



Intrahepatic Cholestasis of Pregnancy: New Diagnostic Insights

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Intrahepatic Cholestasis of Pregnancy (ICP) is the most common pregnancy-related liver disease and is characterized by onset of pruritus and elevated serum transaminases and bile acids (BA) in the third trimester of pregnancy. Symptoms and abnormal liver tests resolve following delivery but frequently recur in subsequent pregnancies. Genetic defects of canalicular transporters have been associated with the development of ICP, which may be further influenced by gestational hormones. In particular, the syndrome has been associated in some patients with heterozygosity for mutations in the genes encoding familial intrahepatic cholestasis protein-1 FIC1 (ICP-type 1), bile salt excretory protein (BSEP) (ICP-type 2), and multidrug resistance protein 3 (MDR3) (ICP-type 3).¹ Patients are classified as mild (BA 10-40) or severe (BA > 40) which has implications with respect to the risk of perinatal complications. ICP has been associated with higher rates of premature delivery, meconium staining of the amniotic fluid, respiratory distress, low Apgar scores, and stillbirth.² These complications are increased in women with severe ICP (BA > 40) while women with mild ICP have similar rates compared to women without ICP.^{3,4} While the mechanisms resulting in fetal complications remain incompletely understood, studies suggest that elevated bile acid levels in the circulation play a significant role. Animal studies have found that infusion of cholic acid caused both stimulation of gut motility resulting in meconium staining of amniotic fluid and myometrial contractility resulting in pre-term labor.⁵ In two recently published studies from China, additional risk factors for fetal complications were found to be early-onset ICP and concomitant hepatitis B infection.^{6,7} Interestingly, women who have an episode of ICP, despite post-delivery resolution of symptoms, are at increased risk for development of various hepatobiliary disorders later in life and require appropriate long-term follow up.⁸

In this issue, Chen, *et al.* describe the characteristics of ICP in the Chinese population. They indicate that, unlike findings in other populations, assay of serum total bile acid levels in their patients was not a reliable indicator of ICP. The present study was designed to identify novel serum biomarkers in these women. The authors followed a cohort of 98 patients diagnosed with ICP based on the presence of pruritus, elevated serum transaminases and total serum BA > 14 and a control group of 50 healthy pregnant women. The patients with ICP were diagnosed as having mild (N = 50) or severe (N = 48) disease based on the degree of elevation of serum levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST), total bilirubin (TB) and cholyglycine (CG). While serum total bile acids levels have been found to be useful in the diagnosis of ICP in other populations, the authors of this study were concerned that they did not significantly differ between healthy pregnant women and women with mild and severe ICP (Figure 1 in the manuscript). As seen in the figure, there was very high variability in these levels within these groups, leading to the concern that consideration of serum bile acids (BA) alone in these groups of pregnant Chinese women had low diagnostic sensitivity to differentiate those with ICP from those with normal pregnancies in the third trimester. The impact of population-specific differences in establishing normal ranges of serum total bile acid levels especially as it relates to diagnosis of ICP are clear.⁹ Prior studies, including a study based in China, evaluating the diagnostic criteria in ICP have found use of serum total bile acid levels superior to ALT and AST, which frequently follow bile acid elevations in the circulation and may represent hepatocellular damage due to cholestasis.^{6,10} Cholyglycine (a serum bile acid) and serum bilirubin levels, in addition to ALT and AST, did correlate significantly with disease severity in this population as shown in table 2 of this report.

Classification of disease severity has helped to determine the risk of fetal complications in patients with ICP. In this study, increased incidence of premature delivery and decreased newborn birth weight were found in patients with both mild and severe disease as compared to births in healthy pregnant women, with the highest rates occurring in those patients with severe disease. Serum total bile acid levels were not reported in regards to fetal complication rate in this study, which is unfortunate as elevated serum BA are not only a marker of disease but also likely play an important role in its pathogenesis. In addition, ursodeoxycholic acid is the standard treatment of ICP and serum BA are monitored throughout therapy (in addition to liver enzymes) to determine successful treatment. The authors do not discuss treatment strategies in the management of their patients with mild or severe ICP.

In the search for a more sensitive biomarker of ICP in the Chinese population, the authors found that elevated serum levels of matrix metalloproteinases MMP-2 and MMP-9 correlated with the degree of severity of ICP. MMP-2 and MMP-9 are secreted by the placenta and have been implicated in various disorders of pregnancy. Prior studies have shown that alterations of MMP-2 and MMP-9 levels during pregnancy may result in adverse maternal and fetal outcomes. The current study has as its basis a recent investigation by these authors in which they found that MMP-2 and MMP-9 were up-regulated in a rat model of cholestasis of pregnancy, with an increase of MMP levels in both the serum and the liver.¹¹ In addition to diseases of pregnancy, MMP-2 and MMP-9 also play a role in remodeling within the liver and have been studied as potential markers of hepatic fibrosis and/or cirrhosis. Elevated levels of these MMPs have been found in a number of liver disorders including hepatitis C, cirrhosis and hepatocellular carcinoma.¹²⁻¹⁴

