

Review article: the evolving role of liver biopsy

M. S. CAMPBELL* & K. R. REDDY†

*Fellow in Gastroenterology and Hepatology, Hospital of the University of Pennsylvania, Division of Gastroenterology, Philadelphia, PA, USA; †Professor of Medicine and Surgery, Director of Hepatology, Medical Director of Liver Transplantation, Hospital of the University of Pennsylvania, Division of Gastroenterology, Philadelphia, PA, USA

Accepted for publication 13 May 2004

SUMMARY

Liver biopsy is traditionally the 'gold standard' for the evaluation of liver diseases. There are several situations in which its role is being challenged. In hepatitis C, liver biopsy helps assess prognosis and treatment candidacy. An important exception is genotype 2 or 3 because treatment is more likely to succeed and therapy is relatively short in duration. For hepatitis B, liver biopsy gives some prognostic information, but serologic tests and hepatic biochemical tests are the primary determinants of treatment candidacy. Non-

alcoholic fatty liver disease can be accurately diagnosed without a liver biopsy and, furthermore, there are no specific therapies available. The role of liver biopsy to assess methotrexate-associated hepatotoxicity remains controversial. Finally, patients with focal liver lesions usually do not require biopsy and, in the case of hepatocellular carcinoma, biopsy carries a risk of needle-track seeding. In short, the need for liver biopsy depends on the specific situation and should be performed when there is sufficient uncertainty about diagnosis, severity of disease, prognosis, and treatment decisions.

INTRODUCTION

In the course of treating patients with liver disease, questions often arise about the role of liver biopsy, traditionally the 'gold standard' for the evaluation of chronic liver diseases. Recently the need for liver biopsy has been challenged in a few clinical situations. With the advent of specific serologic diagnostic markers liver biopsy is not required for diagnosis of hepatitis B or C. Thus, is liver biopsy mandatory prior to starting treatment for hepatitis B or C? Non-alcoholic fatty liver disease (NAFLD), a condition that appears to be increasing in prevalence, can be diagnosed based on clinical and imaging features. Therefore, what role does liver biopsy play in the diagnosis, prognosis, and treatment of patients with NAFLD? Recommendations differ regarding periodic liver biopsy for patients taking

methotrexate. What is the current role of liver biopsy for following patients on methotrexate? Finally the role of liver biopsy in hepatocellular carcinoma has become a more contentious issue because advances in imaging technology have allowed highly specific identification of these lesions. Does every suspected hepatocellular carcinoma require biopsy to confirm diagnosis?

TECHNIQUES OF LIVER BIOPSY

Percutaneous liver biopsies are quick, safe procedures commonly performed in out-patient settings. Suction, cutting, and spring-loaded cutting needles with triggering mechanisms have all been safely used for this purpose. At our institution all three types of devices are used. The 14-gauge Trucut needle is preferred in patients with suspected cirrhosis because it may provide better samples without fragmentation. The benefit from using ultrasound to help determine biopsy site remains controversial. Potential liver biopsy sites marked by percussion were changed in between 3 and 15% of patients

Correspondence to: Professor K. R. Reddy, Hospital of the University of Pennsylvania, Division of Gastroenterology, 3 Ravdin, 3400 Spruce Street, Philadelphia, PA 19104, USA.
E-mail: rajender.reddy@uphs.upenn.edu

after ultrasound was performed.^{1, 2} In addition, a large, randomized, prospective trial found that ultrasonography use lowered the rate of post-biopsy hospitalization (most common reason for hospital admission was pain). Ultrasonography use did not lower rates of hypotension and bleeding.³ Certainly there is a long track record of safety for performing percutaneous liver biopsy without imaging guidance. Thus, the role of ultrasonography to guide percutaneous liver biopsy remains controversial. Use of ultrasound is not mandatory.

A transjugular approach to liver biopsy is often used in patients with a contraindication to percutaneous biopsy, when concomitant Transjugular Intrahepatic Portosystemic Shunt (TIPS) is planned, or according to provider preference. Although the biopsy specimen is often smaller and more fragmented than that acquired from a percutaneous approach, it is usually diagnostic.^{4, 5}

Laparoscopic liver biopsy allows visualization of the peritoneal cavity and liver surface. It is a safe procedure that can be done under conscious sedation, although it requires expertise that is not readily available. In one large study of 1794 patients, laparoscopic liver biopsies performed in a hepatology training programme provided definitive diagnoses of chronic liver disease in 98% of patients with a rate of only 0.45% major complications.⁶

Because a liver biopsy samples only about 1/50 000th of the entire liver, it may not be representative. Sampling error can approach 20–30%. For accurate diagnosis, it has been estimated that between 5 and 11 complete portal tracts are required with a biopsy length of at least 15–25 mm. A recent study used digitized images of whole sections of liver to allow for 'virtual biopsies' of variable lengths between 5 and 200 mm. For reliable staging of fibrosis in hepatitis C patients, a 25 mm biopsy length was adequate.⁷

HEPATITIS C

Chronic hepatitis C infection affects almost 3 million people in the United States. Chronic infection is generally asymptomatic and most often not progressive. Treatment for hepatitis C is long in duration, expensive, subject to side-effects, and associated with less than ideal response rates, particularly in patients with high viral load and genotype 1. Therefore, candidates for treatment need to be carefully selected. Liver biopsy helps prognosticate which patients with hepatitis C will

have progressive liver disease and are therefore more urgent candidates for treatment. Benefits from biopsy must be weighed against its risks, costs, and patient inconvenience. Non-invasive serologic tests and panels of tests are being developed with variable success to correlate with fibrosis and serve as possible substitutes for liver biopsy.

The decision to treat hepatitis C depends on the risk for progressive liver disease in the absence of treatment. Among many natural history studies, rates of progression to cirrhosis range from 1 to 37% after variable follow-up intervals.⁸ Tertiary referral centres, which are subject to selection bias, have reported the higher rates. Community-based cohorts likely give the most accurate prediction of progression to cirrhosis with a mean cumulative incidence of cirrhosis of 7%.⁸

Key histologic predictors of progression to cirrhosis are the degrees of inflammation, fibrosis, and steatosis seen on liver biopsy. Based on retrospective data, most patients with moderate inflammation on initial liver biopsy develop cirrhosis within 20 years, and nearly all patients with severe inflammation or bridging fibrosis develop cirrhosis within 10 years. Patients with mild inflammation and/or minimal fibrosis have a low risk of progression to cirrhosis.⁹ Hepatic steatosis is also emerging as a major risk factor for fibrosis progression in hepatitis C.¹⁰

Clinical information may help refine prognosis, but cannot substitute for the valuable information obtained from a liver biopsy. Poynard's group showed three clinical factors are independently associated with faster progression of fibrosis: age >40 at time of infection, daily alcohol consumption of 50 g or more, and male gender.¹¹ Indeed post-transfusion hepatitis C patients with mean age over 40 at time of infection have a cumulative incidence of cirrhosis of over 20% after 15–20 years of follow-up.^{12, 13} In contrast, studies of young women infected with hepatitis C have shown development of cirrhosis in <5% after 15–20 years.^{14, 15} Other factors predicting progression to cirrhosis include immunosuppression and coinfection with hepatitis B or HIV.^{16, 17}

Investigators have tried to identify serologic surrogates for cirrhosis and advanced fibrosis in hopes of obviating the need for liver biopsy. Serum hyaluronic acid has a 99% negative predictive value to predict the absence of cirrhosis, but only a 30% positive predictive value for cirrhosis.¹⁸ Other serum markers, including N-terminal propeptide of type III collagen, and YKL-40, have had

variable success for predicting fibrosis and cirrhosis.¹⁹ For the purpose of considering hepatitis C therapy, the distinction between minimal and significant fibrosis is more clinically useful than the distinction between the presence and absence of cirrhosis. The MULTIVIRC group studied 339 patients using a panel of five non-routine serum markers (α -2 macroglobulin, haptoglobin, γ -glutamyl transpeptidase, bilirubin and apolipoprotein A1) along with age and gender to predict significant fibrosis. Fifty per cent of the patients could be accurately stratified as having either significant or insignificant fibrosis; the other 50% had indeterminate results.²⁰

Serum alanine aminotransferase (ALT) has been used to predict liver fibrosis and inflammation. At any point in time, the serum ALT shows poor correlation with liver histology.²¹ Twenty-five to forty per cent of chronic hepatitis C patients have persistently normal ALT levels (usually defined as at least three normal ALTs within 6 months).^{22, 23} Patients with persistently normal ALTs tend to have less fibrosis and inflammation than hepatitis C patients with elevated ALTs.²⁴ Furthermore, in one study of 102 patients, the median progression of fibrosis was twice as fast in an elevated ALT group compared with a persistently normal ALT group (increase in Metavir fibrosis stage per year of 0.13 vs. 0.05, $P < 0.001$).²⁵ Despite these reassuring findings, patients with persistently normal ALTs may have significant inflammation (19% with moderate inflammation)²⁶ and even cirrhosis (6%)²⁷ (Tables 1 and 2).

Table 1. Prognostic indicators of hepatitis C – histologic

| Feature | Significance |
|-----------------------|------------------------------|
| Bridging fibrosis | >90% cirrhosis in 10 years |
| Severe inflammation | >90% cirrhosis in 10 years |
| Moderate inflammation | >90% cirrhosis in 20 years |
| Steatosis | Hastens fibrosis progression |

Table 2. Prognostic indicators of hepatitis C – clinical and laboratory

| Feature | Significance |
|---|---|
| Persistently normal alanine aminotransferase (ALT) | Fibrosis rate 1/2 of group with abnormal ALTs |
| MULTIVIRC index | Not routinely available. Accurately predicts fibrosis in 50% (in other 50% result is indeterminate) |
| Hyaluronic acid, N-terminal propeptide of type III collagen, YKL-40 | Not routinely available. Variably predictive of fibrosis |
| Age >40 at time of infection, male gender, alcohol use | Predictors of liver disease progression |
| Coinfection with hepatitis B, HIV, immunosuppression | Faster progression to cirrhosis |

Other benefits from liver biopsy have been proposed. Liver biopsy may reveal unsuspected cirrhosis. Such a diagnosis is important, as surveillance for hepatocellular carcinoma as well as oesophageal varices may then be initiated. Although liver biopsy could potentially identify diseases in addition to hepatitis C, a retrospective study of 126 patients with chronic hepatitis C found that unsuspected other disorders were found in only three patients (2%) who underwent liver biopsy.²⁸ Finally, liver biopsy may help predict the likelihood of response to therapy. In a study of pegylated interferon (α -2b with ribavirin), the absence of bridging fibrosis or cirrhosis was significantly associated with a sustained virological response (57% vs. 44%, $P = 0.001$).²⁹ However, a study of peginterferon α -2a with ribavirin showed that cirrhosis was not an independent risk factor for predicting sustained virologic response.³⁰

Although the information obtained from liver biopsy is useful, liver biopsy is not mandatory before initiating treatment for hepatitis C (Table 3). Many patients with genotype 2 or 3 are potentially candidates for therapy regardless of findings on liver biopsy because therapy is likely to succeed (80%), and duration of therapy is short (24 weeks). Liver biopsy intuitively is more useful for genotype 1 patients. Because therapy is longer (48 weeks) and less successful (<50%), many patients may desire therapy only if the chance of progressive liver disease from hepatitis C is high. Furthermore, liver biopsy may help some patients decide if they wish to continue therapy in the face of significant side-effects. For example, a patient with extensive fibrosis may decide to weather severe side-effects, possibly stopping work or making other life changes, because the risk of progressive liver disease in the absence of treatment is high. In addition, patients who have undergone unsuccessful therapy with an earlier form of therapy (for example, interferon monotherapy) may also benefit

Table 3. Utility of biopsy in hepatitis C

| Issue | Pros | Cons |
|--------------------------------|--|--|
| Diagnosis | | Hepatitis C antibody and HCV RNA reliable Liver biopsy rarely identifies unsuspected aetiology |
| Staging | Degree of fibrosis and inflammation are the best predictors of disease progression (refer to Table 1) | Clinical and laboratory surrogates for biopsy variably useful (refer to Table 2) |
| Identify cirrhosis | Finding clinically unsuspected cirrhosis prompts screening for varices and hepatocellular carcinoma | Cirrhosis usually clinically suspected |
| Predict response to therapy | | Biopsy not a proven predictor |
| Decision to treat | Genotype 1: Therapy longer and less likely to succeed. Biopsy identifies patients most in need of therapy | Genotypes 2 and 3: Therapy shorter and more likely to succeed. Biopsy not necessary for motivated patients |
| Patient preferences | | Some patients decide for or against therapy regardless of biopsy |
| Treatment-related side-effects | Severity of liver disease informs decision of whether to endure or stop therapy | |
| Previously treated | Lower success with retreatment. Identify patients most in need of therapy | |
| Post-treatment biopsy | Evolving role for biopsy after unsuccessful treatment to initiate and follow maintenance therapy | Maintenance therapy still under investigation |

from a liver biopsy. Expected response to peginterferon and ribavirin in such a patient is lower than in a treatment-naïve patient. Therefore, only patients with significant disease are potential candidates for retreatment. Finally, for patients who defer therapy, many physicians would recommend a repeat liver biopsy in 3–5 years to re-evaluate the need for therapy, based on disease progression.³¹

Although histology may improve after successful hepatitis C therapy, post-treatment liver biopsy is generally not useful for guiding patient management. Patients who do not achieve a sustained virologic response after treatment may show improved histology. One study of virologic non-responders who had achieved a histologic response after 6 months of therapy randomized patients to 2 years of maintenance interferon therapy or placebo. Maintenance therapy led to reduction of fibrosis.³² Long-term mortality and clinical benefits from maintenance therapy are not yet known. It is still unclear what role liver biopsy should play in both identifying candidates for maintenance therapy and also following its effectiveness.

HEPATITIS B

Although liver biopsy is frequently performed for hepatitis B patients, histologic findings often do not influence patient management. Liver biopsy can distin-

guish which patients are cirrhotic and may provide some information about expected disease progression. In some cases liver biopsy helps predict the response to therapy. On the other hand, the practical issue of whether to treat a patient for hepatitis B does not generally depend on histologic findings. Only patients who are likely to respond to therapy and who have actively replicating virus are treatment candidates. These patients can be identified based on hepatic biochemical tests, hepatitis B viral DNA levels, and hepatitis B serologic studies.

Performing liver biopsy to identify cirrhosis is of limited utility in hepatitis B. Identification of cirrhosis does prompt endoscopy to screen for oesophageal varices. Although cirrhosis increases the risk for hepatocellular cancer, periodic hepatocellular cancer screening should probably be performed in all hepatitis B surface antigen (HBsAg)-positive patients (particularly in patients from an endemic area), even in the absence of cirrhosis. Other features which lower the threshold for hepatocellular cancer screening include male gender, age of acquisition of hepatitis B, and family history of cancer.³³ Evidence of decompensated cirrhosis would prompt administration of lamivudine rather than interferon; however, biopsy is not needed to identify clinical decompensation.

Often serologic and clinical information are of more prognostic value than liver histology in predicting

progression to cirrhosis. Two natural history studies looked at progression to cirrhosis among HBsAg-positive non-cirrhotic patients. In one study independent risk factors for progression to cirrhosis were severe inflammation on liver biopsy, older age, and persistence of hepatitis B viral DNA in serum.³⁴ In the other study, independent risk factors included the occurrence of acute serologic exacerbation, hepatitis B virus DNA reappearance, and hepatic decompensation. Histologic information did not independently predict progression to cirrhosis.³⁵ A prospective study of 302 HBsAg-positive patients from an endemic region in southern Italy with median 94 month follow-up showed that survival is predicted by three independent factors: young age, the absence of cirrhosis, and a persistently normal ALT.³⁶

Ideal 'wild-type' chronic hepatitis B patients who are candidates for hepatitis B treatment are positive for HBsAg and hepatitis B e antigen (HBeAg), and have hepatitis B virus DNA of $\geq 10^5$ copies/mL. To expect successful therapy, serum transaminases are ideally elevated at least two times above the upper limit of normal. Precore mutant patients are unable to make HBeAg, but are HBsAg-positive. Such patients are candidates for therapy if they have abnormal transaminases and have serum HBV DNA of $\geq 10^4$ copies/mL. Patients from endemic areas with perinatally acquired infection have an initial immune-tolerant phase of infection. HBsAg, e antigen, and high levels of HBV DNA are present; however, serum ALT levels are normal. Liver biopsies performed on these patients show no or mild liver disease.³⁷ AASLD guidelines do not recommend liver biopsy or consideration for treatment of immune-tolerant phase patients until they show consistently elevated ALTs.³³ Liver biopsy and treatment are also not recommended for patients who have cleared hepatitis B infection or who are inactive carriers (surface antigen-positive, undetectable HBV DNA, and e antigen-negative). A German group concludes that, given the wealth of information available from non-invasive laboratory tests, pre-treatment liver biopsy has limited value.³⁸

A liver biopsy has limited value in predicting response (loss of serum HBeAg and viral DNA) to interferon therapy. The majority of studies has shown that low pre-treatment serum hepatitis B viral DNA and elevated ALT predict response to interferon therapy.³⁹⁻⁴² Histologic features are not predictive. For example, a randomized-controlled trial of interferon α -2b performed

in the United States showed that only low serum hepatitis B viral DNA and shorter duration of infection were predictive of response. The rate of cirrhosis was 65% in the study, and response to therapy was independent of liver histology.³⁹

There is, however, contradictory evidence about the role of liver histology in predicting response to lamivudine therapy. The Asian lamivudine trial was a randomized-controlled trial of 345 patients treated with lamivudine. There was only a 16% response to treatment (loss of HBeAg). A re-analysis of the data showed that the only independent predictor of response was an elevated ALT. Neither the degree of inflammation on liver biopsy nor the absence of cirrhosis predicted response.⁴³ On the other hand, an analysis of four controlled lamivudine phase III trials in the United States involving 805 patients showed that response correlated with both an elevated ALT and increased histologic inflammation. Of interest are observations of the small group with minimally elevated ALT (< 1.5 times upper limit of normal) treated with lamivudine. As expected, patients with a minimally elevated ALT had responses to lamivudine of only 2-3%. However, the small subset of patients with minimal elevation of ALT and severe inflammation showed an increased response of 21% to lamivudine.⁴⁴ This observation requires confirmation in prospective studies. Thus, it may be reasonable to consider liver biopsy in patients with fluctuating or minimal elevation of transaminases (< 2 times upper limit of normal). If moderate or severe inflammation is found on liver biopsy, then treatment could be initiated.

For patients who have not achieved serologic goals of therapy (HBeAg seroconversion to HB e antibody with loss of serum HBV DNA), continued therapy is suggested until these end-points are achieved. Such therapy may prevent histologic progression to cirrhosis and hepatocellular carcinoma. Even patients who did not respond serologically to lamivudine in fact showed histologic improvement, including reversal of cirrhosis, during long-term lamivudine therapy.⁴⁵ Histologic improvement is blunted by the emergence of YMDD variants, and perhaps adefovir will play a role in continued histologic improvement among these patients. However, studies with the goal of histologic improvement are awaited to better define the role of long-term therapy in chronic hepatitis B. At present, when either lamivudine or adefovir is used, it is recommended that duration of therapy be dictated by

achievement of serologic end-points (HBeAg seroconversion and persistent loss of quantifiable HBV DNA) and not by changes in histology. The role of liver biopsy (Table 4) in following response to therapy is not yet well-defined.

ABNORMAL HEPATIC BIOCHEMICAL TESTS AND NON-ALCOHOLIC FATTY LIVER DISEASE

Gastroenterologists commonly evaluate patients with chronically (6 months or more) elevated hepatic biochemical tests. Identifying the aetiology involves a careful history and physical examination, a medication and alcohol history, serologic studies, and imaging of the liver. In most cases an aetiology can be identified without a biopsy. In the majority of remaining cases the diagnosis is NAFLD. For example, in one study of 1124

patients presenting for evaluation of chronically elevated hepatic biochemical tests, only 81 patients (8%) had an undetermined aetiology before liver biopsy. On liver biopsy, 73 patients had NAFLD, and the remaining eight biopsies were normal.⁴⁶

Investigators in the pre-hepatitis C era often recommended liver biopsy for patients with chronically abnormal hepatic biochemical tests (Table 5) because prebiopsy diagnoses were inaccurate. Before the availability of serologic tests for hepatitis C, Van Ness and Diehl showed that a pre-biopsy diagnosis of non-alcoholic steatohepatitis (NASH) had only a 56% positive predictive value.⁴⁷ More recent studies have shown that pre-biopsy diagnoses of patients with chronically abnormal hepatic biochemical tests may be accurate enough to obviate the need for liver biopsy. Sorbi's group reported on 36 patients with persistently

Table 4. Utility of biopsy in hepatitis B

| Issue | Pros | Cons |
|--------------------|---|---|
| Identify cirrhosis | Prompts screening for varices and hepatocellular carcinoma | Hepatocellular carcinoma screening recommended for all hepatitis B surface antigen-positive (BsAg+) patients, cirrhotic or not |
| Prognosis | | Degree of fibrosis and inflammation not proven to predict progression |
| Decision to treat | If minimally elevated or fluctuating alanine aminotransferase (ALT), consider biopsy and treat if moderate or severe inflammation | Biopsy generally does not influence treatment decisions. Treat if elevated ALT, hepatitis BsAg+, hepatitis B DNA >10 ⁵ copies/mL, hepatitis BeAg+/- (precore mutants are eAg-, HBV DNA >10 ⁴ copies/mL) |
| Predict response | Degree of inflammation may predict response to lamivudine | Histologic findings do not help predict response to interferon |
| Long-term therapy | If serologic response not achieved, long-term therapy may improve histology | No established role for serial liver biopsies |

Table 5. Utility of biopsy for abnormal hepatic biochemical tests and non-alcoholic fatty liver disease (NAFLD)

| Issue | Pros | Cons |
|---|---|--|
| Elevated alanine aminotransferase (ALT) | Confirm diagnosis | Cause accurately identified in >90% cases without biopsy |
| Diagnose NAFLD | Patients may not have classic NAFLD risk factors | Accurate diagnosis of NAFLD possible without biopsy |
| Identify prognosis and severity of NAFLD | Imaging cannot distinguish simple steatosis from steatohepatitis Biopsy provides best prognostic information | In future, prognostic indices may be available |
| Treatment of NAFLD | Severe findings on biopsy may motivate risk factor modification | No proven therapy for NAFLD Risk factor modification can be recommended regardless of biopsy findings |
| Identify cirrhosis to begin variceal and hepatocellular carcinoma screening | Identify unsuspected cirrhosis | Cirrhosis usually clinically evident |

elevated hepatic biochemical tests in whom no definite diagnosis had been made after a thorough non-invasive investigation. A pre-biopsy diagnosis was then established; in 67% the pre-biopsy diagnosis was NAFLD. Biopsy subsequently changed diagnoses in only 14%, 80% of whom had normal liver biopsies. Treatment plans changed in only 12 patients, 10 of whom were offered investigational therapy. The authors conclude that after a careful non-invasive investigation, liver biopsy rarely aids either diagnosis or treatment decisions.⁴⁸

The definition of NAFLD requires minimal alcohol consumption, suggestive histology, and the absence of viral infection.⁴⁹ NAFLD is often suspected in patients with characteristic risk factors including hyperlipidaemia, diabetes mellitus and obesity. However, Bacon's group showed that a majority of their NAFLD patients did not have risk factors of obesity, diabetes mellitus, hyperlipidaemia and female gender.⁵⁰ Radiological imaging may also help to identify patients with NAFLD. A study of 25 patients with biopsy proven NAFLD (eight with steatosis alone, 17 with NASH) evaluated the use of ultrasound, computed tomography (CT) scan, and magnetic resonance imaging (MRI) for detecting fatty liver. All three imaging modalities were sensitive (93–100%) for detecting a significantly fatty liver; however, no imaging modality could distinguish simple steatosis from steatohepatitis or fibrosis.⁵¹

The major disadvantage to omitting a liver biopsy in a patient with suspected NAFLD is the loss of valuable information about disease severity and prognosis. Patients with NAFLD may have advanced fibrosis or cirrhosis on biopsy. Once cirrhosis is identified, endoscopy to screen for varices as well as periodic hepatocellular carcinoma screening may be performed. Cirrhosis is present in 10–20% of patients, and moderate-to-severe fibrosis in up to 40%.^{50, 52, 53} Histology also identifies patients at risk for increased progression to cirrhosis and higher mortality. Liver-associated death and progression to cirrhosis are increased in patients with evidence of steatonecrosis or fibrosis, but not in patients with less severe forms of NAFLD.⁵⁴ A study of 40 patients with mild NAFLD showed that after a median follow-up of 11 years only one patient progressed to mild fibrosis. Steatosis without inflammation or fibrosis appears to predict a benign prognosis.⁵⁵

Predictors of advanced histology have been investigated. Transaminases are not correlated with severity of

histology.⁵⁶ One study identified age over 45 years, obesity, and diabetes mellitus as independent risk factors for predicting bridging fibrosis or cirrhosis.⁵⁷ Another study developed an index from age, body mass index, hypertriglyceridaemia, and ALT two times over the upper limit of normal to predict more than minimal fibrosis. They were able to define an index cut-off with sensitivity of 100% and specificity of 47%.⁵³ Although predictive models appear somewhat promising, none has yet been validated.

Although there is no proven therapy for NAFLD, weight loss and careful management of comorbidities such as diabetes mellitus and hyperlipidaemia may be recommended. On a practical level, biopsy results are not generally needed to make these conservative recommendations. On the other hand, demonstration of advanced histologic changes may provide an impetus for some patients to lose weight, better control their diabetes mellitus, and cease all alcohol consumption. Furthermore, more severe histologic disease may prompt the use of investigational drugs, such as pioglitazone, rosiglitazone, metformin, and vitamin E in selected individuals.⁵⁸ At this time, follow-up biopsies to assess histologic response to conservative measures such as weight loss and diabetes mellitus control cannot be recommended outside of research studies.

METHOTREXATE-INDUCED HEPATOTOXICITY

Methotrexate is commonly used to treat psoriasis and rheumatoid arthritis. Controversy lies in the role of routine liver biopsy to detect the development of unsuspected significant fibrosis during treatment. Evidence-based recommendations differ between the dermatology and rheumatology literature.

Severe fibrosis and cirrhosis are relatively common (up to 17%) among psoriasis patients treated with long-term methotrexate, and often hepatic biochemical tests are normal. However, studies of methotrexate hepatotoxicity in psoriasis patients have not rigorously controlled for other underlying chronic liver diseases including alcoholic liver disease, viral hepatitis and NASH. Dermatology guidelines developed in the 1980s recommend liver biopsy before therapy, after an initial total dose of 1.5 g of methotrexate, and every 1–1.5 g cumulative dose thereafter. Finding moderate-to-severe fibrosis or cirrhosis would preclude further therapy.⁵⁹

Among rheumatoid arthritis patients taking long-term methotrexate, the prevalence of significant fibrosis and

cirrhosis is rare, probably owing to more rigorous exclusion of chronic liver disease. It is also possible that patients with rheumatoid arthritis, when compared with psoriasis, have a disease-specific reduced risk for methotrexate-induced toxicity. During the course of methotrexate therapy, persistent transaminase elevation has been shown to predict liver injury.⁶⁰ Rheumatology guidelines recommend pre-treatment liver biopsy if chronic liver disease is suspected. Liver biopsy during therapy is recommended if a majority of ASTs over a year's time (repeated every 4–8 weeks) is elevated or if serum albumin is decreased. Furthermore, reduction in methotrexate dose is recommended in response to an elevated AST. If moderate-to-severe fibrosis or cirrhosis is found, then treatment should be discontinued.⁶¹ Long-term liver safety by following the above guidelines has been demonstrated for rheumatoid arthritis patients.⁶²

FOCAL LIVER LESIONS

The differential diagnosis of focal liver lesions includes a broad spectrum of benign, malignant, and infectious aetiologies. Age, gender, use of oral contraceptives, presence of cirrhosis, travel history, fever, and presence of extrahepatic malignancy help narrow the differential diagnosis. Radiological studies are often diagnostic, and tumour markers may support a suspected diagnosis. Although radiological-guided biopsy is at times needed, biopsy may carry risks of bleeding and needle-track seeding.

Biopsy of focal liver lesions is often not necessary. Most focal liver lesions have characteristic findings on CT or MRI. For example, cavernous haemangiomas may have peripheral feeders with the same attenuation as the aorta. Focal nodular hyperplasia may demonstrate a focal enhancement of a central scar during arterial phase.⁶³ Confident characterization by imaging can obviate the need for biopsy. Furthermore, biopsy is often non-diagnostic for hepatic adenomas and focal nodular hyperplasia. Biopsy of hepatic adenomas, focal nodular hyperplasia, and haemangiomas also carries an increased bleeding risk.⁶⁴ One group of investigators studied 160 patients referred for the evaluation of focal liver lesions. Preoperative fine needle biopsy of the lesions was not performed. Patients subsequently underwent surgery, and in 98% of cases the preoperative diagnosis was confirmed.⁶⁵

In some cases imaging is not conclusive, and the patient may need to undergo surgical resection for

definitive diagnosis. For example, if imaging cannot distinguish between focal nodular hyperplasia and hepatic adenoma, surgical resection could be performed. Resection would prevent cancer and catastrophic bleeding, which could develop if the lesion were a hepatic adenoma. Fine needle biopsy may be avoided because of the likelihood of a non-diagnostic result and the risk of bleeding.

The occurrence of a focal liver lesion in a cirrhotic is suspicious for hepatocellular carcinoma. Among cirrhotics the sensitivity of fine needle biopsy has been shown to be between 86 and 90% for the detection of hepatocellular carcinoma. The accuracy of diagnosis is influenced by nodule location and size. Biopsy is more sensitive for lesions over 3 cm.^{66–68} Non-invasive methods of diagnosing hepatocellular carcinoma include tumour markers and radiological studies. Although α -fetoprotein is normal in up to 40% of patients with hepatocellular carcinoma, a significantly elevated value can confirm the diagnosis. For example, one study of cirrhotic patients, 170 of whom were diagnosed with hepatocellular carcinoma, showed that an α -fetoprotein of 200 ng/mL had a 99% specificity for hepatocellular carcinoma. Lower elevations in α -fetoprotein had lower specificity.⁶⁹ Other diagnostic markers including des- γ -carboxy prothrombin, are under study. Cross-sectional imaging has shown excellent diagnostic accuracy in defining the presence of hepatocellular carcinoma. One study showed that CT had 86–89% sensitivity and 99% specificity for identifying hepatocellular carcinoma.⁷⁰

In recent years investigators have determined that biopsy of hepatocellular carcinomas carries a significant risk of needle-track seeding (1.6–5%).^{66, 67, 71} The occurrence of needle-track seeding has provided impetus for defining conditions in which biopsy may be avoided. An α -fetoprotein over 200 ng/mL or features suggestive of hepatocellular carcinoma on CT or MRI is over 99% specific for the presence of hepatocellular carcinoma. A recent consensus guideline suggests that hepatocellular carcinoma may be diagnosed without biopsy if a characteristic mass >2 cm in a cirrhotic liver is detected by two imaging techniques (out of ultrasound, CT and MRI). Elevated α -fetoprotein (over 400 ng/mL) may also confirm the diagnosis.⁷² At our institution, fine-needle liver biopsy of suspected hepatocellular carcinoma is generally reserved for patients in whom no definitive surgical intervention is planned. Biopsy is obtained at the time of non-surgical treatment

(radiofrequency ablation, alcohol ablation, or chemo-embolization).

CONCLUSIONS

Liver biopsy is commonly performed to diagnose liver disease, prognosticate, and help to determine treatment. In hepatitis C, liver biopsy plays an important role in staging and grading the extent of disease and predicting disease progression. Often the severity of liver disease seen on biopsy is the key determinant to whether a patient will undergo a treatment which is long in duration, has a limited chance for success, and carries a high rate of side-effects. An important exception is the patient with genotype 2 or 3, who may be willing to accept treatment regardless of findings on liver biopsy because treatment is shorter in duration and more likely to succeed. For hepatitis B, liver biopsy does give some prognostic information, but hepatitis B serologies and hepatic biochemical tests are the primary determinants of treatment candidacy and response. NAFLD can be accurately diagnosed without a liver biopsy, although liver biopsy can provide important prognostic information. Because there is no proven treatment for NAFLD, liver biopsy does not generally influence treatment recommendations outside of research studies. In some cases advanced findings on biopsy may motivate some patients to consider investigational drugs and pursue risk factor modification. There is a difference of opinion between the rheumatology and dermatology literature regarding the use of liver biopsy to assess for methotrexate-induced hepatotoxicity. Until the controversy is resolved, it would seem reasonable to perform a liver biopsy in selected patients who have risk factors for chronic liver disease and persistent elevations of transaminases and are on chronic methotrexate therapy. Finally, patients with focal liver lesions usually do not require biopsy, and biopsy of hepatocellular carcinoma carries a risk of needle-track seeding. In short, the role of liver biopsy depends on the specific situation. It should be undertaken when a physician and patient feel that a biopsy would help to clarify situations where there is sufficient uncertainty about diagnosis, severity of disease, prognosis and treatment decisions.

ACKNOWLEDGEMENT

No financial support was required for this study.

REFERENCES

- 1 Smith CI, Grau JE. The effect of ultrasonography on the performance of routine liver biopsy. *Hepatology* 1995; 22: 384A.
- 2 Riley TR. How often does ultrasound marking change the liver biopsy site? *Am J Gastroenterol* 1996; 91: 1292–6.
- 3 Lindor KD, Jorgensen RA, Rakela J, *et al.* The role of ultrasonography and automatic-needle biopsy in outpatient percutaneous liver biopsy. *Hepatology* 1996; 23: 1079–83.
- 4 McAfee JH, Keefe EB, Lee RG, *et al.* Transjugular liver biopsy. *Hepatology* 1992; 15: 726–32.
- 5 Lebrec D, Goldfarb G, Degott C, *et al.* Transvenous liver biopsy: an experience based on 1,000 hepatic tissue samplings with this procedure. *Gastroenterology* 1992; 83: 338–40.
- 6 Vargas C, Jeffers LJ, Bernstein D, *et al.* Diagnostic laparoscopy: a 5-year experience in a hepatology training program. *Am J Gastroenterol* 1995; 90: 1258–62.
- 7 Bedossa P, Dargere D, Paradis V. Sampling variability of liver fibrosis in chronic hepatitis C. *Hepatology* 2003; 38: 1449–57.
- 8 Freeman AJ, Dore GJ, Low MG, *et al.* Estimating progression to cirrhosis in chronic hepatitis C virus infection. *Hepatology* 2001; 34: 809–16.
- 9 Yano M, Kumada H, Kage M, *et al.* The long-term pathological evolution of chronic hepatitis C. *Hepatology* 1996; 23: 1334–40.
- 10 Westin J, Norlinder H, Lagging M, *et al.* Steatosis accelerates fibrosis development over time in hepatitis C virus genotype 3 infected patients. *J Hepatol* 2002; 37: 837–42.
- 11 Poynard T, Bedossa P, Opolon P. Natural history of liver fibrosis progression in patients with chronic hepatitis C. The OBSVIRC, METAVIR, CLINIVIR, and DOSVIRC groups. *Lancet* 1997; 349: 825–32.
- 12 Seeff LB, Buskell-Bales Z, Wright EC, *et al.* Long-term mortality after transfusion associated non-A non-B hepatitis. The National Heart, Lung and Blood Institute Study Group. *N Engl J Med* 1992; 327: 1906–11.
- 13 Tong MJ, El-Farra NS, Reijes AR, *et al.* Clinical outcomes after transfusion-associated hepatitis C. *N Engl J Med* 1995; 332: 1463–6.
- 14 Kenny-Walsh E. Clinical outcomes after hepatitis C infection from contaminated anti-D immune globulin. *N Engl J Med* 1999; 340: 1228–33.
- 15 Wiese M, Berr F, Lafrenz M, *et al.* Low frequency of cirrhosis in a hepatitis C (genotype 1b) single-source outbreak in Germany: a 20-year multicenter study. *Hepatology* 2000; 32: 91–6.
- 16 Soto B, Sanchez-Quijano A, Rodrigo L, *et al.* HIV infection modifies the natural history of chronic parenterally acquired hepatitis C with an unusually rapid progression to cirrhosis. A multicenter study on 547 patients. *J Hepatol* 1997; 26: 1–5.
- 17 Tsai JF, Jeng JE, Ho MS, *et al.* Independent and additive effect modification of hepatitis B and C virus infection on the development of chronic hepatitis. *J Hepatol* 1996; 24: 271–6.
- 18 McHutchison JG, Blatt LM, de Medina M, *et al.* Measurement of serum hyaluronic acid in patients with chronic hepatitis C

- and its relationship to liver histology. Consensus Interferon Study Group. *J Gastroenterol Hepatol* 2000; 15: 945–51.
- 19 Fontana RJ, Lok AS. Noninvasive monitoring of patients with chronic hepatitis C. *Hepatology* 2002; 36: S57–64.
 - 20 Imbert-Bismut F, Ratziu V, Pieroni L, *et al.* Biochemical markers of liver fibrosis in patients with hepatitis C virus infection: a prospective study. *Lancet* 2001; 357: 1069–75.
 - 21 Haber MM, West AB, Haber AD, *et al.* Relationship of aminotransferases to liver histological status in chronic hepatitis C. *Am J Gastroenterol* 1995; 90: 1250–7.
 - 22 Marcellin P, Levy S, Erlinger S. Therapy of hepatitis C: patients with normal aminotransferase levels. *Hepatology* 1997; 26: 133S–6S.
 - 23 Inglesby TV, Rai R, Astemborski J. A prospective, community-based evaluation of liver enzymes in individuals with hepatitis C after drug use. *Hepatology* 1999; 29: 590–6.
 - 24 Martinot-Peignoux M, Boyer N, Cazals-Hatem M, *et al.* Prospective study on anti-hepatitis C virus-positive patients with persistently normal serum alanine transaminase with or without detectable serum hepatitis C virus RNA. *Hepatology* 2001; 34: 1000–5.
 - 25 Martin P, Moussalli J, Cadranel JF, *et al.* Slow progression rate of fibrosis in hepatitis C virus patients with persistently normal alanine transaminase activity. *Hepatology* 1998; 27: 868–72.
 - 26 Marcellin P, Levy S, Benhamou JP, *et al.* Management of the asymptomatic HCV carrier with normal ALT levels. *Viral Hepat Rev* 1996; 2: 277–84.
 - 27 Jamal AM, Soni A, Quinn PG, *et al.* Clinical features of hepatitis C-infected patients with persistently normal alanine transaminase levels in the Southwestern United States. *Hepatology* 1999; 30: 1307–11.
 - 28 Saadeh S, Cammell G, Carey WD, *et al.* The role of liver biopsy in chronic hepatitis C. *Hepatology* 2001; 33: 196–200.
 - 29 Manns MP, McHutchison JG, Gordon SC, *et al.* Peginterferon alfa-2b plus ribavirin compared with interferon alfa-2b plus ribavirin for initial treatment of chronic hepatitis C: a randomized trial. *Lancet* 2001; 358: 958–65.
 - 30 Fried MW, Shiffman ML, Reddy KR, *et al.* Peginterferon alfa-2a plus ribavirin for chronic hepatitis C virus infection. *N Engl J Med* 2002; 347: 975–82.
 - 31 Marcellin P, Asselah T, Boyer N. Fibrosis and disease progression in hepatitis C. *Hepatology* 2002; 36: S47–56.
 - 32 Shiffman ML, Hofmann CM, Contos MJ, *et al.* A randomized, controlled trial of maintenance interferon therapy for patients with chronic hepatitis C virus and persistent viremia. *Gastroenterology* 1999; 117: 1164–72.
 - 33 Lok AS, McMahon BJ. Chronic hepatitis B. *Hepatology* 2001; 34: 1225–41.
 - 34 Fattovich G, Brollo L, Giustina G, *et al.* Natural history and prognostic factors for chronic hepatitis type B. *Gut* 1991; 32: 294–8.
 - 35 Liaw YF, Tai DI, Chu CM, *et al.* The development of cirrhosis in patients with chronic type B hepatitis: a prospective study. *Hepatology* 1988; 8: 493–6.
 - 36 Di Marco V, Lo Iacono O, Camma C, *et al.* The long-term course of chronic hepatitis B. *Hepatology* 1999; 30: 257–64.
 - 37 Chang MH, Hwang LY, Hsu HC, *et al.* Prospective study of asymptomatic HBsAg carrier children infected in the perinatal period: clinical and liver histologic studies. *Hepatology* 1998; 8: 374–7.
 - 38 Heintges T, Mohr L, Hensel F, *et al.* Value of liver biopsy prior to interferon therapy for chronic viral hepatitis. *Dig Dis Sci* 1998; 43: 1562–5.
 - 39 Perrillo RP, Schiff ER, Davis GL, *et al.* A randomized, controlled trial of interferon alfa-2b alone and after prednisone withdrawal for the treatment of chronic hepatitis B. *N Engl J Med* 1990; 323: 295–301.
 - 40 Lok AS, Wu P-C, Lai C-L, *et al.* A controlled trial of interferon with or without prednisone priming for chronic hepatitis B. *Gastroenterology* 1992; 102: 2091–7.
 - 41 Brook MG, Karayiannis P, Thomas HC. Which patients with chronic hepatitis B virus infection will respond to alpha-interferon therapy? A statistical analysis of predictive factors. *Hepatology* 1989; 10: 761–3.
 - 42 Hoofnagle JH, Peters M, Mullen KD, *et al.* Randomized, controlled trial of recombinant human alpha-interferon in patients with chronic hepatitis B. *Gastroenterology* 1988; 95: 1318–25.
 - 43 Chien RN, Liaw YF, Atkins M. Pretherapy alanine transaminase level as a determinant for hepatitis Be antigen seroconversion during lamivudine therapy in patients with chronic hepatitis B. *Asian Hepatitis Lamivudine Trial Group. Hepatology* 1999; 39: 770–4.
 - 44 Perrillo RP, Lai C-L, Liaw Y-F, *et al.* Predictors of HBeAg loss after lamivudine treatment for chronic hepatitis B. *Hepatology* 2002; 36: 186–94.
 - 45 Dienstag JL, Goldin RD, Heathcote EJ, *et al.* Histological outcome during long-term lamivudine therapy. *Gastroenterology* 2003; 124: 105–17.
 - 46 Daniel S, Ben-Menachem T, Vasudevan G, *et al.* Prospective evaluation of unexplained chronic liver transaminase abnormalities in asymptomatic and symptomatic patients. *Am J Gastroenterol* 1999; 94: 3010–4.
 - 47 Van Ness MM, Diehl AM. Is liver biopsy useful in the evaluation of patients with chronically elevated liver enzymes? *Ann Intern Med* 1989; 111: 473–8.
 - 48 Sorbi D, McGill DB, Thistle JL, *et al.* An assessment of the role of liver biopsies in asymptomatic patients with chronic liver test abnormalities. *Am J Gastroenterol* 2000; 95: 3206–10.
 - 49 Powell EE, Cooksley WG, