Case Report

Autopsy Findings in an Infant with Primary Hyperoxaluria (Type-1)

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ABSTRACT

Histopathological findings in oxalosis patient are limited in the literature, although it has high mortality. Oxalosis, which is defined as deposition of calcium oxalate crystals in tissues, is the final stage of various hyperoxaluric syndromes. It is often missed and is rare. The diagnosis is often delayed, since it requires special laboratory tests for establishing the diagnosis. Kidneys, blood vessel walls, and bones are the major sites for crystal deposition. We present an infant autopsy case of primary hyperoxaluria, type 1. Diagnosis was established with genetic testing. On autopsy, calcium oxalate crystals which were refringent to polarized light were found in both kidneys.

Keywords: Oxaluria

Introduction

In humans, oxalate is an end product of metabolism and its primary precursor is glyoxylate. It is either transaminated to glycine in the presence of pyridoxine or metabolized under the influence of the pyridoxine dependent, hepatic peroxisomal enzyme alanine: glyoxylate aminotransferase (AGT). The other precursors of oxalate are glycine derived from serine and ascorbic acid. Oxalate is also present in vegetables. Colon and ileum are major sites of absorption of dietary oxalate. Oxalate is freely filtered across the glomerulus and is excreted predominantly by the kidney. Its progressive accumulation in kidney leads to nephrolithiasis, nephrocalcinosis, and renal tubulointerstitial damage, with consequent decrease in glomerular filtration rate. Overproduction of oxalate combined with renal retention are increased oxalate ion concentrations in tissue fluids and consequently insoluble oxalate is deposited in tissues (Oxalosis).[1] Hyperoxaluria can be either primary or secondary. Most common form of primary hyperoxaluria (PH) is Type I hyperoxaluria (PH1), a rare autosomal recessive disorder which occurs due to deficiency of AGT enzyme and is characterized by increased urinary excretion of oxalate, recurrent urolithiasis, nephrocalcinosis, and oxalosis.[2]

Type II primary hyperoxaluria (PH2) is due to reduced activity of glyoxylate reductase (GR), an enzyme that also possesses both D-glycerate dehydrogenase (an enzyme involved in the gluconeogenetic pathway of serine metabolism) and hydroxy pyruvate reductase (HPR) activities, and leads to increased excretion of both oxalate and L glyceric acid.[1,3] Primary hyperoxaluria type 3 (PH 3) is a newly described entity and accounts for 10% of PH cases. The genetic defect is localized to the HOGA1 gene which is located on chromosome 9 and codes for the mitochondrial 4-hydroxy 2-oxoglutarate aldolase.[4] This enzyme breaks down 4-hydroxy 2-oxoglutarate into pyruvate and glyoxalate which in turn is converted into oxalate.

Secondary or acquired hyperoxaluria results from excessive ingestion of substances containing oxalic acid, such as parsley, spinach, pepper, nuts, cocoa, tea, xylitol, or ascorbic acid, or can occur from poisoning with Aspergillus fungus, methoxyflurane anesthesia, and ethylene glycol, which most often occurs in children. Diet deficient in pyridoxine and thiamine as well as intestinal diseases (malabsorption, ileal resection, non-tropical sprue, pancreatic insufficiency) leading to hyperabsorption also cause secondary hyperoxaluria.[1] It can also occur in the setting of acute and chronic renal failure. Patients present with recurrent urolithiasis, nephrocalcinosis, and progressive renal failure at a young age. Kidneys and bone being the principal organs for oxalate deposition. The disease is diagnosed on clinical findings, measurement of urine oxalate, glycolate, and renal excretion rates, and by plasma oxalate measurement.[2]

The outcome of patient depends on the time of diagnosis and the complications of systemic oxalosis. Since special studies requires to be performed, so, delay in diagnosis is common, consequently, almost one third of patients are diagnosed when they have end-stage renal disease (ESRD).[5-7] This report brings out the autopsy findings of a 6 month old boy who presented to our hospital with acute kidney injury and was diagnosed with hyperoxaluria.

