Impaired Expression of ATP-binding cassette transporter G2 and Liver Damage in Erythropoietic Protoporphyrinia

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Erythropoietic protoporphyria (EPP) is a hereditary disease caused by genetic alterations of ferrochelatase (FECH).¹ Symptoms of EPP are characterized by photosensitivity and liver damage that could be a life-threatening complication.² Incidence of liver damage is higher in patients carrying a nonsense mutation/deletion in FECH than in those with a missense mutation, indicating that FECH activity may relate to severity of liver damage. However, FECH mutations are not always associated with concomitant liver damage. Here, we report on a case study of 2 brothers with identical FECH mutations and similar levels of whole blood protoporphyrin (PP), but showing different courses of liver damage.

The older brother (a 22-year-old male) developed photosensitivity in childhood and was diagnosed with EPP because he carried two predisposing alleles of FECH, an exon-9 deletion and IVS3-48 T>C. His whole blood PP levels fluctuated between 1,500 and 2,500 µg/dL (normal, 30-86 µg/dL), and he exhibited concomitant elevation of serum aminotransferase level (alanine aminotransferase [ALT]: 50-250 IU/L; normal, <35 IU/L). On May 27, 2013, he developed severe hepatic dysfunction: ALT, 312 IU/L; alkaline phosphatase, 690 U/L (normal, 85-300 U/L); and total bilirubin (T-bil), 7 mg/dL (normal, 0.2-1.0 mg/dL). Marked elevation of whole blood PP was also observed (4,883 µg/dL), but we could not detect any data suggesting other causes of liver disease; liver dysfunction associated with EPP was strongly suspected (Fig. 1A). On day 10 of hospitalization, a liver biopsy was performed; marked deposition of PP in hepatocytes and hepatocyte necrosis with inflammatory cell infiltration at the perportal area were observed. Because ATP-binding cassette transporter G2 (ABCG2; a canalicular transporter) is responsible for excretion of PP into bile, we performed an immunohistochemical assay to evaluate ABCG2. The results revealed decreased ABCG2 expression in hepatocytes with PP deposition (Fig. 2A). Because T-bil levels further increased and whole blood PP remained high, plasmapheresis was performed. ALT, T-bil, and PP levels improved to 71 U/L, 3.7 mg/dL, and 2,194 µg/dL, respectively (Fig. 1A).

The younger brother did not have a history of liver dysfunction (Fig. 1B). Deep sequencing of his DNA revealed that he had an identical predisposing alleles to those of the older brother. Although his whole blood PP level was also high (1,500-2,500 µg/dL), his liver biopsy tissues showed that PP was mainly observed in bile canaliculi, but not within hepatocytes. More interestingly, in contrast to liver tissue from the older brother, ABCG2 expression was maintained on the cell membrane of hepatocytes in the younger brother (Fig. 2B). Whole exome sequencing revealed no specific ABCG2 mutation in both patients. However, ABCG2 messenger RNA level was markedly lower in the older brother than in the younger brother.

Both brothers had the same FECH mutations; however, they exhibited different degrees of liver damage. Interestingly, both exhibited high levels of whole blood PP; however, localization of PP in hepatocytes differed between the two brothers. Therefore, we sequenced the whole exomes of the two brothers, including genes involved in porphyrin metabolism. We also compared expression levels of ABCG2, which plays a role in excretion of PP.³ Although their genetic background with respect to FECH was identical, ABCG2 expression was much lower in the older brother than in the younger brother.

Abbreviations: ALT, alanine aminotransferase; EPP, erythropoietic protoporphyria; FECH, ferrochelatase; ABCG2, ATP-binding cassette transporter G2; PP, protoporphyrin; T-bil, total bilirubin.

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lower in the older brother than in the younger brother. Liver biopsy of the older brother showed marked porphyrin deposition in hepatocytes, along with bile thrombi. Because ABCG2 functions as a transporter that excretes PP from hepatocytes into bile canaliculi, it is conceivable that accumulation of porphyrin in hepatocytes is a consequence of decreased ABCG2 expression, which, consequently, leads to severe liver damage.

References

Fig. 1. Clinical course of EPP in the 2 patients. (A) Clinical course in the older brother. (B) Clinical course in the younger brother. Abbreviations: AST, aspartate aminotransferase; RBC, red blood cells.

Fig. 2. Immunofluorescence staining of ABCG2 and localization of PP. (A) Liver biopsy from the older brother. Red arrows indicate deposition of PP in hepatocytes and bile canaliculi. ABCG2 expression was barely detectable on hepatocyte membranes. (B) Liver biopsy from the younger brother. PP was observed in bile canaliculi of hepatocytes. Note that ABCG2 staining was maintained (green fluorescence) at the hepatocyte membrane.