Neonatal Hemochromatosis

Amy G. Feldman, Peter F. Whittington

Department of Pediatrics, Feinberg Medical School of Northwestern University, Ann and Robert H. Lurie Children's Hospital of Chicago, 225 East Chicago Avenue, Box 57, Chicago, IL 60611-2605, United States

Neonatal hemochromatosis is a clinical condition in which severe liver disease in the newborn is accompanied by extrahepatic siderosis. Gestational alloimmune liver disease (GALD) has been established as the cause of fetal liver injury resulting in nearly all cases of NH. In GALD, a woman is exposed to a fetal antigen that she does not recognize as "self" and subsequently begins to produce IgG antibodies that are directed against fetal hepatocytes. These antibodies bind to fetal liver antigen and activate the terminal complement cascade resulting in hepatocyte injury and death. GALD can cause congenital cirrhosis or acute liver failure with and without iron overload and siderosis. Practitioners should consider GALD in cases of fetal demise, stillbirth, and neonatal acute liver failure. Identification of infants with GALD is important as treatment is available and effective for subsequent pregnancies. (J CLIN EXP HEPATOL 2013;3:313–320)

Neonatal hemochromatosis (NH) is a clinical condition in which severe liver disease in the newborn is accompanied by extrahepatic siderosis in the distribution seen with hereditary hemochromatosis. Because it was observed to occur in siblings NH was originally classified as part of the family of hereditary hemochromatosis disorders (OMIM 231100). However, clinical evidence accrued over several decades suggested that NH is not a disease per se, but is the consequence of fetal liver injury. Thus, search for an inherited cause of fetal liver disease capable of producing the NH phenotype ensued. In 2010 it was discovered that the liver in cases of NH showed evidence of complement-mediated hepatocyte injury, which under the circumstances must be initiated by IgG antibody binding to fetal hepatocytes. This finding led to the deduction that gestational alloimmune liver disease (GALD) is the cause of fetal liver injury leading to nearly all cases of NH¹ and to the conclusion that while NH is both congenital and familial, it is not heritable. GALD and NH are not synonymous: GALD is a disease or disease process causing severe fetal liver injury, whereas NH is the phenotypic expression in the neonate of severe liver injury initiated in utero, most commonly by GALD. Moreover, GALD can cause liver disease that is not accompanied by

iron overload, including acute liver failure in the fetus and neonate. Therefore, GALD has emerged as a spectrum of diseases with NH as the common but not exclusive phenotype. The discovery of the alloimmune etiology of NH has impacted approaches to its diagnosis, treatment, and prevention.

ETIOLOGY OF NEONATAL HEMOCHROMATOSIS

Early on, NH was described as a hereditary disorder of iron metabolism.² Infants with NH were found to be cirrhotic, raising the suspicion for intrauterine liver injury. However, until recently the cause of such injury remained a mystery. Because it was observed to affect siblings, a genetic defect was suspected, but intense investigation uncovered no gene locus.^{3,4} In addition, the recurrence pattern defied genetic explanation. A woman could have multiple unaffected infants prior to having an affected infant; however, after the index case there was a 90% probability that each subsequent baby born to that mother would be affected.⁵ NH would affect maternal half-siblings but not paternal half-siblings.^{3,6,7} Female survivors of NH went on to have healthy unaffected infants. Thus, NH appeared to be congenital and familial, but not hereditary.⁸ This pattern of recurrence led to the theorem that NH is caused by a maternofetal alloimmune disorder.

PATHOGENESIS

GALD, like other maternofetal alloimmune diseases, is mediated by immunoglobulin G (IgG).⁹ Maternal IgG antibodies are actively transported across the placenta to the fetus starting around the 12th week of gestation when the neonatal crystallizable fragment receptor (FcRn) is first expressed.^{10,11} These IgG antibodies serve

Keywords: acute liver failure, complement, gestational alloimmune liver disease, immunoglobulin

Received: 26.9.2013; **Accepted:** 27.10.2013; **Available online:** 27.11.2013

Address for correspondence: Amy G. Feldman, MD, Ann & Robert H. Lurie Children's Hospital of Chicago, 225 East Chicago Avenue, Box 57, Chicago, IL 60611-2605, United States. Tel.: +1 312 227 7600; fax: +1 312 227 9645

E-mail: afeldman@luriechildrens.org

Abbreviations: GALD: gestational alloimmune liver disease; FcRn: fragment receptor; IgG: immunoglobulin G; IVIG: intravenous immunoglobulin; NH: neonatal hemochromatosis; NTBI: non-transferrin bound iron

<http://dx.doi.org/10.1016/j.jceh.2013.10.004>

to provide the fetus with humoral immunity as the fetal and newborn adaptive immune system is immature and incapable of warding off infection. Gestational alloimmunity occurs when a woman is exposed to a fetal antigen that she does not recognize as “self”. This exposure results in sensitization and production of IgG antibodies against the fetal-derived antigen. Unlike most gestational alloimmune disease such as hydrops fetalis, ABO incompatibility hemolysis, and alloimmune thrombocytopenia in which IgG antibodies are directed against blood elements inherited from the father, in GALD maternal IgG antibodies are directed against fetal hepatocytes.¹ The antigen of target appears to be a hepatocyte specific protein that is either uniquely expressed by fetal hepatocytes or is highly sequestered in mature liver. If the antigen is uniquely expressed during fetal development, the mother may have lost tolerance to this self-antigen over time in the absence of central immune tolerance. Alternatively, if the antigen is sequestered in the mature liver, the same could occur in the absence of central tolerance. In either scenario, maternal exposure to this antigen induces an immune response that targets fetal hepatocytes. Non-hepatocyte liver cells and extrahepatic tissue do not appear to be attacked by this primary immune process. It remains unclear how antigen exposure to the maternal circulation occurs. We hypothesize that antigen crosses the placenta either when it becomes trapped in/on an exocytic vesicle, or when soluble protein is spilled during apoptosis during rapid liver development.

Once sensitization to the fetal antigen has occurred, specific reactive IgG is passed to the fetus where it binds to a hepatocyte antigen and initiates an innate immune response. The terminal complement cascade is activated by the classical pathway resulting in formation of the membrane attack complex.¹ Immunohistochemical staining identifies the C5b-9 complex, the neoantigen created during terminal complement cascade activation, in nearly all hepatocytes of infants with GALD.¹ Thus, complement-mediated hepatocyte injury has become a defining feature of GALD.

LIVER PATHOLOGY

Study of autopsy specimens has provided extensive description of the liver pathology in NH.^{2,12-14} The liver pathology in NH with proven GALD etiology (GALD-NH) is identical to that described in NH prior to discovery of GALD. Liver tissue of infants with GALD-NH displays severe injury with marked loss of hepatocytes (often less than 10% of normal newborn liver).^{15,16} In some cases, no hepatocytes remain.¹⁷ Surviving hepatocytes show siderosis, giant-cell or pseudoacinar transformation, and canalicular bile plugs. Unlike the hazy iron staining of normal newborn liver, the hepatocyte siderosis seen in NH is coarsely granular. Severe pan-lobular parenchymal

fibrosis is a dominant feature, and regenerative nodules are commonly observed. About 50% of patients can be said to have cirrhosis. Portal tracts are relatively spared from injury, and inflammation is minimal. Inflammation in the parenchyma consists of macrophages and neutrophils, innate immune cells that may be recruited by C3a and C5a during activation of the terminal complement cascade. Complement-mediated hepatocyte injury can be demonstrated using immunohistochemical staining for C5b-9 to identify accumulations of the membrane attack complex in hepatocytes and giant cells.

GALD can also cause acute liver injury to the fetal liver resulting in stillbirth or neonatal demise.¹⁸ The livers from these infants demonstrate hepatocyte necrosis without collapse, fibrosis or inflammation indicating a hyperacute process. There may not be any siderosis in the liver or other tissues. It remains unclear why certain infants develop this hyperacute liver failure while others present with congenital cirrhosis.

EXTRAHEPATIC MANIFESTATIONS OF GESTATIONAL ALLOIMMUNE LIVER DISEASE

Extrahepatic siderosis in NH is most frequently seen in the acinar epithelium of the exocrine pancreas, myocardium, epithelia of thyroid follicles, and the mucosal or minor salivary glands of the oronasopharynx and respiratory tree.^{12,15,19,20} In a recent cohort of 33 fetuses with NH, thyroid siderosis was more frequent than pancreatic.¹⁴ Less frequently affected are the gastric glands, Brunner glands, parathyroid glands, thymus (Hassall corpuscles), renal tubules, pancreatic islets, adenohypophysis, and chondrocytes in hyaline cartilage. The reticuloendothelial system is relatively spared, so spleen, lymph nodes, and bone marrow contain trivial quantities of stainable iron.

