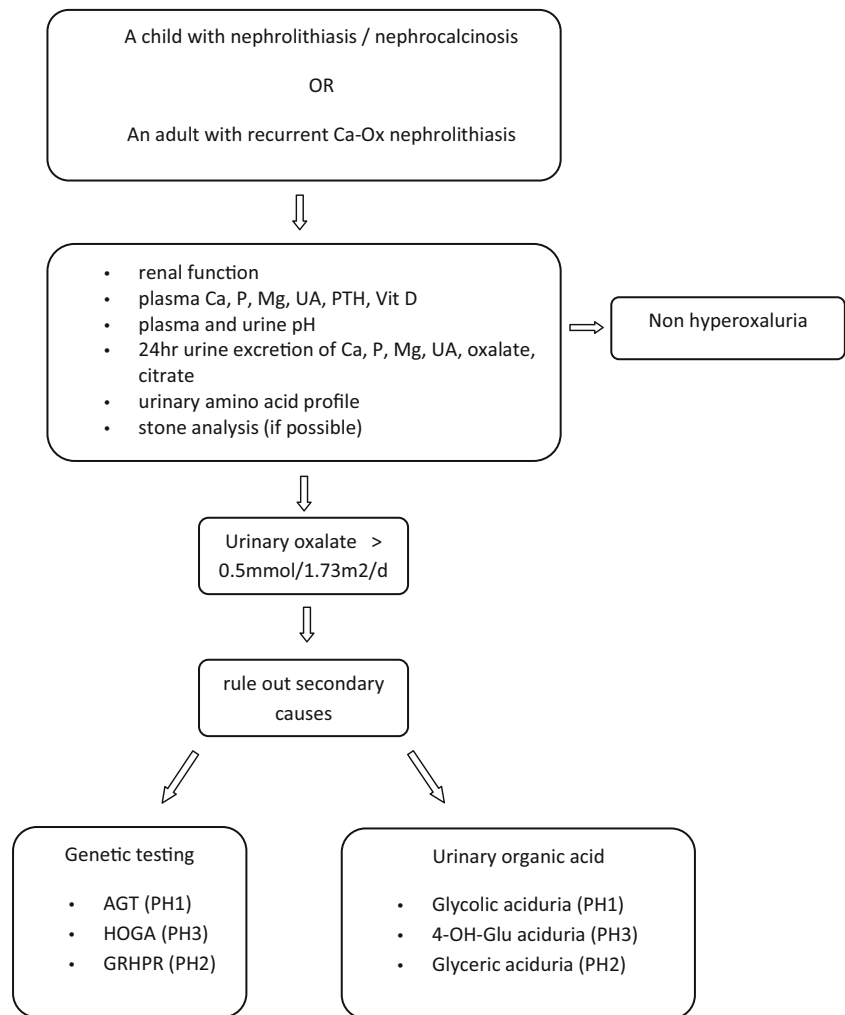
mitochondrial 4-hydroxy-2-oxoglutarate (HOG), which leaks to the cytosol, where it can be converted to glyoxylate by a yet unknown pyruvate aldolase, and oxidized by cytosolic LDH to oxalate [32]. Alternatively, it has been suggested that HOG builds up and down-regulates mitochondrial GRHPR activity, resulting in a phenotype reminiscent of PH2 [68].

## Diagnosis

PH are rare diseases and can present at any age from infancy to late adulthood. A high index of suspicion is essential to avoid delay in diagnosis and provide timely treatment. All children with nephrolithiasis or nephrocalcinosis should be evaluated for an underlying metabolic disorder, as these are prevalent in the pediatric population. This includes measuring urinary excretion of sodium, calcium, phosphorus, magnesium, uric acid, oxalate, and citrate, as well as checking spot urine pH and amino acids and measuring urine volume. Adults with recurrent calcium oxalate kidney stones should also be evaluated for PH. Our suggestion for a simple diagnostic flow-chart is depicted in Fig. 3. Evidence-based guidelines for diagnostic workup have been published [69, 70].