In this present study, elevated serum levels of MMP-2 and MMP-9 were quantified and shown to correlate with the degree of elevation of liver function tests including serum bilirubin, ALT, and AST. It should be noted, although not of major concern regarding this report, that prior studies have shown that assay of MMPs in plasma is more accurate than serum determinations as performed in this study, as MMPs can be variably released from leukocytes and platelets during the preparation of serum following blood clotting in collection tubes.¹⁵ As shown in figure 2, increased levels of MMP-2 and MMP-9 were associated with worsening disease severity. The patients within this study were reportedly without chronic or pre-existing liver disease, including viral hepatitis, suggesting that these elevations are related to ICP. Based on this association, MMP-2 and MMP-9 appear to provide a sensitive marker for liver disease. Their specificity for ICP in this Chinese population will require further study as will de-

termination as to whether there is a direct association of MMP levels with fetal complication rate. It would have been interesting to compare MMP levels to total BA and cholyglycine in order to determine if MMPs are superior to these standard indicators of disease severity and fetal complication rate. However, despite this study's limitations, the finding of elevated MMP-2 and MMP-9 in ICP is novel and intriguing and suggests that further investigation to examine these questions may provide important new insights for diagnosis and management of these patients in the future.

REFERENCES

1. Dixon PH, Williamson C. The pathophysiology of intrahepatic cholestasis of pregnancy. *Clin Res Hepatol Gastroenterol* 2016; 40: 141-53.
2. Keitel V, Dröge C, Stepanow S, Fehm T, Mayatepek E, Köhrer K, Häussinger D. Intrahepatic cholestasis of pregnancy (ICP): case report and review of the literature. *Z Gastroenterol* 2016; 54: 1327-33.
3. Glantz A, Marschall HU, Mattsson LA. Intrahepatic cholestasis of pregnancy: Relationships between bile acid levels and fetal complication rates. *Hepatology* 2004; 40: 467-74.
4. Geenes V, Chappell LC, Seed PT, Steer PJ, Knight M, Williamson C. Association of severe intrahepatic cholestasis of pregnancy with adverse pregnancy outcomes: a prospective population-based case-control study. *Hepatology* 2014; 59: 1482-91.
5. Williamson C, Geenes V. Intrahepatic cholestasis of pregnancy. *Obstet Gynecol* 2014; 124: 120-33.
6. Jin J, Pan SL, Huang LP, Yu YH, Zhong M, Zhang GW. Risk factors for adverse fetal outcomes among women with early- versus late-onset intrahepatic cholestasis of pregnancy. *Int J Gynaecol Obstet* 2015; 128: 236-40.
7. Hu Y, Ding YL, Yu L. The impact of intrahepatic cholestasis of pregnancy with hepatitis B virus infection on perinatal outcomes. *Ther Clin Risk Manag* 2014; 10: 381-5.
8. Ropponen A, Sund R, Riikinen S, Ylikorkala O, Aittomaki K. Intrahepatic cholestasis of pregnancy as an indicator of liver and biliary diseases: a population-based study. *Hepatology* 2006; 43: 723-8.
9. Pathak B, Sheibani L, Lee RH. Cholestasis of pregnancy. *Obstet Gynecol Clin North Am* 2010; 37: 269-82.
10. Walker IA, Nelson-Piercy C, Williamson C. Role of bile acid measurement in pregnancy. *Ann Clin Biochem* 2002; 39: 105-13.
11. Chen Z, Hu L, Lu M, Shen Z. Resveratrol reduces matrix metalloproteinases and alleviates intrahepatic cholestasis of pregnancy in rats. *Can J Physiol Pharmacol* 2016; 94: 402-7.
12. Abdel-Latif MS. Plasma Levels of Matrix Metalloproteinase (MMP)-2, MMP-9 and Tumor Necrosis Factor-alpha in Chronic Hepatitis C Virus Patients. *Open Microbiol J* 2015; 9: 136-40.
13. Huang CY, Tseng KC, Lin MN, Tsai JP, Su CC. Plasma levels of matrix metalloproteinase-2 and -9 in male and female patients with cirrhosis of different aetiologies. *J Clin Pathol* 2015; 68: 917-22.
14. Daniele A, Abbate I, Oakley C, Casamassima P, Savino E, Casamassima A, Sciortino G, et al. Clinical and prognostic role of matrix metalloproteinase-2, -9 and their inhibitors in

breast cancer and liver diseases: A review. *Int J Biochem Cell Biol* 2016; 77: 91-101.

15. Thraillkill K, Cockrell G, Simpson P, Moreau C, Fowlkes J, Bunn RC. Physiological matrix metalloproteinase (MMP) concentrations: comparison of serum and plasma specimens. *Clin Chem Lab Med* 2006; 44: 503-4.

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