The mechanisms of iron overload and the specific distribution of siderosis in NH have been carefully examined. The fetus must closely regulate placental iron transport to ensure adequate iron for growth and oxygen carrying capacity while preventing possible toxic iron overload from the mother's huge iron pool. The fetal liver controls placental iron flux in a manner similar to how the post-natal liver regulates intestinal iron, by sensing iron sufficiency and producing hepcidin as a regulatory feedback molecule.²¹ In states of iron sufficiency, the liver produces hepcidin, which acts to suppress the cell-surface expression of ferroportin, a transmembrane iron transporter that transfers iron out of cells. Hepcidin binds ferroportin resulting in internalization and proteosomal degradation. This decrease in ferroportin results in decreased iron influx. In fetuses with GALD, liver injury results in significantly decreased production of hepcidin.¹⁵ As a result, there is less negative feedback on ferroportin and excess iron is transported from the placenta to the fetal liver. In addition, transferrin gene expression is decreased,

resulting in reduced iron binding capacity. The result is fetal iron overload and an excess of circulating non-transferrin bound iron (NTBI).

The tissue distribution of extrahepatic siderosis in the NH phenotype seems to be a function of the ability of various tissues to manage excess circulating NTBI. Tissues affected by siderosis express ZIP14, a known NTBI transporter.¹⁵ However, several tissues that express ZIP14 do not develop siderosis in GALD. Tissues that are unaffected by siderosis express ferroportin, which permits iron export. Thus, extrahepatic tissues that are ZIP14 positive and ferroportin negative are uniquely susceptible to siderosis. Reticuloendothelial cells are spared siderosis because they express ferroportin, which in the state of hepcidin paucity is fully active. Hepatocytes express transporters for transferrin bound iron and NTBI, and ferroportin. Accrual of hemosiderin in hepatocytes is likely to be a function of injury and perhaps relatively reduced ferroportin expression or function.

Renal hypoplasia with dysgenesis of proximal tubules and paucity of peripheral glomeruli has been described in infants with NH.^{22,23} This pattern of renal abnormality is best explained as a developmental abnormality. Expansion of the proximal tubule and associated glomeruli from the 24th week of gestation onward is dependent upon angiotensinogen, which is synthesized exclusively by the liver. In a study of livers and kidneys from infants with GALD-NH, hepatocyte mass and angiotensinogen gene expression were markedly reduced relative to normal.¹⁶ All GALD infants also had some degree of renal proximal tubule paucity, as demonstrated by quantitative immunohistochemistry staining for the proximal tubule marker fumaryl acetoacetic acid hydrolase. Liver expression of angiotensinogen inversely correlated with proximal tubule density. Therefore, it appears that alloimmune liver injury leads to reduced hepatocyte mass, which results in reduced angiotensinogen production, which in turn leads to defective renal development. This pathologic process dates the onset of liver injury in GALD-NH to before the 24th week of gestation.

CAUSES OF NEONATAL HEMOCHROMATOSIS PHENOTYPE OTHER THAN GESTATIONAL ALLOIMMUNE LIVER DISEASE

Occasionally, the NH phenotype can arise from non-GALD diseases including perinatal infection,^{3,24,25} trisomy 21,^{14,26–28} mitochondrial DNA depletion due to deoxyguanosine kinase deficiency (DGUOK gene mutations),^{29–31} bile acid synthetic defect delta 4-3-oxosteroid 5 beta-reductase deficiency (SRD5B1 mutations),^{32–34} GRACILE syndrome (BCS1L mutation),^{35,36} myofibromatosis,¹⁴ tricho-hepato-enteric syndrome^{37,38}

and Martinez-Frias syndrome.³⁹ In cases of trisomy 21, transient megakaryocytic leukemia may explain the NH phenotype. For the other diseases, fetal liver injury resulting in impaired regulation of placental iron flux is hypothesized to cause NH. Together, these causes account for no more than 2% of NH cases.

CLINICAL FINDINGS

GALD can present anytime from 18 weeks gestation to 3 months post-delivery. The majority of infants present with liver failure within hours of birth. NH is one of the most common causes of liver failure in the neonatal period.⁴⁰ Infants with GALD are often hypoglycemic, coagulopathic, hypoalbuminemic, jaundiced, and edematous. They may have renal involvement and be oliguric. There is frequently a history of intrauterine growth restriction, oligohydramnios, and prematurity. In rarer cases, liver disease may take days to weeks to present. There is a spectrum of disease severity, ranging from acute liver failure to “affected” babies with no clinical disease. Affected twins may have different clinical presentations; with one twin severely affected and the other minimally so.⁴¹ Interestingly, infants with NH often have persistent patency of the ductus venosus on ultrasound.^{2,42} This finding is of unclear clinical significance.

Laboratory evaluation reveals marked hyperbilirubinemia (bilirubin exceeding 30 mg/dL) with elevated conjugated and unconjugated portions. Aminotransferases rarely exceed 100 IU/L, whereas α -fetoprotein levels are very high (100,000–600,000 ng/mL).^{2,7} Iron studies reveal high serum ferritin levels (>800 ng/mL), low transferrin levels and high iron saturations.¹⁵ Serum ferritin levels are a sensitive indicator for NH but are not specific as many liver diseases will show similar elevations.

