OGH Reports 2017; 6(1): 67-71
Peer Reviewed Journal in Oncology, Gastroenterology and Hepatology
www.oghreports.org | www.journalonweb.com/ogh

Case Report

An Itchy Experience - PFIC 3 Masquerading as Wilson's Disease; Learning from Mistakes

Nalini Bansal (Gupta)[1], Mukul Rastogi[2]

ABSTRACT

Introduction: Progressive familial intrahepatic cholestasis (PFIC) type 3 (PFIC3) is an autosomal recessive disorder of biliary phospholipid excretion leading to cholestasis and biliary cirrhosis. These cholestatic disorders show falsely elevated urinary copper and low ceruloplasmin together with increase in copper associated protein content on liver tissue mimicking Wilson's disease leading to diagnostic delay. **Case Report:** We report a case of a 21-year-old male who presented with complaints of gradually progressive jaundice with ascites for past 3 months. His work up revealed low serum ceruloplasmin and high 24-hour urinary copper. He was diagnosed as having Wilson's disease and living donor liver transplant was performed. Explant liver revealed prominent copper associated protein within hepatocytes and numerous Mallory Denk Bodies, findings suggestive of Wilson's etiology for cirrhosis. Patient was discharged in a stable condition. The story continued when 4 months later his 3 siblings (20 year male, 15 year old female and 11 year old female) came with complaints of itching all over the body were evaluated. Possibility of PFIC3 was kept in differential this time due to clinical scenario and liver biopsies were performed in all three. Liver biopsy in all shows prominent bile ductular reaction, increased fibrosis and hepatic copper associated protein. MDR3 immunostains was performed in these cases was negative. Index patient slides were retrieved and MDR3 stain performed showed absent staining confirming the diagnosis of PFIC 3. **Conclusion:** Cholestatic liver diseases frequently mimic Wilson's disease. Criteria for diagnosis of Wilson's does not completely holds true for cholestatic liver diseases in children and adolescents.

Key words: PFIC, Cholestasis, Pediatric, Wilson, PFIC 3.

Nalini Bansal (Gupta)[1], Mukul Rastogi[2]

[1]Fortis Escorts Heart Institute, Okhla Road, New Delhi, INDIA.
[2]Senior consultant Fortis Hospital Noida, INDIA.

Correspondence
Dr Nalini Bansal (Gupta)
MD, DNB, PDCC (Hepatopathology)
Senior Histopathologist SRL Ltd, Fortis Escort Heart Institute Okhla, New Delhi, INDIA.
E-mail: drnalinibansal@yahoo.com

History
- Submission Date: 20-08-2016;
- Review completed: 12-12-2016;
- Accepted Date: 05-01-2017

DOI : 10.5530/ogh.2017.6.2.19

Article Available online
http://www.oghreports.org

INTRODUCTION

Progressive familial intrahepatic cholestasis is group of cholestatic disorders characterized by progressive cholestasis and development of biliary cirrhosis. Type 1 and type 2 show mutation in FIC 1 and FIC 2 genes whereas type 3 is characterized by MDR3 mutation. PFIC 3 also shows high GGT in contrast to PFIC 1 and PFIC2 where GGT remains low. These disease frequently effect several family members.

Frequent mimicking of copper studies similar to Wilson's disease confuses the clinical and pathological picture. Immunohistochemistry and genetic studies only aids in confirming the diagnosis.

