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Investigation of Paediatric Liver Disease

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Summary: The investigation of children with liver disease falls into two categories: the investigation of the cholestatic baby and the investigation of the older child (over 2 years) with hepatomegaly. The approach to investigation is directed by the clinical features and employs many different investigational methods including biochemistry, haematology, radiology, electrophysiology and histology. As the clinical presentation of many diseases is similar, it is appropriate to perform a variety of first-line tests, proceeding to more complex investigations only as indicated.

Investigation of paediatric liver disease can be divided into the investigation of the neonate or infant with cholestasis and the investigation of older children (over 2 years) with hepatomegaly.

NEONATAL LIVER DISEASE

The majority of children with liver disease will present in infancy with cholestasis. The main differential diagnosis is between extrahepatic biliary atresia, neonatal hepatitis or other disorders involving the biliary tree (Odievre, 1990).

A number of clinical clues will be evident from the clinical or family history and physical examination (Odievre, 1990) and the series of investigation will be directed by this information.

The first step in investigating any jaundiced baby is to establish whether there is significant conjugated hyperbilirubinaemia (> 15% of total bilirubin) (Mowat, 1987). Standard liver function tests are unlikely to be helpful in the differential diagnosis between biliary obstruction and 'neonatal hepatitis' (Table 1) but some information about hepatic synthesis and the chronicity of liver disease may be implied by low albumin concentration and prolonged coagulation times that are unresponsive to vitamin K therapy. Poor hepatic function at birth suggests that the disease process has existed *in utero* and may be either an inborn error of metabolism or an infection.

As most neonatal liver disease presents in a similar way, it is usual to perform a series of first-line tests in order to exclude the known causes of intrauterine infection and certain inborn errors of metabolism (Table 2 and Figures 1 and 2) and to establish the patency of the extrahepatic biliary tree. More specialized investigations for specific metabolic disorders are only performed if clinical suspicion is high or the first-line tests suggest an inborn error of metabolism (Table 2).

Table 1 Liver function tests in the diagnosis of neonatal liver disease

	Extrahepatic biliary disorder	*'Neonatal' hepatitis*
Bilirubin, < 20 μmol/L	↑ Conjugated	↑ Conjugated
Aminotransferases		
aspartate	↑	↑↑
alanine < 50 u/L		
Alkaline phosphatase, < 600 u/L	↑↑	↑
Albumin, > 30 g/L	N	Low/N
Cholesterol, 1–4 mmol/L	N	N/High
Bicarbonate, 21–25 mmol/L	N	N/Low
Prothrombin (PT), 12 s	N	N/Abn
Partial thromboplastin (PTT) 35 s	N	N/Abn

Table 2 First line investigations for paediatric liver disease

Bacterial culture of blood and urine
TORCH
Hepatitis A, B, C + HIV
Chromosomes
Sweat test
Plasma glucose and lactate
Free T_4 and TSH
Serum iron and ferritin
α_1-Antitrypsin level and phenotype
Galactose-1-phosphate UDT
Plasma amino acids
Urine:
Reducing sugars
Amino acids
Organic acids

It is essential to exclude associated infection by performing bacterial culture of blood and urine. Serological tests are performed to identify: toxoplasma, rubella, herpes simplex and parvo virus B19 (PHLS Working Party, 1990). IgM antibodies to cytomegalovirus (CMV) imply active infection (although not necessarily hepatitis), particularly if associated with CMV early antigen in urine, or positive throat and urine CMV cultures (Best, 1987). Hepatitis A, B or C are rare causes of neonatal hepatitis.

Chromosome studies should be performed because of the association between neonatal hepatitis and trisomy 13 and 18. A sweat test is carried out to exclude cystic fibrosis, which occasionally presents with neonatal hepatitis. Hypothyroidism is usually associated with unconjugated hyperbilirubinaemia, but it may exacerbate underlying hepatitis. All babies should have been screened for congenital hypothyroidism as part of a neonatal screening programme; however, this cannot be assumed. If there is any doubt, free T4 and TSH should be measured.

It is important to exclude extrahepatic biliary atresia or a choledocal cyst at an

