**Department of Pediatrics**
**Gastroenterology, Hepatology, and Nutrition**
**Pediatric Liver Transplant Program**
**Pediatric Inflammatory Bowel Disease Program**
**Pediatric Transplant Hepatology Program**
500 Parnassus Avenue, MU 4 East
San Francisco, CA 94143-0136
Tel: 415/476-5892
Fax: 415/476-1343
TTY: 415/885-3889
www.ucsfhealth.org/childrens

University of California
San Francisco

**Melvin B. Heyman, MD, MPH**
Professor
Division Chief
Director, Pediatric IBD Program
Director, Pediatric Gastroenterology Fellowship Program

**Alex Green, DO**
Assistant Professor
Director, Pediatric Intestinal Motility Program

**Brandy Lu, MD**
Assistant Professor
Director, Pediatric Endoscopy

**Sue J. Rhee, MD**
Associate Professor
Director, Pediatric Intestinal Rehabilitation and Transplantation Program
Director, Pediatric Transplant Hepatology Fellowship Program

**Philip Rosenthal, MD**
Professor
Director, Pediatric Liver Transplant Program
Director, Pediatric Hepatology

**Walter Sipe, MD**
Associate Professor

**Patrika Tsai, MD, MPH**
Assistant Professor

**Janet Wojcicki, PhD, MPH**
Assistant Professor

**Elizabeth Yen, MD**
Assistant Professor

**Laura Cooke, RN, CPNP, MSN**
Pediatric IBD Nurse Practitioner

**Linda Chu, RN**
Nurse Case Manager

**Betsy Haas-Beckert, RN, PNPc, MSN**
Pediatric GI Nurse Practictioner

**Susan Stritzel-Diaz, RN, PNPc, MSN**
Pediatric Liver Transplant Nurse Practitioner

**Elizabeth McMillan, RD**
Pediatric Registered Dietitian

**Helene Omansky, LCSW**
Pediatric Social Worker

July 23, 2013

Lewis Levy, MD
VP of Corporate Medical Quality
Best Doctors, Inc.
100 Federal Street, 21st floor
Boston, MA 02110
FAX: 617-391-6494

Re: Jianhua Xie
DOB: 9/16/2012

Dear Dr. Levy:

I had the opportunity to review the clinical summary, history and course of Jianhua Xie. I have been requested to consult regarding diagnosis and best treatment options.

In brief this is a 10 month old boy with liver failure of undetermined etiology. The family is seeking a Best Doctors consultation regarding the diagnosis and treatment plan.

He was in his usual state of good health until early April 2013, when he a 3 day history of vomiting, diarrhea, increasing abdominal distension, periorbital and lower extremity edema, and increased work of breathing. He was seen in the emergency department of an academic children's hospital on 4/10/2013, where he was found to be febrile to 40 degrees Celsius. Anasarca and splenomegaly were noted on examination. He did not appear encephalopathic. Labs revealed evidence of fulminant hepatic failure, with an INR of 5.6, a conjugated bilirubin of 4 mg/dL, and mildly elevated AST/ALT (around 100 IU/L). At that time, his initial blood glucose was normal.

Of note, Jianhua's father had had some symptoms of vomiting and diarrhea just prior to this acute presentation. Additionally, Jianhua had had a chronic history of greasy, pale stools ("like olive oil"). There was no history of travel, medications, or herbal supplements.

Jianhua was resuscitated with 30 cc/kg normal saline and given broad-spectrum antibiotics after cultures were obtained. He was admitted to the pediatric intensive care unit for ongoing care, with the diagnosis of acute hepatic failure. His coagulopathy was not responsive to parenteral vitamin K administration. Hypoglycemia ensued, requiring intravenous dextrose administration. A PICC line was placed. For progressive anemia and coagulopathy, Jianhua received multiple transfusions of cryoprecipitate, packed red blood cells, and fibrinogen. He was treated with diuretics, antibiotics, fat-soluble vitamin supplementation (significant deficiencies discovered on testing), and a high MCT formula (Pregestamil).

An extensive work-up to determine the etiology of for his liver failure was unrevealing. Imaging revealed moderate intraperitoneal fluid, mild splenomegaly, evidence of portal hypertension, and multiple small, echogenic foci throughout

the right lobe of the liver. Please see below for full details. A urine toxin screen was negative, and a serum acetaminophen level was undetectable. Testing for autoimmune hepatitis associated antibodies was negative.

An infectious disease consultation was obtained, and evaluation for a number of pathogens was negative (EBV, CMV, HSV, and Hepatitis A/B/C). Blood cultures were negative. A catheterized urine culture grew between 1,000-10,000 cfu/mL of E coli, which was treated with antibiotics.

Hematology was consulted to rule out an infiltrative process. A serum ferritin and triglyceride level were both normal, arguing against hemophagocytic lymphohistiocytosis. By report, soluble IL-2 testing was unremarkable (results unavailable). Leukemia was felt to be extremely unlikely, given the normal neutrophil count. The labs and imaging were not felt to be consistent with lymphoma or a primary liver malignancy. Neuroblastoma was felt unlikely given the imaging findings, and a normal VMA level provided additional support in that regard.

Metabolics/genetics was consulted as well. Carbohydrate deficient glycoprotein, serum amino acids, and urine organic acids were reportedly unremarkable (formal results not available in reviewed medical records). A urinalysis showed trace reducing substances, but subsequent galactosemia screening was reportedly negative (results unavailable). A fecal elastase showed "severe exocrine pancreatic insufficiency (see below)." It appears that this was felt to be a false positive, given what was described as creamy consistency stool. Pancreatic enzyme supplements were not started. Genetic testing ('JaundiceChip' at Cincinnati Children's Hospital) revealed no mutations in the genes involved with alpha-1 antitrypsin deficiency, Alagille syndrome, PFIC-2, or PFIC-3. There was a clinical variant of unknown significance in 1 allele of the PFIC-1 gene (please see lab reports below for full details). A urine bile acid profile was unremarkable (full report listed in labs below).

Jianhua had a normal ophthalmologic exam, without any ocular evidence to suggest a specific metabolic or genetic disease.

He underwent a laparoscopic liver biopsy on 4/18/2013, revealing an established micronodular cirrhosis with bile ductular proliferation. The etiology of the cirrhosis was of unclear etiology.

The liver transplant team followed Jianhua closely during his inpatient stay, and he was listed for transplantation after lengthy discussions with the family and consulting services. By discharge in late April, his jaundice had resolved, and his INR was stable at 2.5 (without the need for any corrective products).

Jianhua has been followed closely by the transplant team as an outpatient, initially weekly and now every 2 weeks. The last available clinic note is from 6/6/2013. At that time, he was doing well. The impression was micronodular cirrhosis of unknown etiology, "probably some developmental gene disorder as the etiology." On examination, he was without jaundice. There were some excoriations on his forehead. His spleen was palpable 3 cm below the left costal margin. His liver was not enlarged, and was palpable and "firm" at the sternal edge. There was no edema. Jianhua's albumin was normal. His conjugated bilirubin was 0.0 mg/dL, and his INR was 1.4. His PELD score was "around 10." So, it was anticipated that the wait for transplant could be months, unless his condition were to deteriorate.

The parents' intake notes that Jianhua was seen again on 6/20/2013, at which time his bilirubin had apparently normalized. His serum ammonia was said to remain elevated. Reportedly, his transplant status has been changed from "active" to "hold", as he is so stable. The family has been told that although he will not likely require a liver transplant in the near term, this will be needed at some point in the future because of the significant damage to his liver.

The current plan is for follow-up with his primary gastroenterologist every two weeks and with the transplant team around July 20th, 2013. He is to have labs performed every 4 weeks.

Currently, Jianhua takes in 30-40 ounces per day of Pregestamil 20 calorie per ounce formula. A number of baby foods have also recently been introduced. He has not gained weight since his discharge in late April 2013, "but this may at least in part reflect the resolution of his fluid retention."

He has 2-3 brownish stools per day. There is no jaundice. He does regurgitate undigested formula at least once per day, particularly with positional changes and overfeeding.

He "sometimes sleeps a lot" and seems weak and listless to his parents. On occasion, the family will note some swelling around his eyes.

He does seem to have itchiness. He "is always trying to scratch himself" and will occasionally excoriate himself to the point of causing bleeding.