GALD (and GALD-NH) should be at the top of the differential for a neonate presenting in acute liver failure. Other processes that can cause neonatal liver failure include mitochondrial diseases, bile acid synthetic defects, tyrosinemia, hemophagocytic lymphohistiocytosis, ABCB11 gene mutations, hereditary galactosemia, hereditary fructose intolerance, and infection. Clinically, NH infants are unique in that they have evidence of fetal insult and neonatal liver failure. They are extremely coagulopathic, but have low serum aminotransferases (in contrast to infants with virally induced acute liver failure who have extremely high serum aminotransferases). Infants with GALD-NH may be misdiagnosed as having tyrosinemia due to elevated tyrosine levels, but they do not have succinylacetone in the urine. Infants with NH may also be misdiagnosed as having a bile acid defect; however, they will not have the classic pattern of bile metabolites found in serum and urine by mass spectroscopy. Finally, infants with GALD-NH should not have markedly elevated lactate levels as seen in infants with mitochondrial abnormalities.

DIAGNOSIS

GALD should be suspected in infants who manifest liver disease antenatally or in the immediate post birth period. It should also be considered in cases of unexplained stillbirth, neonatal demise, or early infant death. GALD is likely underdiagnosed. In cases of stillbirth or fetal loss, practitioners may not think to look for GALD before the index living case with NH occurs. Likewise, in live infants, symptoms of liver failure may be confused with those of global sepsis and practitioners may have difficulty making a diagnosis of NH with currently available tools. Global knowledge of this disorder and its wide spectrum of presentations may help to increase the number of cases that are accurately diagnosed.

Diagnosis of NH rests upon diagnosing extrahepatic siderosis: the complex of severe liver disease and extrahepatic siderosis defines the condition. Siderosis in the liver alone is not diagnostic as the normal newborn liver can contain quantities of iron that are stainable (though quantitatively different to an experienced pathologist). In addition, pathologic hepatic siderosis can be seen in several neonatal liver diseases. There is no known value of iron content in the liver which can accurately discriminate between NH and other causes of neonatal iron overload. Likewise, absence of liver siderosis does not rule out NH, as some GALD infants may have acute injury without iron overload and other GALD cases are associated with complete hepatocyte destruction, which precludes hepatic siderosis.

Extrahepatic siderosis in live infants is demonstrated by iron staining (Prussian blue, Perl's stain) of tissues affected by siderosis or by magnetic resonance imaging (MRI) (Figure 1). Glandular tissue containing iron can most easily be obtained from a biopsy of the oral mucosa.^{43,44} The surgery is minimally invasive and when performed by a competent oral surgeon, no complications are seen. Bleeding, which may potentially be made worse by coagulopathy, is controlled by local measures and has not been a serious problem in any case. No fresh frozen plasma or recombinant factor VII is necessary beforehand. One must be sure to obtain a specimen that contains submucosal glands. T2 weighted MRI can also be used to document siderosis as iron-laden tissue has a different magnetic susceptibility than normal tissue, particularly in the liver and pancreas. Although no formal studies have been done, in our experience an adequate oral biopsy or T2 weighted MRI will each demonstrate abnormal iron in 60% of cases with autopsy proven NH. Together, they have a sensitivity that approaches 80%. On autopsy, NH can be demonstrated by staining typically affected tissues for iron.

The following diagnostic approach can be utilized to avoid missing cases of NH (Figure 2). In a newborn with liver failure or other suspicious clinical circumstance

such as nonimmune hydrops, an attempt should be made to identify extrahepatic siderosis by oral biopsy or MRI. It is not necessary to perform both biopsy and MRI simultaneously as either can confirm the diagnosis. One should be performed, and only if that test is negative should the other be performed. If either is positive, the diagnosis of NH is made. Testing for non-GALD causes of NH (bile acid synthesis defect and mitochondrial DNA depletion due to DGUOK mutation) can then be performed as indicated to ensure the NH is secondary to GALD. If extrahepatic siderosis cannot be demonstrated, liver biopsy for C5b-9 staining can be considered. In cases of fetal loss or stillbirth, NH should always be considered and postmortem studies should be performed to look for extrahepatic siderosis. Stillborn liver tissue is often macerated making C5b-9 testing less useful in these cases.