CASE REPORT

We report a case of a 21 year old male who was apparently alright 3 months back, when he developed weakness and jaundice. Jaundice progressed gradually. He developed ascites one month back and was treated with diuretics. Ascites was not controlled with diuretics and required multiple ascitic tapping twice since last 1 month. He also complained of pain abdomen and melena since last one month. Upper GI endoscopy showed grade III esophageal varices for which endoscopic band ligation was done twice in last 1 month. His hemoglobin was low and blood was transfused. There is no documented history of spontaneous bacterial peritonitis, hepatic encephalopathy and hepatorenal syndrome. Ultrasound abdomen showed normal size liver with splenomegaly and ascites. There was no significant past, present or family history. His height/weight/ BMI: 167 cm / 63 kg /22.58 respectively. His lab investigation revealed haemoglobin 11g/dl,total leucocyte count 6.800 thou/ul, platelet -360 thou/ul, PT/INR 43.4/5.3, fibrinogen 746 mg/dl, bilirubin 39.7 mg/dl Direct -23.95 mg/dl, SGOT 177 u/l, SGPT 123 U/L, SAP 95U/L, GGT 83 U/L, total protein 6.2, albumin 2.6. Renal function test revealed blood urea nitrogen 16, creatinine 0.8, Sodium 129, potassium 4.16, chloride 97, TSH/T3/T4-2.37 U/ml/70.86/3.51ng/dl, Cholesterol <80 mg/dl, HDL/LDL 12/10, Triglyceride 72mg/dl. His markers for HbsAg, HbcAg HCV RNA, HIV qualitative are negative. CMV IgG was positive, alpha fetoprotein level was 3.46 ng/ml, CEA 2.4 ng/ml, CA19.9 198.14 ng/ml. His ceruloplasmin was 0.14 (0.2-0.6g/l), total IgG 22.5 mg/dl and ANA was negative. 24 hours urinary copper was 1732.8 (<60ug/24 hrs). Other investigations were

Cite this article: Bansal N(G), Rastogi M. An Itchy Experience - PFIC 3 Masquerading as Wilson's Disease; Learning from Mistakes. OGH Reports. 2017;6(1):67-71.

all negative. Ultrasound doppler revealed hepatosplenomegaly with chronic liver disease with multiple collaterals seen in perisplenic region and ligamentum teres. On opthalmologic examination no KF ring was seen. Patient was diagnosed as having acute on chronic liver failure and he underwent living donar liver transplant and received right lobe graft without middle hepatic vein. Gross of explant liver measuring 19 x 17 x 7 cms, outer surface was brownish nodular margins were irregular. Serial slicing revealed multiple brownish nodules measuring 0.3-0.5 cms in diameter. Microscopic examination revealed multiple nodules intervened by fibrous septae. Hepatocytes show presence of prominent Mallory Denk Bodies. Orcein stain done show increased copper associated protein within hepatocytes. Case was diagnosed as Cirrhosis likely etiology as Wilson's Disease. (Figure 1).

Patient was discharged in a stable condition. 4 months after surgery he returned with deranged LFT along with his 1 brother and 2 sisters. His brother complaint of itching for past 3 months. His 2nd sister 15 year old complaint of generalized itching for last 2 months and his 3rd sister 11 year old had an episode of epistaxis 10 days back. (Table 1).

All of them underwent liver biopsy for suspected etiology of chronic liver disease. At this point of time differential of PFIC was kept. Liver biopsy tissue showed similar histological findings with prominent bile ductular reaction. Biopsy tissue in all showed increased copper associated protein on orcein stain. (Figure 2).

Differential of Progressive familial intrahepatic cholestasis was thought and immunohistochemistry for MDR 3 gene was perfomed on three biopsy tissue. It turned out to be negative. Explant tissue slides were retrieved and MDR3 was performed on those slides also turned out to be negative. Patient was adviced genetic testing , however family could not afford if due to financial constraints. In the light of immunohistochemical findings all cases were then finally diagnosed as progressive familial intrahepatic cholestasis type 3. All patients were adviced UDCA and responded well to treatment.

DISCUSSION

Etiological diagnosis of cirrhosis developing during childhood and in young patients always poses a diagnostic dilemma. Overlapping clinical and histological features for many diseases further complicates the scenario. In current era of genetic testing, etiological diagnosis should be supplemented with molecular and genetic testing for reaching to final diagnosis.