Jianhua's parents are particularly concerned about the possibility of NICCD (neonatal intrahepatic cholestasis caused by citrin deficiency) or any other unidentified cause that may explain their son's presentation. They are requesting a Best Doctors consultation regarding the diagnosis and optimal treatment plan moving forward.

Past Medical History:
1. Former 38.5-week gestational age infant. During the pregnancy, Jianhua's mother had an abnormal maternal serum screen and therefore underwent amniocentesis. This reportedly demonstrated a double Y chromosome in 10 of 20 cultures. Jianhua was vaginally, without any delivery complications. His birth weight was 7 pounds 5 ounces. He had a normal newborn screen
2. RSV bronchiolitis in January 2013. Recurrent wheezing since then.
3. Presumed milk protein intolerance. He had a history of emesis and diarrhea early in life that resolved upon changing to a soy-based formula.
4. Apart from RSV, no other history of recurrent infections.

Current Medications:
1. Spironolactone 12 mg (2.4 mL) PO BID
2. Ursodiol 18 mg (3.2 mL) PO TID
3. AquaDEKs 1 mL PO daily
4. Vitamin D3 1000 units (0.2 mL) PO daily
5. Aquasol E 50 units (0.3 mL) PO daily
6. Pregestamil formula 30-40 ounces PO daily

Allergies: NKDA

Social History: Jianhua lives with his parents and siblings in the Pacific Northwest.

Family History: Both parents are of Chinese ethnicity. The mother had a heart murmur when she was a young child but is otherwise healthy. The father is healthy. Jianhua has 3 older siblings, all of whom are healthy. There is no family history of liver disease.

Physical Examination:
Current weight – 17 pounds
Current length – approximately 26.5 inches

Laboratory Results:
- Labs, 6/6/2013:

| | |
|---|---|
| Collected Date | 06/06/2013 |
| Collected Time | 12:24:00 |

| Procedure | | Units | Ref Range |
|---|---|---|---|
| Sodium Level | 140 | mEq/L | [135-145] |
| Potassium Level | 4.2 | mEq/L | [3.5-5.5] |
| Chloride Lvl | 105 | mEq/L | [96-110] |
| CO2 Lvl | 22 | mEq/L | [18-27] |
| Anion Gap | 17.2 | mEq/L | [8.0-22.0] |
| Glucose Level | 70 | mg/dL | [60-105] |
| BUN | 8 | mg/dL | [6-20] |
| Creatinine | 0.4 | mg/dL | [0.1-0.4] |
| Estimated GFR (Schwartz Bedside) | **69 Lf** | mL/min/1.73 m2 | |
| Calcium Level, Total | 9.7 | mg/dL | [8.7-10.7] |
| Magnesium Serum | 2.1 | mg/dL | [1.8-2.4] |
| Phosphorus Serum | 5.0 | mg/dL | [3.9-6.5] |
| Bilirubin Total | 0.9 | mg/dL | [0.1-1.3] |
| Bilirubin Conjugated | 0.0 | mg/dL | [0.0-0.3] |
| Bilirubin Unconjugated | 0.3 | mg/dL | [0.0-1.1] |
| Bilirubin Delta | **0.6 H** | mg/dL | [0.0-0.2] |
| AST (SGOT) | **86 H** | IU/L | [3-74] |
| ALT (SGPT) | **47 H** | IU/L | [6-45] |
| Alkaline Phosphatase, Serum | **912 H** | IU/L | [95-380] |
| Gamma Glutamyltransferase | 23 | IU/L | [15-85] |
| Ammonia, Blood Venous i | 33 | mcmol/L | [9-35] |
| Albumin Level | 3.5 | g/dL | [2.9-5.5] |

| Procedure | | Units | Ref Range |
|---|---|---|---|
| White Blood Cell Count | 11.5 | K/mm3 | [6.0-17.5] |
| Platelet Count | **159 L** | K/mm3 | [250-600] |
| Hematocrit | **37.3** | % | [33.0-39.0] |
| Hemoglobin | 12.7 | g/dL | [10.5-13.5] |
| Red Blood Cell Count | 4.42 | M/mm3 | [3.70-5.30] |
| MCV | 84.4 | fl | [70.0-86.0] |
| MCH | 28.7 | pg | [23.0-31.0] |
| MCHC | 34.0 | % | [30.0-36.0] |
| RDW | 13.4 | % | [11.0-16.0] |
| Mean Platelet Volume | 11.1 | fl | [8.9-12.4] |
| Nucleated Red Cells | 0 | /100 WBC | |
| Polys, Absolute | 2852 | /mm3 | [1,500-5,000] |
| Lymphocytes, Absolute | 7199 | /mm3 | [1,500-8,500] |
| Monocytes, Absolute | **1116 H** | /mm3 | [0-500] |
| Eosinophils, Absolute | 264 | /mm3 | [0-300] |
| Basophils, Absolute | 46 | /mm3 | [0-50] |
| Immature Granulocytes, Absolute i | 23 | /mm3 | [0-50] |
| Absolute Neutrophil Ct Cal | 2852 | /mm3 | [1,500-5,400] |
| Polys, % | 24.8 | % | [15.0-45.0] |
| Lymphocytes, % | 62.6 | % | [30.0-75.0] |
| Monocytes, % | 9.7 | % | [0.0-10.0] |
| Eosinophils, % | 2.3 | % | [0.0-4.0] |
| Basophils, % | 0.4 | % | [0.0-1.0] |
| Immature Granulocytes, % i | **0.2 H** | % | [<=0.0] |
| RBC /Platelet Morphology | Performed | | |
| Schistocytes | **1+ *** | | |
| Burr Cells | **1+ *** | | |
| Reactive Lymphocytes | **3+ *** | | |

| Procedure | | Units | Ref Range |
|---|---|---|---|
| Prothrombin Time | **16.6 H** | second(s) | [12.5-15.2] |
| International Normalized Ratio | 1.4 | | |

- Labs, 5/23/2013:

| | |
|---|---|
| Collected Date | 05/23/2013 |
| Collected Time | 14:45:00 |

| Procedure | | Units | Ref Range |
|---|---|---|---|
| Vitamin D, 25-OH Total Level i | **11 L** | ng/mL | [30-100] |
| Vitamin D2 Level | <2 | ng/mL | |
| Vitamin D3 Level | 11 | ng/mL | |

- Labs, 5/15/2013

| | |
|---|---|
| Collected Date | 05/15/2013 |
| Collected Time | 14:10:00 |

| Procedure | | Units | Ref Range |
|---|---|---|---|
| Sodium Level | 140 | mEq/L | [135-145] |
| Potassium Level | 4.7 | mEq/L | [3.5-5.5] |
| Chloride Lvl | 108 | mEq/L | [96-110] |
| CO2 Lvl | 21 | mEq/L | [18-27] |
| Anion Gap | 15.7 | mEq/L | [8.0-22.0] |
| Glucose Level | 78 | mg/dL | [60-105] |
| BUN | 9 | mg/dL | [6-20] |
| Creatinine | 0.2 | mg/dL | [0.1-0.4] |
| Estimated GFR (Schwartz Bedside) | >120 f | mL/min/1.73 m2 | |
| Calcium Level, Total | 9.9 | mg/dL | [8.7-10.7] |
| Magnesium Serum | 2.4 | mg/dL | [1.8-2.4] |
| Phosphorus Serum | 4.8 | mg/dL | [3.9-6.5] |
| Bilirubin Total | 1.3 | mg/dL | [0.1-1.3] |
| Bilirubin Conjugated | 0.0 | mg/dL | [0.0-0.3] |
| Bilirubin Unconjugated | 0.6 | mg/dL | [0.0-1.1] |
| Bilirubin Delta | **0.7 H** | mg/dL | [0.0-0.2] |
| AST (SGOT) | **95 H** | IU/L | [3-74] |
| ALT (SGPT) | **58 H** | IU/L | [6-45] |
| Alkaline Phosphatase, Serum | **1047 H** | IU/L | [95-380] |
| Gamma Glutamyltransferase | 24 | IU/L | [15-85] |
| Albumin Level | 3.7 | g/dL | [2.9-5.5] |