TREATMENT

Literature suggests that the prognosis for severe NH without intervention is very poor. In the past, a cocktail of antioxidants and an iron chelator was used based on the hypothesis that liver injury was secondary to oxidative injury caused by iron overload. Survival rates with this treatment regimen were dismal with rates as low as 10–20%.^{45–47} After the discovery that NH was caused by GALD, a new treatment regimen was initiated utilizing the combination of double-volume exchange transfusion to remove existing reactive antibody followed immediately by administration of high-dose intravenous immunoglobulin (IVIG) (1 g/kg) to block antibody induced complement activation. The published experience with this treatment regimen shows marked improvement in survival compared to historical controls.⁴⁸ Of 16 infants treated, twelve subjects (75%) survived without a liver transplantation compared to 17% of historical control patients ($P < 0.001$). The remaining four infants died, two without a liver transplant and two after liver transplantation. Unpublished experience (data collected in our center) includes over 50 infants treated with IVIG with or without exchange transfusion. Survival of these cases exceeds 80% without liver transplant. Thus, this new therapy appears to offer significant survival benefit in treating NH. It can be utilized in less than sophisticated medical environments. Sepsis and other catastrophic events that intervene are the major cause of treatment failure in 20% of patients.

It is our recommendation that any infant in liver failure should be given one dose of IVIG while NH is being considered. The medication is benign and a single dose poses little risk to the newborn, regardless of disease etiology. If NH is proven, and the infant has not improved, an exchange transfusion should be performed followed by administration of a second dose of IVIG. Normalization of the international normalized ratio (INR) may take 4–6 weeks as this

Figure 1 MRI and histopathology of NH. Liver failure was noted at birth in a 2,900 g, 38-weeks gestation male infant. Decreased fetal movement and oligohydramnios had been noted at 36-weeks gestation. Refractory hypoglycemia and severe coagulopathy were prominent signs of liver failure. NH was suspected. On magnetic resonance imaging performed at 8 days of age, the liver (left arrow) and pancreas (right arrow) had a markedly reduced T2 signal intensity (dark area) relative to the spleen. Serum iron indices were not measured. Supportive therapy failed, and the infant expired on day of life 9. The composite image shows the most prominent histological findings on the postmortem examination. (a) Severe liver damage. A trichrome stain shows a fibrotic portal tract on the upper left. The hepatic parenchyma has been replaced by necrotic and fibrous tissue (blue-stained areas) and ductules; there were very few preserved hepatocytes (original magnification $\times 100$). Perl's Prussian blue stain showed abnormal iron deposition in the (b) liver, (c) pancreas, (d) myocardium, (e) Hassall's corpuscles of the thymus, and (f) thyroid follicles. Original magnifications: a = $100\times$, b–c = $200\times$ and d–f = $400\times$. The liver iron content was modestly elevated at $4,559 \mu\text{g/g}$ wet weight (reference normal 200–2,000). R, right; L, left. (Reprinted from Hepatology 2006; 43:654–660 with permission).

therapy reduces ongoing immune-mediated injury but does not reverse liver disease that has already occurred.⁴⁸

Historically, NH is a frequent indication for liver transplantation in the first 3 months of life.⁴⁶ However, transplantation is extremely challenging in these patients given their frequent history of prematurity, small size, and multiorgan failure.⁴⁹ The overall survival of infants receiving a liver transplant for the indication of NH is approximately 35%.⁴⁸ The timing of liver transplantation in these patients is difficult. Transplant is often considered

when medical management appears to be failing. However, medical management most often fails during settings that would also preclude transplant (concurrent infection, intracranial hemorrhage, multiorgan failure). If liver transplant is performed, it is impossible to know whether medical treatment would truly have failed. Therefore, caution must be used when considering liver transplantation.

There are limited published studies on long-term outcome of babies who receive medical treatment for NH. In our experience, infants recover sufficiently to go

Figure 2 Diagnostic algorithm for neonatal hemochromatosis (NH). In newborns with liver failure or other clinical circumstance suspicious for NH, the infant should be given one dose of intravenous immunoglobulin (IVIG). An attempt should be made to identify extrahepatic siderosis by MRI or buccal biopsy. If either demonstrates iron overload, the diagnosis of NH is made. If both are negative, a liver biopsy for C5b-9 staining should be considered. GALT, gestational alloimmune liver disease.

home in 1–4 months. However, it may take 2–4 years for the liver to fully recover. In two isolated cases, liver biopsies showed reversal of cirrhosis suggesting that the neonatal liver is able to recover even from severe injury.