Passed stones are composed of virtually pure calcium oxalate monohydrate (COM, also known as whewellite) in patients with PH1 [71]. PH stones have a light surface color and are composed of an aggregation of different-sized crystals, as opposed to idiopathic calcium oxalate calculi, which are strongly pigmented. Stones from PH3 patients (and one

**Fig. 3** Diagnostic schema for suspected hyperoxaluria. *Ca-Ox* calcium oxalate, *Ca* calcium, *P* phosphorus, *Mg* magnesium, *UA* uric acid, *PTH* parathyroid hormone, *vit D* vitamin D, *AGT* alanine:glyoxylate aminotransferase, *HOGA* 4-hydroxy-2-oxoglutarate aldolase, *GRHPR* glyoxylate reductase–hydroxypyruvate reductase, *4-OH-Glu* 4-hydroxyglutamate, *PH1/PH2/PH3* primary hyperoxaluria types 1/2/3



treatment-naïve PH1 patient) have exhibited both COM and calcium oxalate dehydrate (COD, weddellite) crystals [72]. Abundant oxalate crystals in a kidney biopsy, especially if associated with interstitial inflammatory reaction and granuloma formation, may also be a clue for the diagnosis of PH.

Urinary oxalate should be measured in a 24-h urine collection, if possible. PH patients generally have oxalate excretion that exceeds twice the upper limit of normal (<0.45 mmol/1.73 m<sup>2</sup>/24 h). Levels >0.5 mmol/1.73 m<sup>2</sup>/24 h should prompt further investigations, including ruling out secondary/enteric hyperoxaluria [69, 70]. If urine collection is not feasible (e.g., infants), a spot urinary oxalate/creatinine ratio can be performed but must be interpreted according to age, as this ratio is highest in premature newborns and decreases rapidly in the first year of life. It should be noted that children with markedly reduced GFR (<30 ml/min/1.73 m<sup>2</sup>) may have normal urinary oxalate excretion. Measurements of oxalate levels in plasma are helpful but not easily available [70]. Elevated urinary glycolic acid is characteristic of PH1 but has also been found in PH3 patients [30, 72]. Glyceric aciduria is suggestive of PH2 although not invariably found. Elevated urinary level of

4OHGlu effectively identifies patients with PH3 [32, 34]. As 4OHGlu levels were found to be markedly elevated in stored newborn screening dried-blood sample, this metabolite may be used to screen for PH3 [34].

Although there is overlap in urinary oxalate excretion between the three forms of PH, it has been suggested that patients with PH3 tend to have lower oxalate levels but higher urinary calcium and uric acid concentrations compared with those with PH1 or PH2 [62].

As mutation analysis technology advances, most patients suspected of having PH are diagnosed by genetic testing. Since PH1 accounts for >80 % of PH patients, it seems logical to test *AGXT* first, and if no mutation is found, *GRHPR* and *HOGA1* should be screened for rarer mutations. Liver biopsy with enzymatic analysis is reserved for the minority of patients with progressive kidney disease who have a phenotype suggestive of PH without pathogenic mutations (unclassified PH). Normal AGT and GRHPR activity would exclude PH1 and PH2, respectively, in these cases, and enable safer decision making about dialysis and transplantation modalities. Molecular diagnosis can provide families with the option of

premarital, prenatal, or pregestational diagnosis to ensure the birth of an unaffected child. Mutation screening should also be performed in asymptomatic siblings of affected individuals in order to consider early treatment. Genetic counseling is of utmost importance in low-resource populations with a high rate of consanguinity where RRT is not readily available and many young children may die of PH1.