Case Report

6-month-old male infant product of third-degree consanguineous marriage born at full term had presented with history of cough and coryza associated with moderate grade fever. On day 5 of admission patient had tonic clonic seizures and deranged renal functions along with generalized edema. Evaluation revealed high serum creatinine of 9.3 mg/dl, conjugated hyperbilirubinemia, bicytopenia and positive dengue serology (IgG and IgM). Ultrasound showed hepatomegaly with pseudo thickening of gall bladder with bilateral pleural effusion and ascites with bulky bilateral kidneys suggestive of acute medical renal disease. 2-D Echo revealed mild pulmonary arterial hypertension and mild tricuspid/aortic regurgitation. EEG features were suggestive of encephalopathy. Patient was managed with broad spectrum antibiotics, diuretics and oral antihypertensive. The patient continued to have high TLC (20,300/cumm) with deranged RFT and persistent hypertension. Repeat ultrasound (post renal biopsy) showed Perinephric collection around left kidney. He was oxygen dependent and received multiple transfusions throughout his stay, he had worsening metabolic acidosis and had 17 episodes of stool following which peritoneal dialysis was stopped. Serial ABG analysis revealed worsening of acidosis. Chest X ray revealed ground glass opacities suggestive of pulmonary edema. The infant developed persistent bradycardia and hypotension and had cardiac arrest.

Table 1: Comparison with other autopsy studies.

S. No	Year	Author	Age/ Sex	Organ Involvement		Other findings
				Ante mortem	**Post mortem**	
1.	2013	Falk N et al [15]	33yr/F	Skin	• Media of small, medium, and large vessels and vessels of subcutaneous skin • Liver • Pancreas • Skeletal muscle • Muscularis propria of GIT • Kidneys • Cardiac myocytes	• Bilateral pulmonary emboli and heart failure (with biventricular myocyte hypertrophy) • Centrilobular necrosis of liver with bridging fibrosis • Splenomegaly
2.	2009	B Doganavsargil et al [23]	4 yr/F	Kidney	• Kidney • Fallopian tubes • Vessels • Uterus • Heart • Thymus • Bone • Salivary glands • Ovary • Pancreas • Bladder • Liver • Tongue • Pleura • Paraortic lymph nodes	• Ventricular hypertrophy • Thrombus in • pulmonary veins
3.	1977	Haqqani MT[24]	50 YR/F	Kidney	• Vessels • Kidneys • Meninges • Brain	• Cardiomegaly • Left ventricular hypertrophy • Atheroma of left coronary artery
4.	2020	Our study	6 month/M	Kidney	• Kidney	• Ectopic liver • Choroid plexus papilloma • Hepatic steatosis
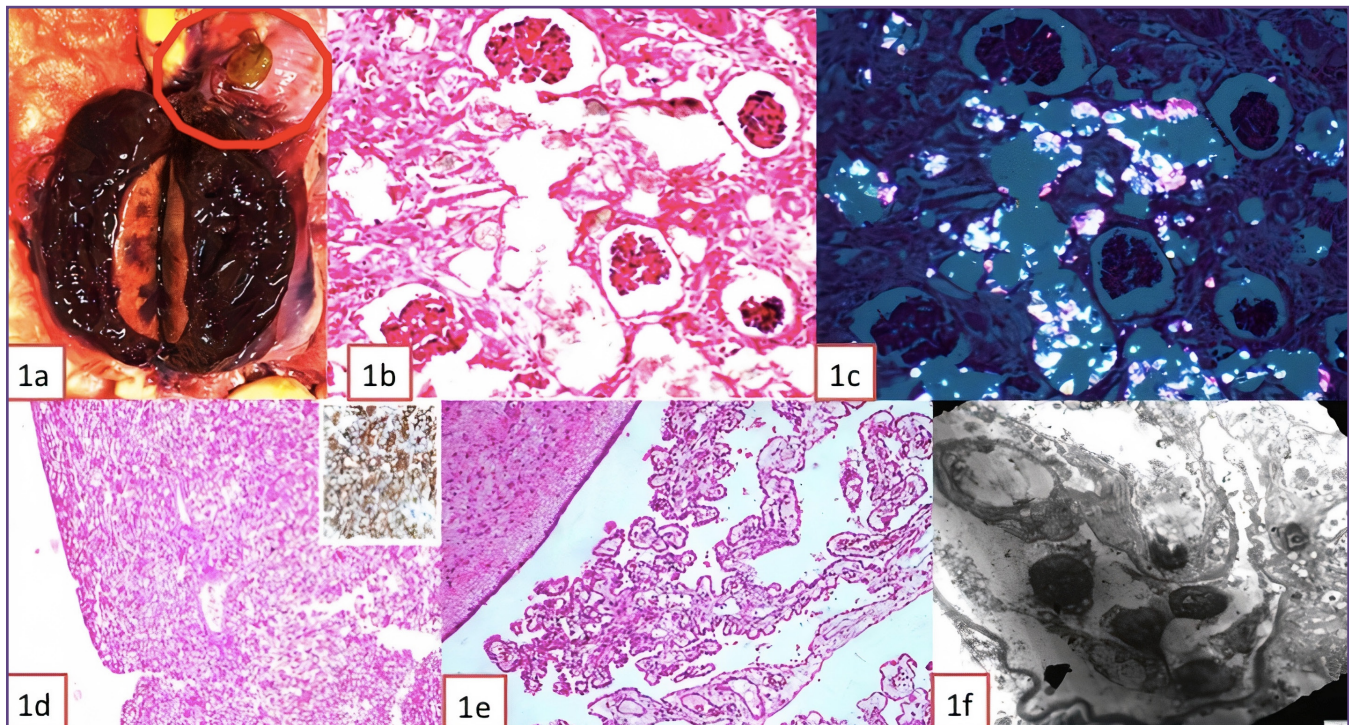
Fig. 1a:Left Kidney with para-renal hematoma and foci of ectopic liver (Encircled red), Figure 1b:Kidney showing oxalate crystals {Hematoxylin and eosin (200X)}, Figure 1c:Birefringent fan-like and polygonal crystals in same area {Hematoxylin and eosin (200X)}, Figure 1d:Ectopic liver {Hematoxylin and eosin (100X)}, Inset showing Hep Par-1 positivity, Figure 1e:HPE of ventricular growth showing Choroid plexus papilloma {Hematoxylin and eosin (100X)}, Figure 1f: Transmission electron microscope photograph of kidney showing black round to ovoid to fan shaped crystals with alternating dark and light bands(10000X).