PREVENTION

Once a woman has delivered an infant with NH, the probability that the next pregnancy will be lethally affected is greater than 90%.⁵ However, recurrence of severe NH can be prevented by treatment with IVIG during gestation.^{5,7} Our current recommendation is that subsequent pregnancies be treated with 1 g/kg body weight (maximum 60 g) of IVIG at 14 weeks, 16 weeks, and then weekly from the 18th week of pregnancy until the end of gestation. Records of 140 pregnancies treated under these guidelines have been collected at our center, with good outcome in 99% of cases. One pregnancy was lost at 22 weeks due to severe acute GALT (treatment for this woman was started at 18 weeks under an old protocol). Two infants were born prematurely at 26 and 32 weeks and both survived intact. All other infants were born full term with no signs of fetal distress or liver disease. Five infants experienced clinical liver disease during childhood. Four of these children had full recoveries, whereas one died at 2 months of age secondary to post viral

encephalomyelitis. Others have reported similar success with this approach.^{50–53} The success of this preventative therapy provides the impetus to make a diagnosis in any affected infant and stillbirth, so the family can have a healthy baby in the future.

CONCLUSIONS AND FUTURE RESEARCH

GALT is the most common cause of neonatal acute liver injury as well as in cases of stillbirth, fetal demise, and early postnatal death. It is likely underdiagnosed. Diagnosis rests upon demonstrating extrahepatic siderosis for identifying NH, which is almost always due to GALT. Once a mother has given birth to an NH infant, the risk for recurrence in subsequent offspring is very high, so IVIG should be used in future pregnancies. Although much has been learned about NH in the past five years, there remain many unanswered questions. How does fetal antigen gain exposure to maternal circulation? Why is there such a spectrum of disease ranging from stillbirth and gestational loss, to acute liver failure at birth, to congenital cirrhosis? Why do some affected infants experience only liver siderosis while others have extensive extrahepatic siderosis? Development of a suitable animal model is necessary to further study this disease, allowing for manipulation of genetic and environmental susceptibility factors.

CONFLICTS OF INTEREST

All authors have none to declare.