| Procedure | | Units | Ref Range |
|---|---|---|---|
| Sodium Level | 140 | mEq/L | [135-145] |
| Potassium Level | 4.7 | mEq/L | [3.5-5.5] |
| Chloride Lvl | 108 | mEq/L | [96-110] |
| CO2 Lvl | 21 | mEq/L | [18-27] |
| Anion Gap | 15.7 | mEq/L | [8.0-22.0] |
| Glucose Level | 78 | mg/dL | [60-105] |
| BUN | 9 | mg/dL | [6-20] |
| Creatinine | 0.2 | mg/dL | [0.1-0.4] |
| Estimated GFR (Schwartz Bedside) | >120 f | mL/min/1.73 m2 | |
| Calcium Level, Total | 9.9 | mg/dL | [8.7-10.7] |
| Magnesium Serum | 2.4 | mg/dL | [1.8-2.4] |
| Phosphorus Serum | 4.8 | mg/dL | [3.9-6.5] |
| Bilirubin Total | 1.3 | mg/dL | [0.1-1.3] |
| Bilirubin Conjugated | 0.0 | mg/dL | [0.0-0.3] |
| Bilirubin Unconjugated | 0.6 | mg/dL | [0.0-1.1] |
| Bilirubin Delta | **0.7 H** | mg/dL | [0.0-0.2] |
| AST (SGOT) | **95 H** | IU/L | [3-74] |
| ALT (SGPT) | **58 H** | IU/L | [6-45] |
| Alkaline Phosphatase, Serum | **1047 H** | IU/L | [95-380] |
| Gamma Glutamyltransferase | 24 | IU/L | [15-85] |
| Albumin Level | 3.7 | g/dL | [2.9-5.5] |

| Procedure | | Units | Ref Range |
|---|---|---|---|
| White Blood Cell Count | 9.5 | K/mm3 | [6.0-17.5] |
| Platelet Count | **102 L** | K/mm3 | [250-600] |
| Hematocrit | 37.2 | % | [33.0-39.0] |
| Hemoglobin | 12.5 | g/dL | [10.5-13.5] |
| Red Blood Cell Count | 4.12 | M/mm3 | [3.70-5.30] |
| MCV | **90.3 H** | fl | [70.0-86.0] |
| MCH | 30.3 | pg | [23.0-31.0] |
| MCHC | 33.6 | % | [30.0-36.0] |
| RDW | **14.8** | % | [11.0-16.0] |
| Mean Platelet Volume | 10.9 | fl | [8.9-12.4] |
| Nucleated Red Cells | 0 | /100 WBC | |
| Polys, Absolute | 1938 | /mm3 | [1,500-5,000] |
| Lymphocytes, Absolute | 6384 | /mm3 | [1,500-8,500] |
| Monocytes, Absolute | **674 H** | /mm3 | [0-500] |
| Eosinophils, Absolute | **456 H** | /mm3 | [0-300] |
| Basophils, Absolute | 38 | /mm3 | [0-50] |
| Immature Granulocytes, Absolute i | 10 | /mm3 | [0-50] |
| Absolute Neutrophil Ct Cal | 1938 | /mm3 | [1,500-5,400] |
| Polys, % | 20.4 | % | [15.0-45.0] |
| Lymphocytes, % | 67.2 | % | [30.0-75.0] |
| Monocytes, % | 7.1 | % | [0.0-10.0] |
| Eosinophils, % | **4.8 H** | % | [0.0-4.0] |
| Basophils, % | 0.4 | % | [0.0-1.0] |
| Immature Granulocytes, % i | **0.1 H** | % | [<=0.0] |
| RBC /Platelet Morphology | Normal for Age | | |

| Procedure | | Units | Ref Range |
|---|---|---|---|
| Prothrombin Time | **18.1 H** | second(s) | [12.5-15.2] |
| International Normalized Ratio | 1.6 | | |

- Labs, 4/10-25/2013 (during his inpatient stay):

| | Collected Date | Collected Time | | | | |
|---|---|---|---|---|---|---|
| | 04/23/2013 | 05:00:00 | 04/14/2013 | 04:20:00 | 04/12/2013 | 05:00:00 |
| Procedure | | | | | Units | Ref Range |
| Vitamin D, 25-OH Total Level i | **7 L** | | **7 L** | | ng/mL | [30-100] |
| Vitamin D2 Level | <2 | | <2 | | ng/mL | |
| Vitamin D3 Level | 7 | | 7 | | ng/mL | |
| Vanilmandelic Acid | | | | 8 | ng/mL | [<=20] |
| Homovanillic Acid | | | | **58 H** | ng/mL | [<=30] |

| Collected Date | 04/23/2013 | 04/22/2013 | 04/21/2013 | | |
|---|---|---|---|---|---|
| Collected Time | 05:00:00 | 06:25:00 | 04:55:00 | | |
| Procedure | | | | Units | Ref Range |
| Phosphorus Serum | 5.2 | 4.9 | 5.2 | mg/dL | [3.9-6.5] |
| Bilirubin Total | | **4.3 H** | | mg/dL | [0.1-1.3] |
| Bilirubin Conjugated | 0.0 | 0.0 | 0.1 | mg/dL | [0.0-0.3] |
| Bilirubin Unconjugated | **2.6 H** | **2.7 H** | **2.9 H** | mg/dL | [0.0-1.1] |
| Bilirubin Delta | | **1.7 H** | | mg/dL | [0.0-0.2] |
| AST (SGOT) | **87 H** | | **107 H** | IU/L | [3-74] |
| ALT (SGPT) | **79 H** | | **88 H** | IU/L | [6-45] |
| Alkaline Phosphatase, Serum | **1229 H** | | **1342 H** | IU/L | [95-380] |
| Gamma Glutamyltransferase | 34 | | 37 | IU/L | [15-85] |
| Ammonia, Blood Venous | **53 H** | **82 H** | **105 H** | mcmol/L | [9-33] |
| Lactic Acid Blood, Venous | 0.9 | | 1.4 | MMOL/L | [0.5-2.2] |
| Albumin Level | 3.3 | 3.2 | 3.9 | g/dL | [2.9-5.5] |
| Vitamin A Level | **38 L** | | | mcg/L | [125-300] |
| Vitamin E Level | **2.5 L** | | | mg/L | [5.0-18.0] |
| Zinc Level | **1.53 Hf** | | | mcg/mL | [0.60-1.20] |
| Retinol Binding Protein | **<1.2 L** | | | mg/dL | [2.3-3.4] |

| Collected Date | 04/14/2013 | 04/14/2013 | 04/14/2013 | 04/14/2013 | | |
|---|---|---|---|---|---|---|
| Collected Time | 16:23:00 | 10:40:00 | 08:00:00 | 04:24:00 | | |
| Procedure | | | | | Units | Ref Range |
| Bilirubin Unconjugated | **6.9 H** | | | | mg/dL | [0.0-1.1] |
| AST (SGOT) | **91 H** | | | | IU/L | [3-74] |
| ALT (SGPT) | **71 H** | | | | IU/L | [6-45] |
| Alkaline Phosphatase, Serum | **729 H** | | | | IU/L | [95-380] |
| Gamma Glutamyltransferase | 45 | | | | IU/L | [15-85] |
| Ammonia, Blood Venous | **44 H** | | | | mcmol/L | [9-33] |
| Lactic Acid Blood, Venous | 1.4 | | | | MMOL/L | [0.5-2.2] |
| Albumin Level | **2.5 L** | | | | g/dL | [2.9-5.5] |