Autopsy findings

Anthropometrical measurements were within normal range for age and sex. The thoracic cavity drained 7ml of serous pleural effusion bilaterally. Left lung was heavy and boggy. Left ventricular hypertrophy and inter ventricular septal hypertrophy was noted in heart. Spleen showed mild congestion on cut section. Kidneys were firm. Cut surface showed altered discolouration and was gritty with small yellow-white granules. A yellow-coloured tissue seen along with abdominal ligament close to left kidney (Fig No. 1a). The brain (weighed normal for age), on sectioning, showed friable, villiform growth measuring 02x01 cm in both lateral ventricles. Culture report of Peritoneal fluid, CSF and Heart blood showed *Providencia rettgeri*.

Histopathological examination of both lungs showed features of bronchopneumonia, septal inflammation, congestion, and thickening of alveolar septae. In addition, other features of diffuse alveolar damage (DAD) were seen, including marked intra-alveolar hemorrhage, type II pneumocyte hyperplasia, and hyaline membrane formation. Liver showed extensive steatosis along with normal hepatic parenchyma. No cholestasis/ inflammation

seen. Few hepatocytes showed feathery degeneration. Both kidneys showed scattered immature glomeruli with increased urinary space. The tubules were seen stuffed with variable shaped crystals (Fig No. 1b) which polarized (Fig No. 1c) and had destroyed and distorted the lining epithelium. Predominant pattern of crystals was 'fan shaped'. The interstitium was edematous with areas of early fibrosis. Small arteries and arterioles were noted in cortical parenchyma and appear unremarkable. No thrombi/segmental necrosis/crescent or collapse seen. Foci of ectopic liver seen abutting the capsule of left kidney and descending colon (Fig No.1d). Bilateral ventricles showed choroid plexus papilloma in the form of complex array of branching fibrovascular fronds covered by monolayer of uniform cuboidal epithelial cells exhibiting minimal nuclear atypia (Fig No.1e). Transmission electron microscopy of kidney revealed black round to ovoid to fan shaped crystals with alternating dark and light bands (Fig No. 1f). Next generation sequencing was performed on post mortem liver biopsy (Report came 8 weeks thereafter), and showed a pathogenic sequence c.33dup found in AGXT gene which is consistent with Autosomal recessive primary hyperoxaluria, type I.

Discussion

Most common among primary hyperoxaluria is PH1, occurring 0.11 to 0.26 per 100,000 births. The prevalence of PH1 is approximately 1-3 cases per million population. [8,9] Several methods can be used in diagnosing PH1. One is 24-hour urine collection oxalate measurement corrected for body surface area. Until genetic analysis for the detection of the AGXT gene, liver biopsy was used to measure AGT catalytic activity was essential for the diagnosis. Liver biopsy is still being used in patients with no identifiable mutation. The normal AGXT gene is a single copy gene located on chromosome 2q37.3. This protein has 392 amino acids, 11 exons with a molecular mass of 43- kDa, and is vitamin B6 (pyridoxine)-dependent. Almost 50% of mutations cosegregate with a minor allele having a 74-bp duplication in intron 1 with a proline to leucine change in position 11. The most frequent mutation, which is found in 20 to 40% of patients, is p.Gly170Arg. DNA which is obtained from crude chorionic villi or amniocytes can be used for prenatal diagnosis. Other mutations include insertions, deletions as well as missense, nonsense, and splice mutations. More than 99% of PH1 patients have been found to have mutations in the AGXT gene.[10-15]

PH1 has damaging effects on the urinary system. There is increased synthesis and urinary excretion of CaOx. CaOx, being insoluble in urine; presents as urolithiasis and/or nephrocalcinosis. Oxalate deposition in other organs, principally skin occurs when saturation point of plasma oxalate is reached and the glomerular filtration rate falls. Other involved organs include heart, central nervous system, joints, skin, soft tissues, retina.[10,11,16] In our case, we found oxalate crystals only in kidneys. With worsening renal function, oxalosis involves myocardium and conducting system and cardiac vessels.[17,18]

Differential diagnoses of crystalline nephropathies can be divided into three subgroups: Renal ischemia due to vascular calcifications or crystal embolism (type 1); intratubular and extra tubular crystalline precipitates causing tubular injury (type 2) and nephrolithiasis causing obstructive nephropathy (type 3). Except cysteine which has a larger size, crystal size is usually in the micrometer range. Cholesterol crystal embolism (Type-1) occurs due to emboli of cholesterol crystals (CCs) which arise from the rupture of an atherosclerotic plaque from aortic or other major arteries and block smaller arteries (150–200 μm in diameter). Mechanism of injury is by killing of tubular epithelial cells by activating receptor-interacting serine/threonine-protein kinase 3 (RIPK3) and mixed lineage kinase domain-like (MLKL). Sudden onset of crystal formation in type 2 is due to acute supersaturation leading to acute episodes of crystal-induced tubular cell injury along with interstitial inflammation, and impairment of renal function, i.e., acute kidney injury (AKI). Most commonly associated crystals include Calcium oxalate (Mono-and dehydrate), calcium phosphate, uric acid, myoglobin, bile pigment and monoclonal light chains. Type 3 crystal nephropathy includes:-nephrolithiasis, ureterolithiasis, or cystolithiasis which causes nephron loss, renal inflammation, and fibrosis as a result of obstructive nephropathy. Hereditary metabolic diseases which are associated with an increased production of cysteine, uric acid, or oxalate are typical examples showing pathogenesis of crystalluria and stone formation.[19]

Treatment is primarily conservative and is directed at increasing the urinary solubility of CaOx and decreasing the oxalate production. High fluid intake along with CaOx crystallization inhibitors, such as potassium/sodium citrate and phosphorus, are used to decrease calcium absorption, and consequent calciuria, and CaOx crystals. 10 to 30% of PH1 patients are sensitive to pyridoxine, which chelates oxalate precursors and is metabolized to pyridoxal phosphate, which is the main co-factor of AGT, and should be used once ESRD is reached. [10,11]

For unresponsive and patients with GFR rates between 15 and 40 mL/min per 1.73 m², liver transplantation or combined liver-kidney transplantation is the next option. Since the biochemical defect is in the liver and due to high chances of recurrence; Isolated kidney transplantation is no longer recommended.[17] New modalities under evaluation are gene therapy, chaperone treatment, liver cell transplantation and proteomic analysis of urine for management of patients with primary hyperoxaluria. [18]

Promoting earlier diagnosis through increased awareness of this genetic entity will enable treatment and the prevention of ESRD and further systemic sequelae of PH1.

In an autopsy series reported by Fayemi et al thyroid and myocardium were the most frequently involved organs other than the kidney.[21] Brain and eye involvement are well documented in the literature.

Conclusion

In conclusion, patients with PH have a high risk of early renal failure, and, if misdiagnosed (or if diagnosis is made late), of early death. Despite being a rare entity, with its early diagnosis hinging on clinical suspicion, it should be considered in patients with recurrent urolithiasis or urinary tract infections. The case is presented to highlight the unusual histological picture of an end-stage disease and rarity of disease.

Acknowledgements

Competing Interests

Nil

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