REFERENCES

- Pan X, Kelly S, Melin-Aldana H, Malladi P, Whittington PF. Novel mechanism of fetal hepatocyte injury in congenital alloimmune hepatitis involves the terminal complement cascade. *Hepatology*. Jun 2010;51(6):2061–2068.
- Knisely AS, Mieli-Vergani G, Whittington PF. Neonatal hemochromatosis. *Gastroenterol Clin North Am*. Sep 2003;32(3):877–889. vi–vii.
- Kelly AL, Lunt PW, Rodrigues F, et al. Classification and genetic features of neonatal haemochromatosis: a study of 27 affected pedigrees and molecular analysis of genes implicated in iron metabolism. *J Med Genet*. Sep 2001;38(9):599–610.
- Hardy L, Hansen JL, Kushner JP, Knisely AS. Neonatal hemochromatosis. Genetic analysis of transferrin-receptor, H-apoferritin, and L-apoferritin loci and of the human leukocyte antigen class I region. *Am J Pathol*. Jul 1990;137(1):149–153.
- Whittington PF, Kelly S. Outcome of pregnancies at risk for neonatal hemochromatosis is improved by treatment with high-dose intravenous immunoglobulin. *Pediatrics*. Jun 2008;121(6):e1615–1621.
- Verloes A, Temple IK, Hubert AF, et al. Recurrence of neonatal haemochromatosis in half sibs born of unaffected mothers. *J Med Genet*. Jun 1996;33(6):444–449.
- Whittington PF, Hibbard JU. High-dose immunoglobulin during pregnancy for recurrent neonatal haemochromatosis. *Lancet*. Nov 6–12 2004;364(9446):1690–1698.
- Whittington PF. Neonatal hemochromatosis: a congenital alloimmune hepatitis. *Semin Liver Dis*. Aug 2007;27(3):243–250.
- Hoftman AC, Hernandez MI, Lee KW, Stiehm ER. Newborn illnesses caused by transplacental antibodies. *Adv Pediatr*. 2008;55:271–304.
- Simister NE. Placental transport of immunoglobulin G. *Vaccine*. Jul 28 2003;21(24):3365–3369.
- Palmeira P, Quinello C, Silveira-Lessa AL, Zago CA, Carneiro-Sampaio M. IgG placental transfer in healthy and pathological pregnancies. *Clin Dev Immunol*. 2012;2012:985646.
- Knisely AS. Iron and pediatric liver disease. *Semin Liver Dis*. Aug 1994;14(3):229–235.
- Silver MM, Valberg LS, Cutz E, Lines LD, Phillips MJ. Hepatic morphology and iron quantitation in perinatal hemochromatosis. Comparison with a large perinatal control population, including cases with chronic liver disease. *Am J Pathol*. Nov 1993;143(5):1312–1325.
- Collardeau-Frachon S, Heissat S, Bouvier R, et al. French retrospective multicentric study of neonatal hemochromatosis: importance of autopsy and autoimmune maternal manifestations. *Pediatr Dev Pathol*. Nov–Dec 2012;15(6):450–470.
- Bonilla S, Prozialeck JD, Malladi P, et al. Neonatal iron overload and tissue siderosis due to gestational alloimmune liver disease. *J Hepatol*. Jun 2012;56(6):1351–1355.
- Bonilla SF, Melin-Aldana H, Whittington PF. Relationship of proximal renal tubular dysgenesis and fetal liver injury in neonatal hemochromatosis. *Pediatr Res*. Feb 2010;67(2):188–193.
- Gilmour SM, Hughes-Benzie R, Silver MM, Roberts EA. Le foie vide: a unique case of neonatal liver failure. *J Pediatr Gastroenterol Nutr*. Dec 1996;23(5):618–623.
- Whittington PF, Pan X, Kelly S, Melin-Aldana H, Malladi P. Gestational alloimmune liver disease in cases of fetal death. *J Pediatr*. Oct 2011;159(4):612–616.
- Whittington PF. Fetal and infantile hemochromatosis. *Hepatology*. Apr 2006;43(4):654–660.
- Silver MM, Beverley DW, Valberg LS, Cutz E, Phillips MJ, Shaheed WA. Perinatal hemochromatosis. Clinical, morphologic, and quantitative iron studies. *Am J Pathol*. Sep 1987;128(3):538–554.
- Chua AC, Delima RD, Morgan EH, et al. Iron uptake from plasma transferrin by a transferrin receptor 2 mutant mouse model of haemochromatosis. *J Hepatol*. Mar 2010;52(3):425–431.
- Bale PM, Kan AE, Dorney SF. Renal proximal tubular dysgenesis associated with severe neonatal hemosiderotic liver disease. *Pediatr Pathol*. May–Jun 1994;14(3):479–489.
- Johal JS, Thorp JW, Oyer CE. Neonatal hemochromatosis, renal tubular dysgenesis, and hypocalvaria in a neonate. *Pediatr Dev Pathol*. Sep–Oct 1998;1(5):433–437.
- Kershisnik MM, Knisely AS, Sun CC, Andrews JM, Wittwer CT. Cytomegalovirus infection, fetal liver disease, and neonatal hemochromatosis. *Hum Pathol*. Sep 1992;23(9):1075–1080.
- Metzman R, Anand A, DeGiulio PA, Knisely AS. Hepatic disease associated with intrauterine parvovirus B19 infection in a newborn premature infant. *J Pediatr Gastroenterol Nutr*. Jul 1989;9(1):112–114.
- Cheung PC, Ng WF, Chan AK. Neonatal haemochromatosis associated with Down syndrome. *J Paediatr Child Health*. Jun 1995;31(3):249–252.
- Ruchelli ED, Uri A, Dimmick JE, et al. Severe perinatal liver disease and Down syndrome: an apparent relationship. *Hum Pathol*. Dec 1991;22(12):1274–1280.
- Neil E, Cortez J, Joshi A, et al. Hepatic failure, neonatal hemochromatosis and porto-pulmonary hypertension in a newborn with trisomy 21 – a case report. *Ital J Pediatr*. 2010;36:38.
- Hanchard NA, Shchelochkov OA, Roy A, et al. Deoxyguanosine kinase deficiency presenting as neonatal hemochromatosis. *Mol Genet Metab*. Jul 2011;103(3):262–267.
- Nobre S, Grazina M, Silva F, Pinto C, Goncalves I, Diogo L. Neonatal liver failure due to deoxyguanosine kinase deficiency. *BMJ Case Rep*. 2012;2012.
- Pronicka E, Weglewska-Jurkiewicz A, Taybert J, et al. Post mortem identification of deoxyguanosine kinase (DGUOK) gene mutations combined with impaired glucose homeostasis and iron overload features in four infants with severe progressive liver failure. *J Appl Genet*. Feb 2011;52(1):61–66.
- Shneider BL, Setchell KD, Whittington PF, Neilson KA, Suchy FJ. Delta 4-3-oxosteroid 5 beta-reductase deficiency causing neonatal liver failure and hemochromatosis. *J Pediatr*. Feb 1994;124(2):234–238.
- Siafakas CG, Jonas MM, Perez-Atayde AR. Abnormal bile acid metabolism and neonatal hemochromatosis: a subset with poor prognosis. *J Pediatr Gastroenterol Nutr*. Sep 1997;25(3):321–326.
- Ueki I, Kimura A, Chen HL, et al. SRD5B1 gene analysis needed for the accurate diagnosis of primary 3-oxo-Delta4-steroid 5beta-reductase deficiency. *J Gastroenterol Hepatol*. May 2009;24(5):776–785.
- Rapola J, Heikkilä P, Fellman V. Pathology of lethal fetal growth retardation syndrome with aminoaciduria, iron overload, and lactic acidosis (GRACILE). *Pediatr Pathol Mol Med*. Mar–Apr 2002;21(2):183–193.
- Visapaa I, Fellman V, Vesa J, et al. GRACILE syndrome, a lethal metabolic disorder with iron overload, is caused by a point mutation in BCS1L. *Am J Hum Genet*. Oct 2002;71(4):863–876.
- Verloes A, Lombet J, Lambert Y, et al. Tricho-hepato-enteric syndrome: further delineation of a distinct syndrome with neonatal hemochromatosis phenotype, intractable diarrhea, and hair anomalies. *Am J Med Genet*. Feb 11 1997;68(4):391–395.
- Fabre A, Martinez-Vinson C, Goulet O, Badens C. Syndromic diarrhea/tricho-hepato-enteric syndrome. *Orphanet J Rare Dis*. 2013;8:5.
- Martinovici D, Ransy V, Vanden Eijnden S, et al. Neonatal hemochromatosis and Martinez-Frias syndrome of intestinal atresia