| Collected Date | 04/10/2013 | 04/09/2013 | 04/09/2013 | 04/09/2013 | | |
|---|---|---|---|---|---|---|
| Collected Time | 02:51:00 | 22:20:00 | 22:15:00 | 20:35:00 | | |
| Procedure | | | | | Units | Ref Range |
| Sodium Level | | **134 L** | 135 | 135 | mEq/L | [135-145] |
| Potassium Level | | 5.0 | 5.1 | 5.2 | mEq/L | [3.5-5.5] |
| Chloride Lvl | | 110 | | | mEq/L | [96-110] |
| CO2 Lvl | | **17 L** | | | mEq/L | [18-27] |
| Anion Gap | | 12.0 | | | mEq/L | [8.0-22.0] |
| Glucose Level | | | 89 | **120 H** | mg/dL | [60-105] |
| BUN | | 9 | | | mg/dL | [6-20] |
| Creatinine | | 0.2 | | | mg/dL | [0.1-0.4] |
| Total Protein | **4.1 L** | | | | g/dL | [5.6-7.2] |
| Calcium Ionized | **1.15 L** | | 1.28 | 1.27 | MMOL/L | [1.16-1.45] |
| Magnesium Serum | | 2.4 | | | mg/dL | [1.8-2.4] |
| Phosphorus Serum | | **3.4 L** | | | mg/dL | [3.9-6.5] |
| Ferritin Lvl | 93 | | | | ng/mL | [10-95] |
| Bilirubin Conjugated | **4.0 H** | | | | mg/dL | [0.0-0.3] |
| Bilirubin Unconjugated | **10.3 H** | | | | mg/dL | [0.0-1.1] |
| AST (SGOT) | **107 H** | | | | IU/L | [3-74] |
| ALT (SGPT) | **91 H** | | | | IU/L | [6-45] |
| Alkaline Phosphatase, Serum | **1270 H** | | | | IU/L | [95-380] |
| Uric Acid | 4.3 | | | | mg/dL | [2.0-6.0] |
| Gamma Glutamyltransferase | | 54 | | | IU/L | [15-85] |
| Triglycerides i | **47 L** | | | | mg/dL | [60-135] |
| Amylase Serum | | <30 | | | IU/L | [0-115] |
| Lipase Level | | **<10 L** | | | IU/L | [25-120] |
| Lactate Dehydrogenase | 810 | | | | IU/L | [425-975] |
| Lactic Acid Blood, Venous | | | 2.1 | **3.1 H** | MMOL/L | [0.5-2.2] |
| Albumin Level | **2.3 L** | | | | g/dL | [2.9-5.5] |
| C Reactive Protein | | <0.8 | | | mg/dL | [<=0.8] |

| Collected Date | 04/25/2013 | 04/24/2013 | 04/23/2013 | 04/22/2013 | | |
|---|---|---|---|---|---|---|
| Collected Time | 04:30:00 | 05:30:00 | 05:00:00 | 06:25:00 | | |
| Procedure | | | | | Units | Ref Range |
| Prothrombin Time | **25.5 H** | **25.3 H** | **24.1 H** | **24.4 H** | second(s) | [12.5-15.2] |
| International Normalized Ratio | 2.5 | 2.4 | 2.3 | 2.3 | | |
| APTT | **>150 Cf** | **62 H** | **68 H** | **69 H** | second(s) | [25-35] |
| Fibrinogen Level | **137 L** | **125 L** | **125 L** | **139 L** | mg/dL | [230-450] |
| Thrombin Time | | | | **20 H** | second(s) | [14-17] |

| Collected Date | 04/21/2013 | 04/20/2013 | 04/19/2013 | 04/18/2013 | | |
|---|---|---|---|---|---|---|
| Collected Time | 04:55:00 | 04:55:00 | 04:40:00 | 05:46:00 | | |
| Procedure | | | | | Units | Ref Range |
| Prothrombin Time | **23.7 H** | **23.6 H** | **23.7 H** | **18.3 H** | second(s) | [12.5-15.2] |
| International Normalized Ratio | 2.2 | 2.2 | 2.2 | 1.6 | | |
| APTT | **>150 Cf** | **77 H** | **52 Hf** | **44 H** | second(s) | [25-35] |
| Fibrinogen Level | **159 L** | **162 L** | **156 L** | **204 L** | mg/dL | [230-450] |
| Thrombin Time | **79 H** | | 17 | | second(s) | [14-17] |

| Collected Date | 04/14/2013 | 04/13/2013 | 04/13/2013 | 04/12/2013 | | |
|---|---|---|---|---|---|---|
| Collected Time | 04:20:00 | 16:45:00 | 03:45:00 | 20:00:00 | | |
| Procedure | | | | | Units | Ref Range |
| Prothrombin Time | **21.0 H** | **38.4 Cf** | **33.7 Cf** | **31.8 Cf** | second(s) | [12.5-15.2] |
| International Normalized Ratio | 1.9 | 4.1 | 3.5 | 3.2 | | |
| APTT | **40 H** | **62 H** | **54 H** | **55 H** | second(s) | [25-35] |
| Fibrinogen Level | **183 L** | **89 L** | **121 L** | **134 L** | mg/dL | [230-450] |
| Thrombin Time | | **20 Hf** | | | second(s) | [14-17] |

| Collected Date | 04/10/2013 | 04/10/2013 | | |
|---|---|---|---|---|
| Collected Time | 11:00:00 | 02:51:00 | | |
| Procedure | | | Units | Ref Range |
| Prothrombin Time | **46.3 Cf** | **48.6 Cf** | second(s) | [12.5-15.2] |
| International Normalized Ratio | 5.3 | 5.6 | | |
| APTT | **69 Hf** | **75 H** | second(s) | [25-35] |
| Fibrinogen Level | **80 L** | **60 L** | mg/dL | [230-450] |
| Thrombin Time | **19 Hf** | | second(s) | [14-17] |

| Collected Date | 04/14/2013 | 04/13/2013 | 04/13/2013 | | |
|---|---|---|---|---|---|
| Collected Time | 10:40:00 | 16:45:00 | 11:16:00 | | |
| Procedure | | | | Units | Ref Range |
| Respiratory Viral PCR Battery Result i | Negative | | | | |
| Specimen, Viral culture (VCS) | | | STOOL f | | |
| Result, Prelim culture (VCS) | | | SEE COMMENTS f | | |
| Result, Final culture (VCS) | | | SEE COMMENTS f | | |
| Specimen, Hepatitis B Viral DNA Qnt | | Serum f | | | |
| Hepatitis B DNA Quant | | SEE COMMENTS f | | | |
| Hepatitis B Result PCR Quant | | None detected. | | IU/mL | [NDET] |
| HBV DNA IU/mL (Log 10) | | Not Calculated | | | |

| | | | | | |
|---|---|---|---|---|---|
| Collected Date | 04/11/2013 | 04/11/2013 | | | |
| Collected Time | 16:40:00 | 00:20:00 | | | |
| Procedure | | | | Units | Ref Range |
| Human Herpes Virus 6 by PCR | SEE COMMENTS f | | | | |
| HH6 Result PCR Quant | None detected. | | | copies/mL | [NDET] |
| HH6 DNA Copies/mL (Log 10) | Not Calculated | | | | |
| Specimen, HSV PCR | Plasma f | | | | |
| Result, HSV PCR | None detected. | | | copies/mL | [NDET] |
| HSV DNA Copies/mL (Log 10) | Not Calculated | | | | |
| HSV Interpretation PCR Quant | SEE COMMENTS f | | | | |
| Hepatitis B surface antigen | See Below | | | | |
| Hepatitis B Surface Antibody | See Below | | | | |
| Hepatitis C Antibody | See Below | | | | |
| HIV Ag and Ab Interpretation | SEE COMMENTS f | | | | |
| HIV Ag and Ab Result | Nonreactive | | | | [NREAC] |
| CMV IgG | | See Below | | | |
| CMV IgG | | Positive | | | [Negative] |
| CMV IgM | | See Below | | | |
| CMV IgM | | Negative | | | [Negative] |
| EBV IgG result | | Positive | | | [Negative] |
| EBV IgM result | | Negative | | | [Negative] |
| Epstein Barr Nuclear Antibody | | Positive | | | [Negative] |
| Epstein Barr Early Antigen Antibody | | Negative | | | [Negative] |
| Interpretation, EBV | | See Below | | | |

04/11/2013 16:40:00 Hepatitis B surface antigen
Nonreactive; No evidence of hepatitis B surface antigen. HBsAg can be detected in serum 30-60 days after exposure to hepatitis B virus.

04/11/2013 16:40:00 Hepatitis B Surface Antibody
Nonreactive, <5.0 mIU/mL Antibody to surface antigen is the major protective antibody against hepatitis B virus. It is detected following recovery from HBV, and inoculation with hepatitis B vaccine. A nonreactive antibody test indicates lack of immunity to hepatitis B.

04/11/2013 16:40:00 Hepatitis C Antibody
Nonreactive. Approximately 45% of patients are positive for anti-HCV within 6 weeks of illness; the remainder may not have detectable antibody until 6 months later. Nonreactive HCV tests should be repeated for up to one year if clinical symptoms suggest HCV infection.

