# Neonatal Hemochromatosis: Diagnostic Work-Up Based on a Series of 56 Cases of Fetal Death and Neonatal Liver Failure

Sophie Heissat, MD[1], Sophie Collardeau-Frachon, MD, PhD[2], Julien Baruteau, MD[3], Estelle Dubruc, MD[2], Raymonde Bouvier, MD[2], Monique Fabre, MD[4], Marie Pierre Cordier, MD[5], Pierre Broué, MD[3], Vincent Guigonis, MD, PhD[6], and Dominique Debray, MD, PhD[7]

**Objective** To define an algorithm to improve diagnosis of neonatal hemochromatosis (NH) related to gestational alloimmune liver disease (GALD), which is diagnosed by immunohistochemistry demonstrating activated complement at hepatocytes (IDACH).

**Study design** We assessed 56 instances of fetal death or neonatal liver failure (NLF; 2006-2009), 29 (7 stillborns, 22 NLF) with NH, and 27 (5 stillborns, 22 NLF) without NH (non-NH). Immunohistochemistry was retrospectively performed in 21 cases. Cases were grouped as follows: (1) GALD as demonstrated by IDACH (n = 17); (2) indeterminate for GALD (n = 28); or (3) alternate diagnosis found (n = 11). We compared cases of immunohistochemically proven GALD with those with an alternate diagnosis.

**Results** Of the 12 stillborns, 7 had NH because of GALD (NH-GALD), one was undeterminate, and 4 had alternate diagnoses (GALD excluded). Of the 22 newborns with NH, 6 had NH-GALD, one had mitochondrial respiratory chain disorder (MRCD), and 15 were indeterminate for GALD. Of 22 non-NH newborns, extrahepatic siderosis (EHS) was not assessed in 13 (3 GALD, 1 alternate diagnosis [MRCD] and 9 indeterminate GALD) and excluded in 9 (5 alternate diagnoses and 4 indeterminate GALD). The only clinical features found to be associated with GALD were intrafamilial recurrence, prematurity, and EHS.

**Conclusions** In unexplained fetal death or NLF, the diagnosis of subsets of NH requires tissue analysis (autopsy) to assess EHS. In patients with NH, if MRCD is ruled out, NH-GALD is likely. The rate of IDACH in the diagnosis of GALD in cases without NH requires further study. *(J Pediatr 2015;166:66-73)*.

Neonatal hemochromatosis (NH) (OMIM 231100) is a severe liver disorder associated with extrahepatic siderosis (EHS) sparing the reticuloendothelial system (RES) and fetal death or neonatal liver failure (NLF).[1] NH is characterized by high rates of mortality and recurrence (about 80%) in the progeny of affected women.[1,2] NH refers to the phenotypic expression of fetal liver disease regardless of etiology. Most NH is ascribed to gestational alloimmune liver disease (GALD, NH because of GALD [NH-GALD]).[3] It has also become clear in recent years that GALD can produce liver disease without EHS and that the absence of pathologic siderosis in the newborn liver does not exclude the diagnosis of GALD.[4,5] In 2010, it was reported that C5b-9 complex immunohistochemistry demonstrating activated complement in hepatocytes (IDACH) defined GALD among cases of severe neonatal liver disease.[6,7] Distinguishing between NH-GALD and NH because of other causes is important because gestational intravenous immunoglobulin (IV-IG) can palliate NH-GALD. Its value in non-NH-GALD is unknown. Moreover, exchange transfusion (ET) with IV-IG at birth lessens the severity of the illness in affected neonates and improves outcome.[7-9]