In our patient, biochemical testing led to an erroneous diagnosis of Wilson disease; due to low ceruloplasmin levels and elevated urinary copper much above typical diagnostic levels. Utilizing the guidelines laid down by European society at the 8th International Meeting on Wilson's disease in Leipzig in 2001 that includes graded values for clinical, biochemical and molecular testing. Our patient met criteria for a diagnosis of WD based on elevated urine copper excretion (2), low ceruloplasmin (1) and copper granules (1), giving a total score of 4 (a score above 3 is considered diagnostic for WD).[1] PFIC3 is an autosomal recessive disorder results from mutations in the *ABCB4* gene located on chromosome 7

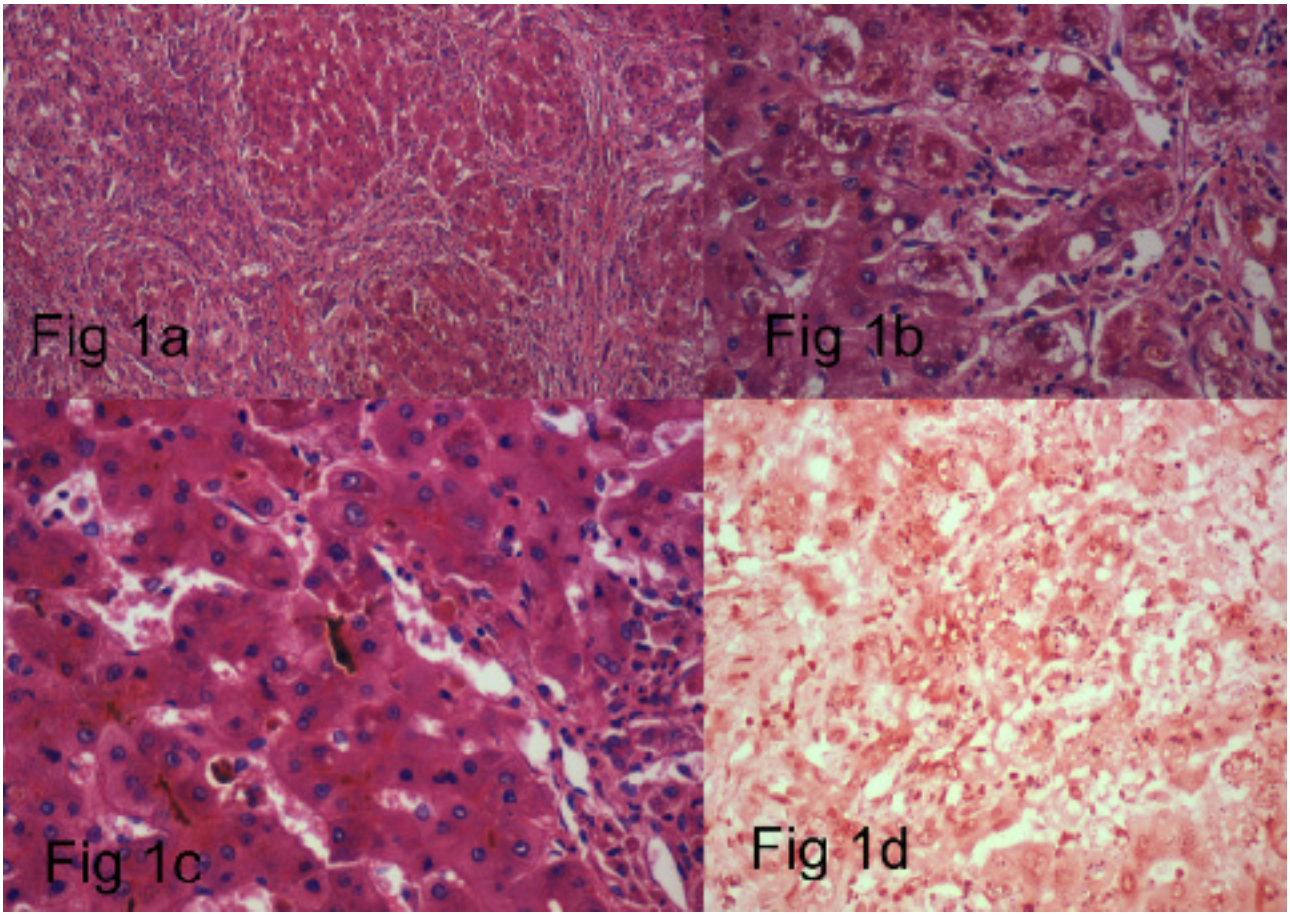

Figure 1a: Showing cirrhotic nodules (H&EX4) Fig 1b-Showing MDB (H&EX40) Fig 1c-Showing cellular and canalicular cholestasis (H&EX20) Fig 1d-Orcein stain showing copper associated proteins within hepatocytes.