04/11/2013 00:20:00 CMV IgG
IgG antibody to CMV detected. This indicates a current or past CMV infection.

04/11/2013 00:20:00 CMV IgM
No significant level of detectable CMV IgM antibody. No current infection with CMV.

04/11/2013 00:20:00 Interpretation, EBV
Evidence of past EBV infection.

| | | | | | |
|---|---|---|---|---|---|
| Collected Date | 04/10/2013 | 04/10/2013 | 04/10/2013 | | |
| Collected Time | 11:00:00 | 04:45:00 | 02:51:00 | | |
| Procedure | | | | Units | Ref Range |
| Rotavirus | | Negative | | | [Negative] |
| Specimen, CMV PCR | Plasma f | | | | |
| CMV by PCR, QUANTitative | SEE COMMENTS f | | | | |
| CMV PCR Quant Result (IU/mL) | None detected. | | | IU/mL | [NDET] |
| CMV DNA IU/mL (Log 10) | Not Calculated | | | | |
| Specimen, Epstein Barr Virus PCR | Plasma f | | | | |
| EBV quant PCR blood/CSF | SEE COMMENTS f | | | | |
| EBV PCR Quant Result (Copies/mL) | None detected. | | | copies/mL | [NDET] |
| EBV DNA Copies/mL (Log 10) | Not Calculated | | | | |
| Hepatitis A IgM | | | See Below | | |
| Hepatitis A Total Antibody | | | See Below | | |

04/10/2013 02:51:00 Hepatitis A IgM
Nonreactive. A nonreactive IgM anti-HAV test and a positive Total anti-HAV is consistent with past infection or immunization with hepatitis A vaccine. This indicates immunity to HAV.

04/10/2013 02:51:00 Hepatitis A Total Antibody
Reactive. Total antibody to hepatitis A virus tests for immunoglobulins G and M. A reactive test result is an indicator of recent, past infection or immunization with hepatitis A vaccine and immunity to HAV. IgM anti-HAV will be performed. The presence of maternal antibodies could also cause a reactive test, and may not indicate long term immunity.

| | | | |
|---|---|---|---|
| Collected Date | 04/10/2013 | | |
| Collected Time | 02:51:00 | | |
| Procedure | | Units | Ref Range |
| Acetaminophen Level | <10 | mcg/mL | [10-20] |

| | | | | | |
|---|---|---|---|---|---|
| Collected Date | 04/12/2013 | 04/12/2013 | 04/11/2013 | | |
| Collected Time | 10:25:00 | 05:00:00 | 16:40:00 | | |
| Procedure | | | | Units | Ref Range |
| Anti Nuclear Antibody Screen | Negative | | | | [Negative] |
| Aspergillus galactomannan Index | | 0.073 f | | | [0.000-0.499] |
| Result, Aspergillus galactomannan EIA | | SEE COMMENTS f | | | |
| Leptospira Antibody | | Negative f | | | [Negative] |
| Liver Kidney Microsomal Antibody | | | Negative | | [Negative] |

| Procedure | | Units | Ref Range |
|---|---|---|---|
| Immunoglobulin G Level | 479 | mg/dL | [156-829] |
| Anti Smooth Muscle Antibody | Negative | | [Negative] |

- Urine bile acid profile, 4/22/2013:

## Bile Acids

**FAB No:** 12089

**Client Patient Chart/Ref#:** 1275567

**Referring Physician/Lab:**

Seattle Childrens Hopsital
Sendout Lab - A-6901
4800 Sand Point Way NE
Seattle, WA 98105

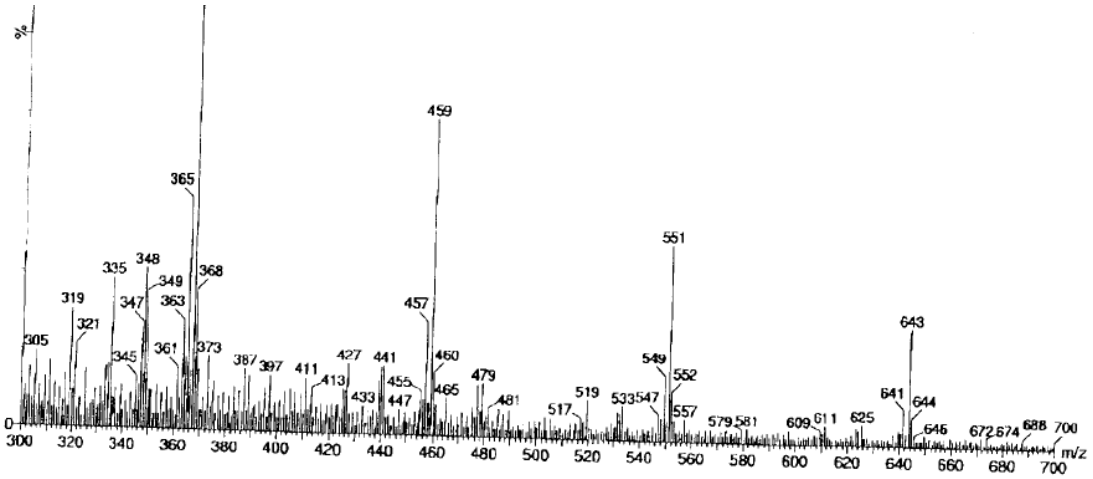
**Sample received: April 24th 2013**
**Urine volume: 11.6 mL**

A urine sample, collected April 22nd 2013 from the above named patient, has been analyzed using fast atom bombardment ionization mass spectrometry for the detection of potential inborn errors in the cholesterol-bile acid biosynthetic pathway.

The negative ion FAB-MS spectrum of the urine from this patient is attached and is unremarkable. The profile indicates a normal urinary bile acid profile, with no evidence for a cholestatic condition. Based upon this analysis, there is no indication for a defect in the cholesterol-bile acid biosynthetic pathway, or any abnormality in peroxisomal beta-oxidation of bile acid intermediates.

If you require any further information, please contact me at 513-636-4548.

*This test was developed and its performance characteristics were determined and validated by the Clinical Mass Spectrometry Laboratory at Cincinnati Children's Hospital Medical Center. It has not been cleared or approved by the U.S. Food and Drug Administration. This laboratory is certified under the Clinical Laboratory Improvement*




- JaundiceChip Resequencing Array, 4/15/2013:

HL-13-00155

# JaundiceChip Resequencing Array

Collection Date and Time: 4/15/2013 4:27 PM
**Laboratory Accession # : HL-13-00155**
Specimen : 2.0mL blood/good
Ordering Physician : Phelan, Rachel
Received Date and Time : 4/17/2013 12:43 PM
Ethnicity : Asian

## Genes Tested:

***SERPINA1* (a1-antitrypsin deficiency)**
**Allele 1**: No Mutation Identified
**Allele 2**: No Mutation Identified

***JAG1* (Alagille syndrome)**
**Allele 1**: No Mutation Identified
**Allele 2**: No Mutation Identified

***ATP8B1* (PFIC1)**
**Allele 1**: 2364 A>C (E788D)
**Allele 2**: No Mutation Identified

***ABCB11* (PFIC2)**
**Allele 1**: No Mutation Identified
**Allele 2**: No Mutation Identified

***ABCB4* (PFIC3)**
**Allele 1**: No Mutation Identified
**Allele 2**: No Mutation Identified

## RESULT: Variant of uncertain clinical significance

We have completed the sequence analysis of the five most common genes associated with heritable liver disease in childhood. This patient is heterozygous for a previously unreported sequence variant in the *ATP8B1* gene. Its relationship with the patient's liver disease is uncertain at this time. This patient does not have an identified mutation in any of the other genes related to heritable liver disease specified above.

**CLINICAL SIGNIFICANCE**

We have no clinical history on this patient. This patient is heterozygous for a missense variant of uncertain clinical significance in the *ATP8B1* gene. The presence of this variant in this individual neither confirms nor excludes this variant as a causative factor in the patient's disease.