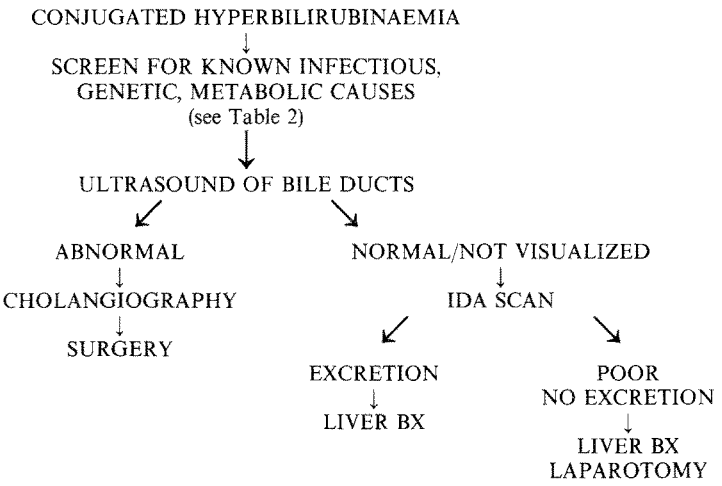

Figure 1 Evaluation of liver disease in childhood

early stage as corrective or palliative surgery is possible if performed early enough (Ohi *et al.*, 1985). The most useful investigation is an abdominal ultrasound, which may demonstrate a choledocal cyst or an absent gallbladder. If the ultrasound is normal or equivocal it is necessary to perform a radioisotope scan using technetium-IDA (iminodiacetic acid) to demonstrate hepatic uptake of isotope and excretion into the bowel. There is usually poor uptake but good excretion of isotope in neonatal hepatitis syndromes, whereas good uptake but no excretion into bowel suggests either extrahepatic biliary atresia or severe intrahepatic cholestasis.

Liver histology may differentiate between intra- and extrahepatic disorders, but there is considerable overlap in the pathological features. Classically, neonatal hepatitis of any aetiology will show giant cell transformation and rosette formation of hepatocytes with an inflammatory cell infiltrate. Excessive fat deposition is suggestive (but not diagnostic) of a metabolic disease. In extrahepatic biliary obstruction of any kind there will be fibrous expansion of the portal tracts with proliferation of bile ducts, but features of each disorder may be present and it may be necessary to perform a laparotomy and an operative cholangiogram to outline the extrahepatic biliary tree before the diagnosis is secure.

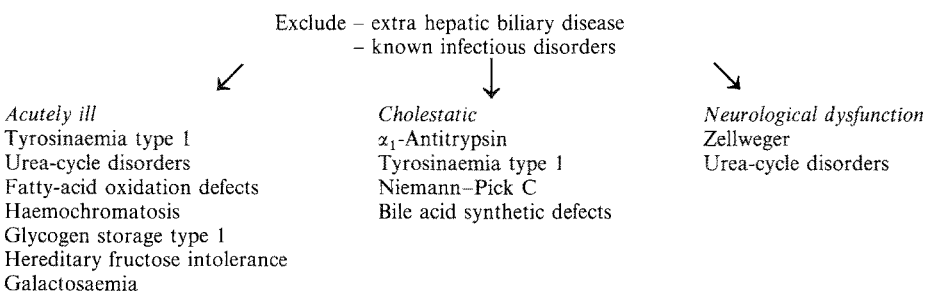

Figure 2 Differential diagnosis of metabolic liver disease in infants

INBORN ERRORS OF METABOLISM

The commonest inborn error of metabolism to present with neonatal cholestasis is α_1-antitrypsin deficiency (Table 2 and Figure 2), which is easily diagnosed by establishing a reduced serum level (< 0.8 g/L) and the phenotype PIZZ. Liver histology will confirm storage of PAS-positive granules in the endoplasmic reticulum (Mowat, 1990).

Inborn errors of carbohydrate metabolism are suggested by the detection of reducing sugars in urine or a low fasting glucose (< 3.0 mmol/L) and/or a raised plasma lactate (> 3.0 mmol/L) (Table 2). Babies with severe liver disease may have gross galactosuria secondary to severe liver disease and so it is necessary to exclude galactosaemia by estimating the enzyme galactose-1-phosphate uridyl transferase in red cells (Gitzelmann and Hansen, 1980). Hereditary fructose intolerance presents later in infancy when sucrose has been introduced to the diet and is confirmed by measuring fructose-1,6-diphosphate aldolase in liver or intestinal biopsies (Steinmann and Gitzelmann, 1981).

Glycogen storage disease does not present with neonatal cholestasis but with hepatomegaly and in some variants hypoglycaemia and a raised plasma lactate. It can be confirmed by measuring the appropriate enzymes in liver, leukocytes or muscle (Fernandes, 1990).

In patients with liver disease, tyrosinaemia type I is the commonest disorder of amino-acid metabolism and is suggested by the finding of elevated tyrosine and methionine in plasma and urine. A non-specific increase of tyrosine and methionine can occur in severe liver disease from any cause and therefore measurement of urinary succinyl-acetone is essential as this is elevated in tyrosinaemia type I. The diagnosis is then confirmed by demonstrating a deficiency of fumaryl-acetoacetate in leukocytes or cultured skin fibroblasts (Kvittingen, 1990). An elevated alpha-fetoprotein (> 800 IU/L) is additional suggestive evidence of tyrosinaemia and may indicate early malignant change in the liver, which can be confirmed by abdominal ultrasound, computed tomography or liver histology.

Niemann–Pick C is the commonest lipid storage disease to present with neonatal hepatitis. There is no screening test but the diagnosis must be excluded by looking for the characteristic storage cells in liver, in bone marrow and in ganglion cells on rectal biopsy (Lake, 1990).