## Treatment

### Supportive measures

In the early stages of the disease, conservative measures are essential to preserve renal function and prevent nephrolithiasis. Adequate hydration and oral citrate supplements are important to increase calcium oxalate solubility and prevent crystallization [73]. High fluid intake has been shown to be an effective interventional measure in stone formers [74]. The recommended daily fluid intake in patients with PH is 2–3 l/m<sup>2</sup> body surface area. In infants and toddlers, nasogastric tube or gastrostomy feeding around the clock is often required to ensure adequate water intake and consequently dilute urine. Special attention should be paid during situations of increased fluid losses (diarrhea, vomiting, fever) or limited fluid intake. Fluid administration IV is indicated in these circumstances to prevent calcium oxalate deposits. Administration of potassium citrate at 0.1–0.15 mg/kg body weight leads to alkalinization of the urine, which increases calcium oxalate solubility but also inhibits crystallization by forming complexes with calcium. With declining kidney function and the risk of hyperkalemia, sodium citrate may be given instead.

Pyridoxine (vitamin B6), a cofactor of AGT, was reported to be beneficial in about a third of patients with PH1, specifically those with p.Gly170Arg or Phe152Ile mutations [55]. Fargue [75] showed that pyridoxine increases net expression, catalytic activity, and peroxisomal import of the most common mistargeted mutant form of AGT (i.e., Gly170Arg in the background of the polymorphic minor allele) in Chinese hamster ovary (CHO) cell line. Partial effect of pyridoxine has also been observed for two other common AGT mutations in the minor allele background. The first prospective study of vitamin B6 treatment in patients with PH1 with preserved kidney function was recently published by Hoyer-Kuhn et al. [76]. The starting dose was 5 mg/kg body weight administered in increments of 5 mg/kg every 6 weeks up to a final dose of 20 mg/kg daily. Six of 12 patients showed >30 % reduction in urinary oxalate levels, but none achieved complete biochemical remission. Whether moderate reduction in urinary oxalate excretion has bearing on the rate of kidney disease progression remains to be proven. Higher doses of vitamin B6 should be

avoided, as sensory neuropathy syndromes have been described in adults taking >200 mg/kg daily [77].

The main source of oxalate is endogenous production by the liver. Strict restriction of oxalate-containing food is therefore unnecessary, especially as intestinal oxalate absorption is reduced in patients with PH [78]. Theoretically, diet restricted in meat and gelatin, sources of hydroxyproline, may benefit patients with PH3, but this approach has not been studied systematically to date.

Extracorporeal shock-wave lithotripsy (ESWL) in patients with PH is inadvisable. ESWL demonstrated suboptimal results in children with PH, as calcium oxalate stones are difficult to fragment [79]. Moreover, repeated treatments may cause parenchymal damage and shockwave-induced renal damage has been shown to promote calcium oxalate deposition in a rat model of hyperoxaluria [80].

*Oxalobacter formigenes* is an anaerobic oxalate-degrading bacterium. Uncontrolled observations have reported a reduction in urinary and plasma oxalate levels in PH patients treated orally with *O. formigenes* [81, 82]. A randomized controlled trial of orally administered *O. formigenes* in PH1 patients with preserved renal function failed to show a significant reduction in urinary oxalate excretion [83].

## Dialysis

Dialysis in children with PH and ESRD serves a dual purpose: clearance of excess fluids and uremic toxins, and removal of the oxalate load produced by the liver that can no longer be cleared via the kidneys. As oxalate clearance in peritoneal dialysis is inefficient, the modality of choice is intensified daily hemodialysis [84, 85]. A combination of daily nocturnal continuous-cycling peritoneal dialysis (CCPD) with hemodialysis has also been suggested. However, no dialysis strategy can offset oxalate generation by the liver [85]. Ultimately, PH1 patients on chronic dialysis will develop systemic oxalate deposition, with severe clinical consequences. Hemodialysis should therefore be limited to patients awaiting liver or combined liver and kidney transplantation (LKT) and/or those who are too small to be transplanted. Hemodialysis or hemofiltration is also used after transplantation to help remove heavy oxalate loads.