**GENETIC SIGNIFICANCE**

This variant, 2364 A>C, results in the substitution of aspartic acid for glutamic acid at amino acid 788. This amino acid sequence is not conserved across mammalian species examined in our laboratory. Our laboratory has not identified other symptomatic patients with this sequence variant. This variant is listed in the EntrezGene dbSNP (rs#77317429), but no minor allele frequency is available. According to the Grantham scale, this amino acid substitution is predicted to be conservative. In addition, through the use of mutation prediction software, this amino acid substitution is predicted to be "tolerated" and "benign". The accuracies of various in silico prediction methods are in the range of 0.60-0.82 (Thusberg et al 2011). **Based on all of the available information, we are unable to predict whether this sequence variant represents an unrecognized disease-causing mutation or a benign polymorphism.**

Genetic counseling is recommended for this family.

**CLINICAL RECOMMENDATIONS**

At this point, we recommend completion of the diagnostic evaluation as directed by the referring physician.

If you have any questions about this test result or its implications, please contact the Molecular Genetics Laboratory at 513-636-4474.

- Fecal elastase, 4/10/2013:

| | | | | |
|---|---|---|---|---|
| Collected Date | 04/10/2013 | 04/10/2013 | | |
| Collected Time | 10:17:00 | 04:45:00 | | |
| Procedure | | | Units | Ref Range |
| Pancreatic Elastase 1 (PE1) | | <50.0 | mcgE1/g | |
| Stool Alpha 1 Antitrypsin | <0.5 | | mg/g | [0.1-3.0] |
| Stool Occult Blood | | Negative | | [Negative] |
| Pancreatic Elastase 1 Interp, Stool | | See Comments f | | |

04/10/2013 04:45:00 Pancreatic Elastase 1 Interp, Stool:
Severe exocrine pancreatic insufficiency*

*Please Note: This specimen was creamy/mucous-like in consistency. A formed stool should be tested for more accurate results.
Reference Values For Pancreatic Elastase in Stool

200 to >500 ug Elastase/g stool = Normal

100 to 200 ug Elastase/g stool = Moderate to slight exocrine pancreatic insufficiency

<100 ug Elastase/g stool = Severe exocrine pancreatic insufficiency

- Urinalysis, 4/9/2013:

| | | | | | |
|---|---|---|---|---|---|
| Collected Date | 04/22/2013 | 04/11/2013 | 04/09/2013 | | |
| Collected Time | 06:15:00 | 21:00:00 | 23:05:00 | | |
| Procedure | | | | Units | Ref Range |
| Urine Color | | | Orange | | |
| Urine Specific Gravity | | | 1.021 | | [1.001-1.035] |
| Urine pH | | | <= 5.0 | | [4.5-8.0] |
| Urine Leukocyte Esterase | | | Negative | | [Negative] |
| Urine Nitrite | | | Negative | | [Negative] |
| Urine Protein Qualitative | | | **Trace** * | | [Negative] |
| Urine Glucose Qualitative | | | Negative | | [Negative] |
| Urine Ketones i | | | **Trace** * | | [Negative] |
| Urine Urobilinogen | | | **1.0** | | |
| Urine Bilirubin | | | See Note | | [Negative] |
| Urine Occult Blood | | | Negative f | | [Negative] |
| Urine Reducing Substances | | **Trace** * | | | [Negative] |
| Urine Collection Type | | | I/O Cath | | |
| Urine Microscopic | | | Done | | |
| Urine Cultured? | | | Yes | | |
| Urine Volume | | | 3.0 | mL | |
| Urine RBC | | | 0-2 | /HPF | [None] |
| Urine WBC | | | **0-5** * | /HPF | [None] |
| Urine Bacteria | | | Not Seen | /HPF | |
| Urine Epithelial Cells | | | **Few** * | /LPF | [None] |
| Renal Tubular Cells | | | 1-5 | | |
| Urine Amorphous Crystals | | | **Few** * | /LPF | |
| Hyaline Casts | | | Rare | /LPF | |
| Notes On UA | | | See Note f | | |
| Urine Bile Acid Profile i | See Below | | | | |

- Urine culture, 4/9/2013:

PROCEDURE: Urine Culture
SOURCE: U Foley Cath
FREE TEXT SOURCE: Plated by core

COLLECTED: 04/09/2013 23:05
RECEIVED: 04/10/2013 04:48
ACCESSION: 13-099-1240

*** FINAL REPORT ***

Final Report
Between 1,000 and 10,000 cfu/ml Escherichia coli
<1,000 cfu/ml Gram Negative Flora

Verified:04/11/2013 18:17

*** PRELIMINARY REPORT ***

Preliminary Report
Between 1,000 and 10,000 cfu/ml Escherichia coli
<1,000 cfu/ml Gram Negative Flora
, Final Report to Follow.

Verified:04/11/2013 02:51

*** SUSCEPTIBILITY RESULTS ***

| | Escherichia coli |
|---|---|
| | Interp |
| Augmentin | Suscept |
| Ampicillin | Suscept |
| Amikacin | Suscept |
| Ceftazidime | Suscept |
| Ceftriaxone | Suscept |
| Cefuroxime IV Use | Suscept |
| Cefuroxime Oral use | Suscept |
| Cefazolin | Suscept U |
| Cefepime | Suscept |
| Nitrofurantoin | Suscept |
| Gentamicin | Suscept |
| Meropenem | Suscept |
| Trimethoprim/Sulfamethoxazole | Suscept |
| Pip/Taz | Suscept |
| Comments(1) | Yes |

Imaging Reports:

- Chest and abdominal X-rays, 4/9/2013:

Impression:

1. Colonic thumbprinting could indicate infectious or inflammatory colitis. An infiltrative process cannot be completely excluded on the basis of these radiographs.
2. No gross evidence of small bowel obstruction or ileus, although a paucity of gas in the bowel makes evaluation somewhat difficult.
3. Possible mild intraperitoneal free fluid.

- Abdominal ultrasound, 4/9/2013:

Findings:
The liver is normal in size and echogenic texture. There is no focal hepatic mass. The gallbladder is normal in size. Gallbladder wall demonstrates mild diffuse thickening. There is no cholelithiasis. There is no pericholecystic fluid. The common bile duct measures 1.2 mm. There is no intrahepatic biliary dilatation.

There is mild splenomegaly, measuring 8.5 cm. (normal range 5.2-7.0 cm). Splenic echogenic texture is normal.

The pancreas is poorly visualized, but demonstrates no gross abnormality. The right kidney measures 5.6 cm in length. The left kidney measures 6.0 cm in length. Normal renal length for age is 6.2 cm +/- 1.3 cm. There is trace bilateral pelviectasis. Renal parenchyma is unremarkable. The urinary bladder is decompressed, but demonstrates no focal abnormality. There is no distal ureteral distention.

Visualized images of the abdominal aorta and IVC are unremarkable.

There is a moderate amount of intraperitoneal free fluid.

Impression:

1. Moderate intraperitoneal free fluid.
2. Mild diffuse gallbladder wall thickening without cholelithiasis or pericholecystic fluid, likely related to volume overload with third spacing of fluid.
3. Mild splenomegaly.
4. Trace bilateral pelviectasis (SFU grade 1).

- Abdominal ultrasound with Doppler, 4/10/2013:

Impression:

1. Edematous appearance to the liver with interval development of multiple small echogenic foci throughout the right lobe of the liver, the latter findings suggesting a diffuse inflammatory etiology including septic emboli. Differential diagnosis for the echogenic hepatic foci includes metastatic lesions and the possibility of hemorrhagic infarcts.

2. Persistent splenomegaly.
3. Contracted gallbladder likely reflecting nonfasting state.
4. Trace central pelviectasis right kidney, decreased from prior.
5. Small amount of anechoic free intraperitoneal fluid.
6. Abnormally elevated resistive indices measuring 1.0 throughout the liver.
7. Recanalization/patency of the umbilical vein, a finding which, in conjunction with splenomegaly, suggests portal hypertension.

- Transthoracic echocardiogram, 4/10/2013:

Summary:

1. No intracardiac vegetation.
2. No intracardiac shunting.
3. Normal valvular structure and function.
4. No pericardial effusion.
5. Indirect markers for impaired relaxation of the left ventricle include abnormal annular and septal tissue Doppler patterns, 'a' wave reversal in pulmonary venous Doppler.
6. There is mild increase in velocity across aortic, pulmonary and mitral valves, indicating hyperdynamic or fluid overloaded status.