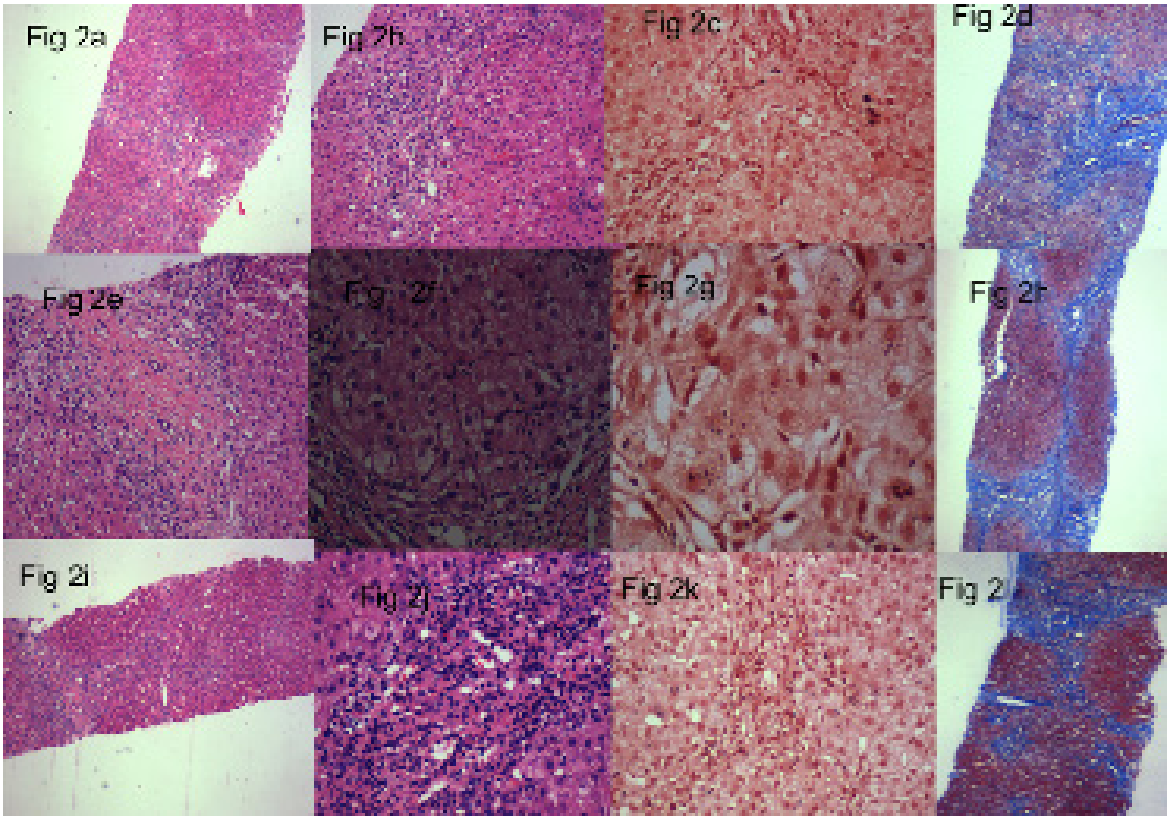

Figure 2: Figure 2 a, b, c and d-Biopsy tissue of youngest sibling 11 year old with expanded tracts (2a&b), copper associated protein on Orcein stain (2 c) and increased periportal fibrosis on MT stain (2d) Figure 2 e, f, g and h-Biopsy tissue of IInd sibling 15 year old with expanded tracts, bile ductular reaction (2 e & f), copper associated protein on Orcein stain (2g) and bridging fibrosis on MT stain (2 h) Figure 2 i, j, k and h-Biopsy tissue of 3rd sibling 20 year old with expanded tracts, portal inflammation and ductular reaction (2 i & j), copper associated protein on Orcein stain (2 k) and cirrhosis on MT stain (2 l).