- and diabetes mellitus in a consanguineous newborn. *Eur J Med Genet.* Jan–Feb 2010;53(1):25–28.
40. Durand P, Debray D, Mandel R, et al. Acute liver failure in infancy: a 14-year experience of a pediatric liver transplantation center. *J Pediatr.* Dec 2001;139(6):871–876.
 41. Ekong UD, Kelly S, Whittington PF. Disparate clinical presentation of neonatal hemochromatosis in twins. *Pediatrics.* Dec 2005;116(6):e880–e884.
 42. Tsai A, Paltiel HJ, Sena LM, Kim HB, Fishman SJ, Alomari AI. Neonatal hemochromatosis and patent ductus venosus: clinical course and diagnostic pitfalls. *Pediatr Radiol.* Aug 2009;39(8):823–827.
 43. Knisely AS, O'Shea PA, Stocks JF, Dimmick JE. Oropharyngeal and upper respiratory tract mucosal-gland siderosis in neonatal hemochromatosis: an approach to biopsy diagnosis. *J Pediatr.* Nov 1988;113(5):871–874.
 44. Smith SR, Shneider BL, Magid M, Martin G, Rothschild M. Minor salivary gland biopsy in neonatal hemochromatosis. *Arch Otolaryngol Head Neck Surg.* Jun 2004;130(6):760–763.
 45. Flynn DM, Mohan N, McKiernan P, et al. Progress in treatment and outcome for children with neonatal haemochromatosis. *Arch Dis Child Fetal Neonatal Ed.* Mar 2003;88(2):F124–F127.
 46. Rodrigues F, Kallas M, Nash R, et al. Neonatal hemochromatosis—medical treatment vs. transplantation: the king's experience. *Liver Transpl.* Nov 2005;11(11):1417–1424.
 47. Vohra P, Haller C, Emre S, et al. Neonatal hemochromatosis: the importance of early recognition of liver failure. *J Pediatr.* Apr 2000;136(4):537–541.
 48. Rand EB, Karpen SJ, Kelly S, et al. Treatment of neonatal hemochromatosis with exchange transfusion and intravenous immunoglobulin. *J Pediatr.* Oct 2009;155(4):566–571.
 49. Sundaram SS, Alonso EM, Whittington PF. Liver transplantation in neonates. *Liver Transpl.* Aug 2003;9(8):783–788.
 50. Faas D, Axt-Flidner R, Zimmer KP, Heckmann M. Prophylactic prenatal therapy with intravenous immunoglobulins for patients at risk for neonatal haemochromatosis. *Z Geburtshilfe Neonatol.* Dec 2011;215(6):246–249.
 51. Nicholl MC. Successful pregnancy outcome with the use of antenatal high-dose intravenous immunoglobulin following previous neonatal death associated with neonatal haemochromatosis. *Aust N Z J Obstet Gynaecol.* Aug 2010;50(4):403–405.
 52. Tanaka H, Haba R, Itoh S, Sakamoto H, Hata T. Prenatal high-dose immunoglobulin treatment for neonatal hemochromatosis: a case report and review of the literature. *J Obstet Gynaecol Res.* Dec 2011;37(12):1891–1894.
 53. Venkat-Raman N, Venkata-Krishnan RV, Howarth ES. Successful pregnancy outcome following maternal intravenous immunoglobulin treatment in a woman with recurrent perinatal haemochromatosis. *Prenat Diagn.* Dec 2006;26(13):1256–1259.