- CT angiogram of the abdomen, thorax, and pelvis, 4/11/2013:

Impression:

1. Diffuse hazy mesentery could represent edema in the setting of stigmata of portal hypertension including recanalized paraumbilical vein, minimal gastric varices and moderate splenomegaly as well as diffuse subcutaneous edema. Additional diagnostic considerations include infectious processes of the mesentery given coexisting bowel wall thickening and mucosal hyperenhancement of the sigmoid colon or possibly an infiltrative process of the mesentery such as lymphoma or leukemia.

2. Numerous small hypodense hepatic lesions are too small to further characterize and are thus indeterminate. However, diagnostic considerations should include infection or metastases. Fat density is seen within several lesions and therefore fat containing metastases such as liposarcoma can produce this pattern although there is no evidence of primary tumor on the current exam, benign fat-containing lesions such as angiomyolipomas, xanthomas could be included in the differential of fat containing lesions.

3. Sigmoid colon and rectal wall thickening with mucosal hyperenhancement could represent infectious colitis.

4. Bilateral dependent airspace opacities are most compatible with atelectasis, although infectious etiology cannot be completely excluded.

Pathology Reports:

- Liver biopsy, 4/18/2013:

Gross Description:

Received fresh labeled LIVER BIOPSY is a single needle core biopsy fragment measuring 0.1 cm in diameter x 1.3 cm in length. The specimen is placed in formalin then wrapped and submitted in its entirety in a single cassette designated A1. [MA]

Microscopic Description:

H&E stained sections show a cirrhotic liver with bridging fibrosis and marked bile ductular proliferation. There is moderate inflammation in the portal tracts with scattered neutrophils, lymphocytes, and eosinophils. Bile ducts are present, although difficult to see with the marked portal expansion and ductal proliferation. Lobular inflammation is variable and predominantly lymphocytic in nature. There is variable cholestasis, with some nodules showing canalicular plugging and others showing only mild intracellular cholestasis. Acidophil bodies are readily identified. Hepatocytes are swollen with lacy cytoplasm and reactive appearing nucleoli. Rare areas show features of early hepatocyte necrosis with the blurring of cell junctions and hypereosinophilia. There is patchy mild macrosteatosis (<5%). A trichrome stain highlights the presence of bridging fibrosis, as well as pericellular fibrosis. Periodic acid shift stains with a without diastase reveal no diastase resistant globules.

Slide and stain summary: H&E 3 slides; PAS 1 slide; PAS-D 1 slide; trichrome 1 slide

Electron Microscopy Findings:

Toluidine blue stained thick sections reveal cirrhotic liver with both periportal and pericellular fibrosis. There is mild portal inflammation. Examination by transmission electron microscopy shows mild cholestasis, and occassional hepatocytes with membranous debris but no evidence of storage disease. Bile canaliculi show normal microvillous architecture, without Bylers-type bile. Probable peroxisomes are identified. Mitochondria are present in normal numbers, and are without circular or paracrystalline arrays of cristae.

Final Diagnosis:

LIVER, NATIVE, CORE BIOPSY: CIRRHOSIS AND BILE DUCTULAR PROLIFERATION, SEE COMMENT

**Comment**

The biopsy shows established cirrhosis with marked bile ductular proliferation and variable cholestasis. There is ongoing hepatocyte injury, with scattered apoptotic cells, as well. PASd stain reveals no evidence of alpha-1 antitrypsin deficiency, and no abnormal mitochondria, bile or storage-type material are seen on electron microscopy. A definitive etiology for the patient's chronic liver failure is not evident from this biopsy.

- The pathology slides were reviewed as consults at Massachusetts General Hospital. The MGH review clarified the diagnosis, with the following interpretation:

**FINAL PATHOLOGIC DIAGNOSIS**

Micronodular cirrhosis with cholestasis, moderate macrovesicular steatosis and PAS/D positive globules. See note

Note:
The trichrome stain shows nodules composed of hepatocytes and surrounded by fibrosis. A keratin 19 immunohistochemical stain was evaluated. The stain shows marked bile ductular reduplication. In addition, native bile ducts are also identified arguing against a diagnosis of paucity of bile ducts. An iron stain is negative.

A copper stain shows a diffuse increase in intracellular copper. However this finding does not necessarily indicate a diagnosis of Wilson's disease. Elevated copper levels are often found in other chronic diseases of the liver particularly in children as well as in individuals with significant fibrosis as in this instance. Nevertheless, if clinically indicated, further evaluation to exclude a diagnosis of Wilson's disease is suggested.

A PAS/D stain shows numerous periseptal hepatocellular globules. However, these globules are not typical for out of A1AT deficiency. Nevertheless, an immunohistochemical stain for AIAT also shows the presence of periseptal globules. This data raises the possibility of A1AT deficiency and appropriate investigations to either support or exclude this possibility are required. The diagnosis of other heritable metabolic disorders would require appropriate testing and cannot be excluded based on this material alone.

Treating Physician's Assessment and Plan:
Jianhua is an 10 month-old boy that presented in April with fulminant hepatic failure. He has subsequently been diagnosed with chronic liver failure of undetermined etiology. Histologically, he has been shown to have a micronodular cirrhosis with marked bile ductular proliferation. An extensive work-up to date has been largely unremarkable, with the exception of a single allele mutation in the PFIC1 gene of unknown clinical significance. A fecal elastase revealed "severe exocrine pancreatic insufficiency," though this may have been felt to be a false positive because of loose stools (as pancreatic enzyme supplements were not started). Recently, his liver transplant status has been changed from "active" to "hold", as he is so stable. It is anticipated by his treating hepatology team that he will at some point need transplantation, though the timing of this is not clear. The family is seeking a Best Doctors consultation regarding the diagnosis and treatment plan.

It is my opinion that though this child presented symptomatically with what appeared to be fulminant liver failure, the cirrhosis and history suggest that this child actually had a chronic liver condition likely from birth which finally clinically became apparent with the need for admission to the hospital. I will respond to the queries as follows:

**Questions:**

**1) What is your overall assessment of the patient's condition?** It is my opinion that this child has a chronic liver disorder, has cirrhosis and though currently compensated will require a liver transplant in the future as a result.

**a) What is the differential diagnosis for this patient?** A very thorough work-up of this child has failed to determine a precise diagnosis. Very often when the liver is already cirrhotic it is impossible to detect what the etiology of the cirrhosis was. The treatment will not necessarily change, but it would be helpful to confirm a diagnosis. The usual viral, metabolic, and genetic causes have all been searched for. There are several clues however that make me very suspicious of the diagnosis of progressive familial intrahepatic cholestasis (PFIC) type 1- the gene chip test from Cincinnati suggested a PFIC 1 abnormality of uncertain significance. Also, there is a normal GGT with elevated alkaline phosphatase and bile duct proliferation on liver biopsy which is seen with PFIC. Also the abnormal exocrine pancreatic function was felt to be a lab error- however, in PFIC type 1 pancreatic insufficiency can be seen.

**b) The family is particularly concerned about the possibility of NICCD (neonatal intrahepatic cholestasis caused by citrin deficiency). This is based upon their observations that: a) his birth weight was the lowest of their 4 children, b) the presence of fatty stool, c) his "improved condition after changing from soy to Pregestamil formula," and d) "his matching lab results of the AST more elevated than the ALT, high alkaline phosphatase, etc…" The parents ask if Jianhua might have NICCD, and why or why not?**

Citrin deficiency can manifest in newborns as neonatal intrahepatic cholestasis caused by citrin deficiency (NICCD). Children younger than age one year have growth retardation with transient intrahepatic cholestasis, hepatomegaly, diffuse fatty liver and parenchymal cellular infiltration associated with hepatic fibrosis, variable liver dysfunction, hypoproteinemia, decreased coagulation factors, hemolytic anemia, and/or hypoglycemia. Although NICCD is generally not severe and symptoms often resolve by age one year with appropriate treatment, some infants succumb to infection and liver cirrhosis and others require liver transplantation. The diagnosis of citrin deficiency is suspected from clinical and biochemical findings (in general, increased blood or plasma concentration of ammonia, plasma or serum concentration of citrulline and arginine, plasma or serum threonine-to-serine ratio, and serum concentration of pancreatic secretory trypsin inhibitor [PSTI]). Identification of biallelic mutations in SLC25A13, the only gene in which mutations are known to cause citrin deficiency, confirms the diagnosis. Citrin deficiency is inherited in an autosomal recessive manner. When both parents are carriers, each sib of an affected individual has, at conception, a 25% chance of being affected, a 50% chance of being an asymptomatic carrier, and a 25% chance of being unaffected and not a carrier. When one parent is a carrier and the other parent has two mutated SLC25A13 alleles, each sib of an affected individual has, at conception, a 50% chance of being affected and a 50% chance of being an asymptomatic carrier. Carrier testing for at-risk relatives and prenatal testing for pregnancies at increased risk are possible if the disease-causing mutations in the family are known. It might be worthwhile to test both Jianhua and the parents for this mutation.