encoding for the Class 3 Multidrug Resistance Protein (MDR3,[2] This gene product is predominantly expressed in the hepatocellular canalicular membrane and is responsible for phospholipid transport into bile. MDR3 is a transporter of phosphatydylcholine in bile. In mutation in MDR 3 there is absence of phosphatidylcholine in bile hence bile salts exert full detergent action on biliary epithelial cells resulting in destruction of these cell-releasing GGT in circulation. Further, the altered ratio of phospholipids and bile salts leads to increased lithogenicity of bile, resulting in crystallization of cholesterol with cholelithiasis. Injury to the biliary epithelium results in cholestasis, biliary cirrhosis and hepatocellular failure. Wilson's disease in contrast is caused by mutation in ATP 7B gene on chromosome 13q 14.3. The gene codes for a copper transporting protein ATPase 2. This gene aids in binding of copper with alpha globulin to form cerruloplasmin. In absence of this gene low ceruloplasmin is formed, and remaining copper accumulates within liver.[3,4] Literature search revealed 3 case reports of PFIC being misdiagnosed initially as Wilson's disease due to falsely elevated copper levels (Table 2).[5-7] A series of 4 pediatric patients from India also highlighted findings of falsely elevated urinary copper and hepatic copper content in patients with cholestatic liver disease.[8] The elevated levels fulfills the criteria laid down by EASL and AASLD for diagnosis of Wilson's disease. Histology also adds little to differentiate the etiological differentials in these cases. As was seen in our case where explant liver showed high levels of copper associated protein on orcein stain and numerous Mallory Denk Bodies. Testing for molecular and genetic markers is of utmost importance for rendering a final etiological diagnosis thereby guiding future therapy.

Patients of PFIC 3 present in slightly older age group than PFIC 1 and 2. The age group of presentation overlaps with that of Wilson' disease (5-15 years). Clinically patients of PFIC presents in first decade of life with itching all over the body due to cholestasis and abdominal pain. Later they show features of cirrhosis like portal hypertension and splenomegaly. Whereas those of wilson's show features of liver disease, KF ring and neuropsychiatric manifestation. History of itching in siblings should raise suspicion of cholestatic disease over Wilson's. In our patient there were no neurological symptoms.[9,10]

Laboratory investigation in PFIC 3 show a high GGT in contrast to PFIC 1 and 2. SGOT, SGPT and alkaline phospahatase can be mildly deranged. Serm bile acid levels are usually elevated. Wilson's disease show AST: ALT >2.2 and alkaline phosphatase: bilirubin < 4 especially in acute liver failure due to wilson's. Diagnostic value of serum ceruloplasmin and 24 hour urinary copper for ruling out wilson's needs to be redefined as in our patient the values fitted the guidelines for diagnosing wilson's . Copper overload has been described as a secondary manifestation of the chronic cholestasis with mechanisms including disturbed copper transport via bile due to collapse of ducts and also due to defect in hepatic lysosomal function which is the main source of biliary copper. These may thus mimick as wilson's disease and confuse the clinical picture. Definite utility of ceruloplasmin as a screening test for wilson's is uncertain. Being an acute phase phase reactant levels can be falsely elevated (estrogen use, inflammation etc) or decreased (low serum albumin as in end stage liver disease, protein loss etc) in a number of other conditions. Penicillamine D challenge test is a useful surrogate in such cases wherein ceruloplasmin level should decrease on giving penicillamine and lead to correct diagnosis.

Liver histology in PFIC 3 cases show portal inflammation, bile ductular reaction and subsequent fibrosis. Wilson's show overlapping features with PFIC 3. Presence of prominent Mallory Denk bodies and high cop-