Neurological problems in babies with neonatal liver disease may be primary (e.g. Zellweger syndrome) or secondary to unrecognized hypoglycaemia, hyperammonaemia or intracranial haemorrhage in α_1-antitrypsin deficiency (Hope *et al.*, 1982). Many of these babies will not be cholestatic and if they are acutely ill one needs to exclude urea-cycle defects and fatty-acid oxidation defects (Figure 2) by measuring plasma ammonia, amino acids, urinary amino acids, organic and orotic acid.

A peroxisomal disorder (such as Zellweger) is investigated by initially estimating very-long-chain fatty acids (VLCFA) in plasma (Heymans *et al.*, 1990). Serum iron is usually elevated. The absence, or presence, of abnormal peroxisomes can be demonstrated in liver tissue using special stains (Lake, 1990).

Patients with persisting cholestasis in whom all other investigations for inborn

errors of metabolism are negative should have urine screened for inborn errors of bile acid synthesis using FAB-MS (Clayton, 1990).

LIVER DISEASE IN CHILDREN OVER 2 YEARS

The commonest form of liver disease in this age group is acute or chronic hepatitis secondary to viral infection (Table 3). It is important to exclude the other diseases mentioned in Table 3 as they have a different clinical course and therapy.

Standard biochemical tests may show a hepatitis (raised transaminases 4–10 times upper limit of normal) or evidence of poor hepatocellular function (low albumin and abnormal coagulation). Abdominal ultrasound may show a small liver and an enlarged spleen, suggesting cirrhosis and portal hypertension from any cause. Evidence of chronic liver disease may be gained from X-rays of the wrist, which may show osteopenia or rickets. Severe rickets suggests a renal tubular disorder that is usually secondary to an inborn error of metabolism, e.g. Wilson disease, hereditary fructose intolerance, tyrosinaemia type I. As the most common diagnosis in this age group is acute or chronic viral hepatitis, a viral aetiology must be excluded at all times.

Autoimmune liver disease presents in either sex, although the incidence is higher in girls (3:1). Non-organ-specific autoantibodies may be demonstrated in 70% of children and there is always an increase in IgG (> 20 g/L).

As Wilson disease may present with almost any form of liver disease it must always be considered in children over 3 years old. Classically the diagnosis is established by detecting a reduced serum copper (< 10 μmol/L), ceruloplasmin (< 200 mg/L) and excess urine copper (> 1.0 μmol/24 h). Approximately 24% of children presenting with hepatic disease may have normal or borderline ceruloplasmin but all should have elevated urinary copper excretion, particularly after treatment with penicillamine (20 mg/kg) (Werlin *et al.*, 1978). There will be increased hepatic copper on liver histology (> 250 mg/g dry weight) which is higher than the amount detected in chronic cholestasis.

In equivocal cases, radioactive copper studies using either ^{64}Cu or ^{67}Cu will demonstrate reduced incorporation into ceruloplasmin compared to normals.

Table 3 Causes of chronic liver disease in children

Chronic persistent/active hepatitis
Post-viral hepatitis B, C, undefined
Autoimmune hepatitis
Drugs (nitrofurantrin, α-methyldopa)
Wilson disease (> 3 years)
α_1-Antitrypsin deficiency
Cystic fibrosis secondary to
Neonatal liver disease
Bile duct lesions

Table 4 Causes of acute liver failure in children

Infection
Viral hepatitis A, B, C, undefined
Poisons/drugs
Paracetamol, isoniazide
Halothane
Amanita phalloides
Metabolic
Wilson disease
Tyrosinaemia type 1
Fatty-acid oxidation defects
Autoimmune hepatitis
Reye syndrome

ACUTE LIVER FAILURE

Acute liver failure may present at any age. The clinical features are varied. Jaundice is not universally present, but there is always hypoglycaemia, encephalopathy and abnormal coagulation. The commonest cause is fulminant viral hepatitis (usually an unidentified virus) but a search for other aetiologies is mandatory (Table 4). It is important to perform a toxological screen for paracetamol as a history of drug ingestion is not always available.

Biochemical evidence of acute hepatitis with raised transaminases (10–100 times upper limit of normal) is always present in the early stages. Falling transaminases with a rising bilirubin and increasing coagulation times implies a poor prognosis.

An elevated ammonia is non-specific, but may suggest an inborn error of metabolism that should be excluded as described earlier. Encephalopathy can be monitored using an electroencephalogram (EEG), which correlates with the clinical state. The appearance of triphasic waves is characteristic of hepatic encephalopathy and the development of diffuse slow activity suggests a poor prognosis. Computed tomography of the brain may demonstrate cerebral oedema in acute fulminant viral hepatitis or Reye syndrome, which is important for management but not diagnosis. Liver biopsy to confirm the diagnosis is usually impossible because of coagulation abnormalities.

A Reye-like syndrome with acute liver failure and convulsions may be due to an underlying metabolic disorder, in particular fatty-acid oxidation defects. These disorders should be excluded by measuring plasma ammonia and amino acids, urinary amino acids and organic acids in the urine. A liver biopsy demonstrating microvesicular fatty deposition in the hepatocytes (Ballistreri, 1990) occurs both in Reye syndrome and fatty-acid oxidation defects and is not diagnostic.

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