**c) What do you think is the most likely diagnosis and why?** I am concerned for both PFIC type 1 and NICCD as possible explanations for the liver disease observed. I think it appropriate for further genetic testing to assess for PFIC type 1 genes and NICCD mutations in both parents and the child.

**2) What additional diagnostic testing do you recommend?** Genetic testing of both parents and the child. My concern for PFIC type 1 is further substantiated by a genetic database query suggesting that the mutation seen in this child on the jaundice chip might be possibly damaging.

**3) The parents ask if the specialist can estimate the percentage of living liver cells from his liver biopsy?** Obviously there are adequate liver cells currently as the child is compensated. Livers can regenerate so this is not the major issue for Jianhua. The issue is the extent of scarring in the liver which has resulted already in cirrhosis- this will cause problems with blood flow through the liver resulting likely in the development of portal hypertension again as was witnessed with the ascites and edema. With portal hypertension there will also be the risk of bleeding from esophageal varices and the development of thrombocytopenia (low platelets) from splenomegaly and hypersplenism.

**4) Based upon his significant improvement, can the natural history of this condition be predicted moving forward?** It is encouraging that the condition has stabilized, but I believe careful and frequent evaluations and monitoring of Jianhua are necessary. The disease may progress and the child will again develop signs of decompensated liver disease. This may be triggered by a simple cold, for example.

**a) His family asks if it is possible to determine the likelihood that he will or will not need a liver transplant?** While it is impossible to make an accurate prediction, based upon the extent of liver injury at such a young age unless there is complete cessation of the process, it is highly likely this child will eventually require a liver transplant.

**5) Jianhua has not been gaining weight well since discharge from the hospital (though the resolution of his fluid retention is likely at least in part the cause). His parents ask how weight gain can be optimized?** Supplementation as is being done with fat soluble vitamins and MCT containing formula to allow ready absorption of fat is the appropriate nutritional intervention.

**a) They note that they tried increasing his protein by giving him more chicken, but this seemed to make his pruritis worse. Green peas led to edema. Are any specific dietary measures recommended?** No- while the family may have noted the chicken and peas resulting in these symptoms, it is unlikely they were directly related. There is nothing in the medical literature pertaining to peas resulting in edema. A balanced nutritional diet is appropriate for this child to maximize growth and development.

**6) What treatment options exist?** For NICCD supplementing diet with fat-soluble vitamins and use of lactose-free formula (in those with galactosemia) or formulas containing medium-chain triglycerides is appropriate. This would also be appropriate for PFIC type 1 or other similar liver disorders. Careful monitoring for the need for liver transplant would of course be indicated.

**a) Apart from liver transplant, what medical and/or nutritional therapies are indicated?** See above- these nutritional therapies are supportive not curative.

**b) What are the relative advantages and disadvantages of each option?** Again, liver transplant is likely the only treatment with potential for cure at this point. The other therapies are merely supportive.

**c) How would you recommend proceeding in this case?** As previously mentioned, genetic testing for both PFIC type 1 and NICCD in my opinion are very appropriate.

**7) How would you recommend following this patient?** Frequent assessment of liver function, growth, development, electrolytes, stooling, vitamin levels are all appropriate and monitoring for development of portal hypertension with platelet count, and abdominal ultrasound periodically.

**8) If not addressed by the questions above, please provide any further recommendations that you believe will aid in understanding this patient's findings or in guiding future therapeutic decisions.** Again, further genetic testing and frequent monitoring of this child will provide the best long-term outcome.

**9) Please provide scientific references that may be helpful to the treating physicians and the patient, and/or that lend support to your recommendations.**

**Kobayashi K, Saheki T, Song YZ. Citrin Deficiency. In: Pagon RA, Adam MP, Bird TD, Dolan CR, Fong CT, Stephens K, editors. GeneReviews™ [Internet]. Seattle (WA): University of Washington, Seattle; 1993-2013.**

**Suporn Treepongkaruna, Suttiruk Jitraruch, Porawee Kodcharin, Dussadee Charoenpipop, Pim Suwannarat1, Paneeya Pienvichit, Keiko Kobayashi and Duangrurdee Wattanasirichaigoon. Neonatal intrahepatic cholestasis caused by citrin deficiency: prevalence and SLC25A13 mutations among Thai infants. BMC Gastroenterology 2012, 12:141 doi:10.1186/1471-230X-12-141**

**Raffaella A. Morotti, Frederick J. Suchy, Margret S. Magid. Progressive Familial Intrahepatic Cholestasis (PFIC) Type 1, 2, and 3: A Review of the Liver Pathology Findings. Semin Liver Dis 2011; 31(1): 003-010 DOI: 10.1055/s-0031-1272831**

**Pawlikowska L, Strautnieks S, Jankowska I, Czubkowski P, Emerick K, Antoniou A, Wanty C, Fischler B, Jacquemin E, Wali S, Blanchard S, Nielsen IM, Bourke B, McQuaid S, Lacaille F, Byrne JA, van Eerde AM, Kolho KL, Klomp L, Houwen R, Bacchetti P, Lobritto S, Hupertz V, McClean P, Mieli-Vergani G, Shneider B, Nemeth A, Sokal E, Freimer NB, Knisely AS, Rosenthal P, Whitington PF, Pawlowska J, Thompson RJ, Bull LN. Differences in presentation and progression between severe FIC1 and BSEP deficiencies. J Hepatol. 2010 Jul;53(1):170-8. doi: 10.1016/j.jhep.2010.01.034.**

Please feel free to contact me if there are any additional questions regarding his condition I can answer.

Sincerely,

Philip Rosenthal, MD
Professor of Pediatrics & Surgery

**Addendum:**

I have been asked to clarify a few points.

1. **You mention that additional genetic testing for PFIC-1 would be appropriate. Is there more detailed testing available than that offered by the JaundiceChip? If so, could you please elaborate?**

Here is the information from Cincinnati Children's Hospital about the jaundice chip-

**“**Inherited intrahepatic cholestasis is a heterogeneous group of disorders typically presenting as neonatal jaundice and leading to persistent liver dysfunction in children and adults. Some types of cholestasis present with a clear pattern of extrahepatic symptoms and laboratory and pathologic findings, but there remains considerable clinical overlap. Five genes represent the most common heritable causes of cholestatic liver disease in young children: JAG1 (Alagille syndrome), ATP8B1 (PFIC1), ABCB11 (PFIC2), ABCB4 (PFIC3), and SERPINA1 (alpha-1-antitrypsin deficiency). The Molecular Genetics Laboratory at Cincinnati Children's Hospital Medical Center offers diagnostic testing for these individual genes, or in cases of clinical overlap, the innovative Jaundice Chip resequencing assay , the first chip of its kind, which provides rapid and cost effective analysis of all five genes.**”**

The jaundice chip is a fast and cost effective means to assess for the most common causes of neonatal cholestasis. It is not a replacement for formal gene sequencing for rarer causes of cholestasis (for example NICCD). My recommendation is for formal gene sequencing of Jianhua and his parents. Consultation with a Geneticist should be able to direct appropriate additional genetic testing especially for NICCD and a more thorough evaluation for PFIC.

2. **He is currently receiving Pregestimil formula. As you gathered, the family is particularly interested in whether any nutritional changes are warranted. Would you recommend any change away from this formula at this point?**

Pregestemil is the appropriate formula for a child with cholestasis. It is medium chain triglyceride based which does not require bile acids for fat absorption and is the formula routinely utilized in children with cholestasis. Obviously, monitoring and supplementation of fat soluble vitamins (A,D,E,K) is also advised.