## Transplantation

Historically, kidney transplantation outcome in patients with PH has been poor. The 1990 report of the European Dialysis and Transplant Registry [20] reports 3-year graft survival of 17–23 % in PH patients. Graft loss was mostly due to oxalate deposition in the transplanted kidney. LKT prevents disease

recurrence in the renal allograft [86, 87], as liver transplantation with removal of the native liver corrects the metabolic defect [88]. PH patients receiving combined or sequential LKT have a better death-censored graft survival than those receiving kidney transplantation alone (KTA) [89, 90]. The 5-year survival of children with PH undergoing KTA or LKT is 14 % and 76 %, respectively [10]. These results are inferior to those reported in adults with PH who undergo kidney transplantation alone and may reflect the impact on young children of the most severe form of PH. Patients with ESRD due to PH accumulate a massive amount of oxalate in various body tissues, as described above (systemic oxalosis). Recovery of renal function hence results in immediate and significant oxaluria, even after removal of the native liver and liver transplantation. In order to prevent oxalate-induced damage to the renal allograft, Kidney Disease Improving Global Outcomes (KDIGO) guidelines [91] recommend urinary dilution with aggressive hydration and frequent dialysis for patients with delayed graft function in the immediate posttransplantation period. Harps [92] reported excellent results in nine pediatric patients who received continuous venovenous hemofiltration (CVVH) treatments immediately after combined LKT for PH1. Adequate hydration (3 l/m<sup>2</sup>/day) and anti-crystallization therapy with orally administered potassium/sodium citrate should be continued until urinary oxalate excretion returns to normal levels, a process that may take several years [93]. Orthophosphate and magnesium oxide may also help prevent calcium oxalate deposition in the transplanted kidney [91]. A recent study reported successful long-term results of KTA in four of five patients with the p.Gly170Arg mutation maintained with pharmacological doses of pyridoxine [94]. After clearing oxalate stores, urine oxalate levels were normal in most samples. These results must be interpreted cautiously because of the small number of patients and the fact that all of them were >30 years, suggesting a milder disease phenotype.

Preemptive liver transplantation has been shown to preserve renal function [95, 96]. Taking into account the fact that disease progression is highly variable, even within families, and that liver transplantation carries significant risks, it is advisable to delay the procedure until GFR declines substantially. It should be emphasized that the native liver, which is completely normal aside from the enzymatic defect, has to be removed, as opposed to auxiliary liver transplantation implemented in some patients with acute hepatic failure. Sequential transplantation with the liver first may benefit patients who do not have a suitable kidney donor. Liver transplantation, combined with intensive dialysis prior to renal transplantation, allows for a decrease in total body oxalate load. Both living and deceased donors have been used. Furthermore, sequential liver and kidney transplantation from a single living donor has been reported [97].

There is scarce data on transplantation outcome in PH2 patients [98]. Kidney transplantation alone has been performed with varying success, and graft failure due to oxalate deposition has been reported [27]. At this time, there are no reports of PH3 patients requiring RRT.

### Future strategies for treatment

Several experimental therapies for PH are under investigation. Neonatal liver-cell transplantation transiently reduced plasma oxalate levels, without populating the native liver, in an infantile oxalosis patient [99]. This procedure could serve as a bridge to liver transplantation. Its effectiveness is limited due to persistent oxalate production by native hepatocytes, which is the underlying difficulty in any potential therapeutic modality for patients with PH that does not provide adequate repopulation of the native liver tissue. The main problem remains the accumulation of substrates, mainly glyoxylate—which is oxidized to oxalate—rather than enzymatic deficiency per se.

*AGXT* gene transfer using adenoassociated virus vector in a mouse model of PH1 reduced oxaluria to normal levels and restored AGT activity in whole liver extracts [100]. Human studies have not been performed as yet.

Some AGT mutations have been shown to cause protein homeostasis defects, such as destabilization, misfolding, and mistargeting. This suggests that kinetic stabilizers and protein homeostasis modulators may be suitable pharmacological therapies in PH1 [101, 102], similar to the effect of pyridoxine, which acts not only as a coenzyme increasing catalytic activity but also as a molecular chaperone stabilizing the mutated monomers and improving the productive folding and dimerization of the enzyme [103].