IV-IG infusion is expensive, may have adverse effects, and availability can be limited.[10] Therefore, expert counseling for family planning purposes is of importance. For this reason, since 2006, pediatricians and pathologists in France have been invited to submit cases of unexplained fetal death or NLF to a national NH advisory group to assess if GALD without IDACH was likely enough to recommend gestational IV-IG for future pregnancies.[11]

| | | | |
|---|---|---|---|
| DGUOK | Deoxyguanosine kinase | IUGR | Intrauterine growth restriction |
| EHS | Extrahepatic siderosis | IV-IG | Intravenous immunoglobulin |
| ET | Exchange transfusion | MRCD | Mitochondrial respiratory chain disorder |
| GA | Gestational age | MRI | Magnetic resonance imaging |
| GALD | Gestational alloimmune liver disease | NH | Neonatal hemochromatosis |
| HLH | Hemophagocytic lymphohistiocytosis | NH-GALD | Neonatal hemochromatosis because of gestational alloimmune liver disease |
| HUS | Hemolytic uremic syndrome | NLF | Neonatal liver failure |
| IDACH | Immunohistochemistry demonstrating activated complement in hepatocytes | RES | Reticuloendothelial system |

From the [1]Department of Pediatric Gastroenterology and Hepatology, Hôpital Femme Mère Enfant, Hospices Civils de Lyon et Université Lyon 1; [2]Department of Pathology, Hôpital Femme Mère Enfant, Hospices Civils de Lyon et Université Lyon 1, Lyon, France; [3]Department of Pediatric Hepatology and Metabolic Diseases, CHU Toulouse, Toulouse, France; [4]Department of Pathology, Villejuif, France; [5]Department of Genetics, Hôpital Femme-Mère-Enfant, HCL Lyon, Lyon, France; [6]Department of Pediatrics, Hôpital Mère et Enfant, CHU Limoges, Limoges, France; and [7]Medical-Surgical Center, Hepatology, and Transplantation AP-HP, Hôpital Necker Enfants Malades, Paris, France

The authors declare no conflicts of interest.


http://dx.doi.org/10.1016/j.jpeds.2014.09.030

Here, we report the results of a retrospective study of cases of fetal death and cases of NLF that were reviewed by the NH advisory group between 2006 and 2009. Immunostaining for the C5b-9 complex was performed a posteriori in all stillborns and in 9 newborns. Our aim was to characterize features suggesting GALD in stillborns and newborns with NLF. To this end, we compared cases of immunohistochemically proven GALD with those with an alternate diagnosis. We propose a work-up intended for use in the primary care setting to facilitate the diagnosis of GALD.

## Methods

All cases of fetal death or NLF referred to the national NH advisory group between 2006 and 2009 were included in the study. Infants born to women treated with IV-IG were excluded from the study. The study was conducted with the approval of the ethics committee of Limoges University Hospital, France.

Cases were analyzed independently by 3 pediatric hepatologists. Additional data were requested from the referring physicians whenever necessary. Liver biopsy and autopsy tissue samples were reviewed independently by 3 pathologists. NH was defined as severe liver injury associated with EHS sparing the RES. Cases without evidence of EHS, whether because of not finding EHS in a full autopsy or not having a full autopsy or abdominal magnetic resonance imaging (MRI) to examine, were referred to as cases without NH.

All data available from medical records were reviewed: maternal and gestational histories, gestational ages (GAs), initial symptoms, time from birth to initial symptoms, main initial clinical signs and laboratory results, abdominal MRI results (if available), histology results, treatment, and outcome. For the purpose of the study, immunostaining with monoclonal antibodies to human C5b-9 neoantigen (Quidel, San Diego, California) was retrospectively performed in 21 available liver biopsy samples as previously described.[6,7] All parents gave their consent for reviewing the clinical and histopathologic data.

Among the 56 cases included in the study, there were 12 stillborn fetuses and 44 newborns who presented with NLF, born to 45 women. Eight women had 2 or 3 stillborns or newborns included in the study (**Figure 1**).