TABLE 1

	Our patients			
Age of pt	**21 Year M**	**20 year M**	**15 year F**	**11 year F**
Bil T/D mgdl	39.7/23.9	0.57/0.24	0.73/0.32	0.08/0.25
SGOT IU/L	177	241	137	171
SGPT IU/L	123	357	155	170
SAP IU/L	95	256	348	332
SGGTP IU/L	83	355	218	121
Albumin mg/dl	2.6	3.7	6.9	8.7
Ceruloplasmin (18-46 g/dl)	0.14	0.57	1.9	2.1
24 hour urine copper (3-50 ug/ml)	1732	164.6	50.29	26.2
Hepatic Cu content ug/g	ND	ND	ND	ND
Serum copper (90-190 ug/dl)	ND	ND	ND	ND
Liver Biopsy	Cirrhosis with MDB in hepatocytes, copper associated protein	Bile ductular reaction with bridging fibrosis	Bile ductular reaction with bridging fibrosis	Bile ductular reaction with bridging fibrosis

TABLE 2

	Our patients				Boga *et al*	Ramraj *et al*		Shneider *et al*
Age of pt	21 Year M	20 year M	15 year F	11 year F	15 year	11 year	6 year	2 year F
Bil T/D mgdl	39.7/23.9	0.57/0.24	0.73/0.32	0.08/0.25	2.8/1.6	0	0.6	
SGOT IU/L	177	241	137	171	141	85	192	
SGPT IU/L	123	357	155	170	114	80	139	
SAP IU/L	95	256	348	332	596	774	397	
SGGTP IU/L	83	355	218	121	734	293	608	
Albumin mg/dl	2.6	3.7	6.9	8.7	4.1	2.6	4.5	
Ceruloplasmin (18-46 g/dl)	0.14	0.57	1.9	2.1	38	26.7	44.6	
24 hour urine copper (3-50 ug/ml)	1732	164.6	50.29	26.2	342	125	66	
Hepatic Cu content ug/g	ND	ND	ND	ND	1.47	860	863	248
Serum copper (90-190 ug/dl)	ND	ND	ND	ND		84		
Liver Biopsy	Cirrhosis with MDB in hepatocytes, copper associated protein	Bile ductular reaction with bridging fibrosis	Bile ductular reaction with bridging fibrosis	Bile ductular reaction with bridging fibrosis	portal inflammmation, ductular reaction, Bridging fibrosis ? cirrhosis	Balloning degeneration of hepatocytes, portal inflammation, micronoular cirrhosis	hepatic inflammation, bridging fibrosis, detectable copper histochemically	

per content within hepatocytes could be identified in both disease as was seen in our case. Other subtle features of Wilson's include mild periportal steatosis and presence of glycogenated nuclei.[11-13] Confirmation through immunohistochemical markers for MDR3 or genetic testing done could help in making correct diagnosis.

IHC for MDR3 show a variable expression and positive staining should not be taken as evidence against a diagnosis of PFIC3 disease as gene could also be present and non functional.[14,15]

The diagnosis of PFIC3 is confirmed by molecular genetic analysis of the ABCB 4 gene and for Wilson's by genetic testing for ATP 7B 1 gene.

The primary medical therapies utilized in PFIC include ursode- oxycholic acid, phenobarbital and rifampicin.

Decrease in serum bile acid concentration on UDCA therapy has been reported by Jacquemin *et al.*[2] and could be used as a guide to therapy. Partial bile diversion by a cholecystojejunal cutaneous conduit and/or ileal diversion have also been tried. Liver transplantation has been offered in patients who present in later stages of cirrhosis.

CONCLUSION

The current case report highlighted several points-Chronic cholestatic pediatric disorders presenting late in the disease course show mimicking clinical, biochemical and histological features with wilson's disease. The standard criteria for diagnosis of Wilson's needs to be revised. Penicillamine challenge test offers a good hope in diagnosing these cases in resource constraint setting.

Future research are indicated in cholestatic cases for giving new insight to the pathogenesis of MDR3 deficiency leading to defective copper hepatic excretion.

ACKNOWLEDGEMENT

Nil

CONFLICT OF INTEREST

Nil

ABBREVIATION USED

PFIC: Progressive familial intrahepatic cholestasis; MDR: Multidrug resistant protein; UDCA: Ursodeoxycholic acid; WD: Wilson disease.