Enzyme replacement therapy, which has been successful in a number of metabolic derangements, mainly lysosomal storage diseases, would be challenging in PH, as the enzyme would have to be directed into the peroxisomes of the hepatocytes and in an efficient system that would correct the vast majority of native cells. Another potential strategy for treating patients with PH is substrate reduction therapy (SRT), which is an attempt to decrease oxalate production by inhibiting synthesis of its precursor, glyoxylate. The most appealing target for treating PH1 would be to develop specific inhibitors of *HAOI*, encoding GO, which would block oxidation of glycolate to glyoxylate. Preliminary results show that *Hao1*-deficient mice are asymptomatic and that introducing this knockout gene into PH1 mice results in a marked reduction in urinary oxalate excretion [104]. This observation concurs with the report of loss-of-function mutations in the *Hao1* gene, resulting in a child with asymptomatic glycolic aciduria [105].

## Conclusion

The three types of PH demonstrate a wide clinical spectrum, ranging from relatively benign nephrolithiasis to early-onset ESRD and severe systemic oxalosis. PH should be suspected in every child with kidney stone disease or nephrocalcinosis. Early diagnosis is essential for optimal treatment. Genetic testing can offer families prenatal or pregestational diagnosis. Patient advocacy groups may assist families to cope with the disease by obtaining information and joining support groups, e.g., the Oxalosis and Hyperoxaluria Foundation (OHF, <http://www.ohf.org>); European Hyperoxaluria Consortium (Oxaleurope, <http://www.oxaleurope.org>); and the Rare Kidney Stone Consortium (RKSC, <https://rarediseasesnetwork.epi.usf.edu/RKSC/>). Hopefully, future advances will provide better therapeutic modalities for children with PH.

## Summary points

1. PH is characterized by hepatic oxalate overproduction, leading to renal deposition of calcium oxalate crystals.
2. Three distinct PH entities have been described: mutations in *AGXT* cause PH1, the most severe form, often leading to ESRD. *GRHPR* and *HOGAI* mutations cause PH2 and PH3, respectively. PH2 is rare and clinically similar to PH1. PH3 has a milder phenotype and may be underdiagnosed.
3. PH treatment is challenging. Patients with ESRD need frequent intense hemodialysis to prevent systemic oxalosis.
4. Liver transplantation prevents recurrence in transplanted kidney.
5. Genetic diagnosis and counseling can ensure the birth of unaffected children.

## Multiple-choice questions (answers are provided following the reference list)

1. Which of the following is an indication for metabolic screening of lithogenic risk factors, including hyperoxaluria?
  - a. A single renal stone in a 3-year-old boy
  - b. Four episodes of renal colic in a 25-year-old woman
  - c. Renal failure with severely hyperechoic kidneys in a 4-month-old baby
  - d. All of the above
2. What is the recommended approach to treating early-stage hyperoxaluria?
  - a. Sodium restriction and urine alkalization
  - b. Extracorporeal shockwave lithotripsy

- c. Hyperhydration and citrate supplements
  - d. Restriction of oxalate-rich food
3. A 7 year-old boy is referred after having passed two kidney stones. Two older sisters suffered a few episodes of renal colic at a young age, but repeated US scans are negative for stones or nephrocalcinosis. All three siblings have elevated urinary oxalate levels. Family history of ESRD is negative. What is the most likely diagnosis?
    - a. PH1
    - b. PH2
    - c. PH3
    - d. Secondary hyperoxaluria
  4. Which of the following is not recommended for treating a child with advanced renal failure due to PH?
    - a. Early start of peritoneal dialysis
    - b. Preemptive liver transplantation
    - c. Combined liver and kidney transplantation
    - d. Daily hemodialysis

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## Answers

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3. c
4. a