Cases were divided into 2 groups. Group 1 (cases with NH) included 7 fetuses and 22 newborns with NH diagnosed either at autopsy or using MRI or oral mucosal biopsy. Group 2 (non-NH) included 5 fetuses and 22 newborns for whom EHS was either not demonstrated or not assessed. These groups were further divided into 3 subgroups according to GALD status based on results

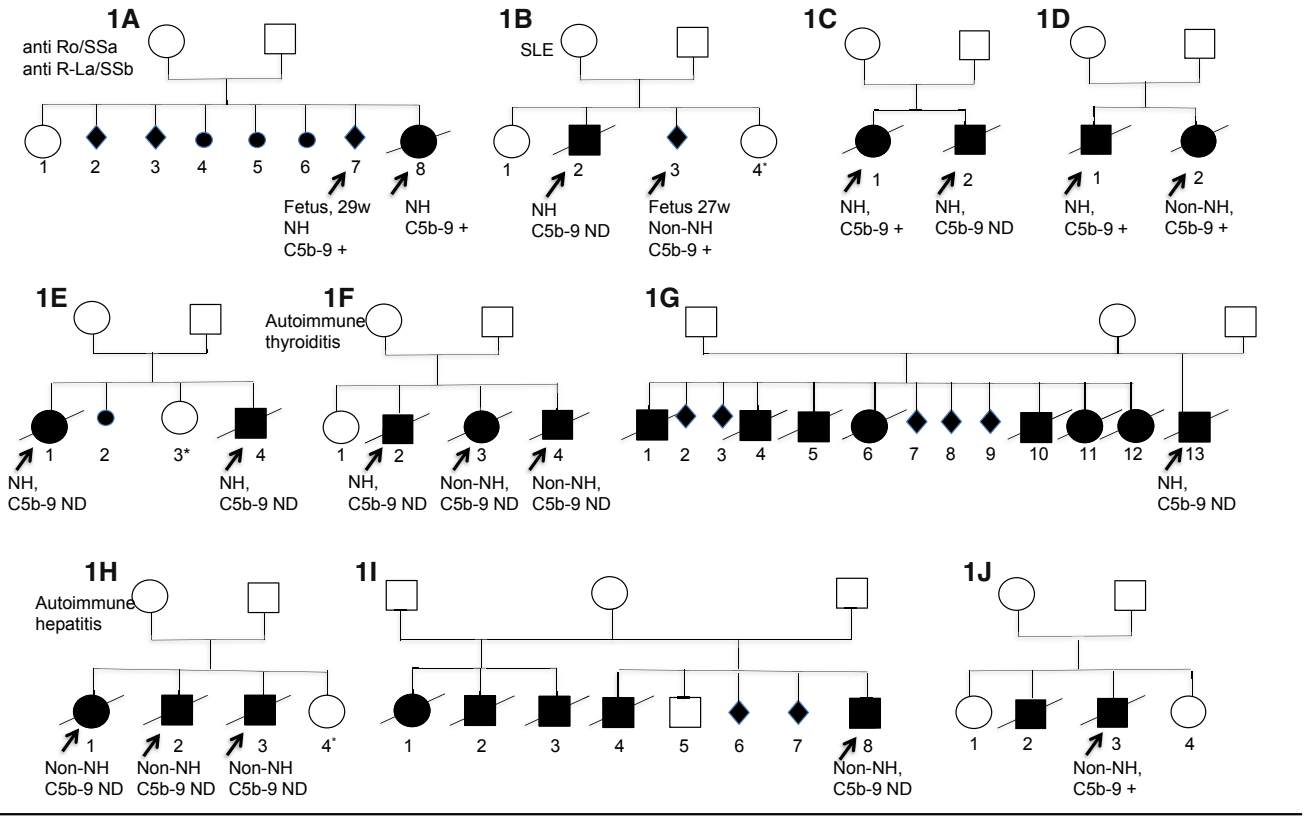


**Figure 1.** Outcome of gestations of 10 mothers with stillborn fetuses or NLF recurrences. *Arrows* indicate cases included in the series. *Large solid circles* and *squares* indicate affected newborns with NLF. *Large clear circles* and *squares* indicate healthy newborns. *Black diamonds* indicate fetal death, and *small solid circles* indicate miscarriage. *Survived following gestational IV-IG. *AIH*, autoimmune hepatitis; *SLE*, systemic lupus erythematosus.