REFERENCES

1. European Association for Study of Liver. EASL Clinical Practice Guidelines: Wilson's disease. J Hepatol. 2012;56:671-85. https:/doi.org/10.1016/j.jhep.2011.11.007 PMid:22340672.
2. Jacquemin E. Progressive familial intrahepatic cholestasis. Genetic basis and treatment. Clin Liver Dis. 2000;4(1):753-63. https:/doi.org/10.1016/S1089-3261(05)70139-2.
3. Jacquemin E. Role of multidrug resistance 3 deficiency in pediatric and adult liver disease: one gene for three diseases. Semin Liver Dis. 2001;21:551-62. https:/doi.org/10.1055/s-2001-19033 PMid:11745043.
4. Baussan C, Cresteil D, Gonzales E, Raynaud N, Dumont M, Bernard O, *et al.* Genetic cholestatic liver diseases: the example of progressive familial intrahepatic cholestasis and related disorders. Acta Gastroenterol Belg. 2004; 67:179–183 PMid:15285575.
5. Shneider BL. ABCB4 disease presenting with cirrhosis and copper overload-potential confusion with Wilson disease. J Clin Exp Hepatol. 2011;1(2):115-7. https:/doi.org/10.1016/S0973-6883(11)60131-X.
6. Ramraj R, Finegold MJ, Karpen SJ. Progressive familial intrahepatic cholestasis type 3: overlapping presentation with Wilson disease. Clin Pediatr (Phila). 2012;51:689-91. https:/doi.org/10.1177/0009922812451076 PMid:22669981 PMCid:PMC3678905.
7. Boga S, Jain D, Schilsky ML. Presentation of Progressive Familial Intrahepatic Cholestasis Type 3 Mimicking Wilson Disease: Molecular Genetic Diagnosis and Response to Treatment. Pediatric Gastroenterology. Hepatology and Nutrition. 2015;18(3):202-8. https:/doi.org/10.5223/pghn.2015.18.3.202 PMid:26473142 PMCid:PMC4600706.
8. Sood V, Rawat D, Khanna R, Alam S. Cholestatic liver disease masquerading as Wilson disease. Indian Journal of Gastroenterology. 2015:34(2);174-7. https:/doi.org/10.1007/s12664-015-0552-9 PMid:25937485.
9. Merle U, Schaefer M, Ferenci P, Stremmel W. "Clinical presentation, diagnosis and long-term outcome of Wilson's disease: a cohort study". Gut. 2007;56(1):115-20. https:/doi.org/10.1136/gut.2005.087262 PMid:16709660 PMCid:PMC1856673.
10. Brewer GJ, Askari FK. "Wilson's disease: clinical management and therapy". Journal of Hepatology. 2005;42(Suppl 1):13-21. https:/doi.org/10.1016/j.jhep.2004.11.013 PMid:15777568.
11. Ludwig J, Moyer TP, Rakela J. The liver biopsy diagnosis of Wilson's disease. Methods in pathology. Am J Clin Pathol. 1994;102(4):443-6. https:/doi.org/10.1093/ajcp/102.4.443 PMid:7942601.
12. Langner C, Denk H. Wilson disease. Virchows Arch. 2004;445:111-8. https:/doi.org/10.1007/s00428-004-1047-8 PMid:15205951.
13. Stromeyer FW, Ishak KG. Histology of the liver in Wilson's disease: a study of 34 cases. Am J Clin Pathol. 1980;73:12-24. https:/doi.org/10.1093/ajcp/73.1.12.
14. Wendum D, Barbu V, Rosmorduc O, Arrivé L, Fléjou JF, Poupon R. Aspects of liver pathology in adult patients with MDR3/ABCB4 gene mutations. Virchows Arch. 2012;460(3):291-8. https:/doi.org/10.1007/s00428-012-1202-6 PMid:22331132.
15. Sannier A, Ganne N, Tepper M, Ziol M. MDR3 immunostaining on frozen liver biopsy samples is not a sensitive diagnostic tool for the detection of heterozygous MDR3/ABCB4 gene mutations. Virchows Arch. 2012;460(5):535-7. https:/doi.org/10.1007/s00428-012-1231-1 PMid:22527017.

Cite this article: Bansal N(G), Rastogi M. An Itchy Experience - PFIC 3 Masquerading as Wilson's Disease; Learning from Mistakes. OGH Reports. 2017;6(1):67-71.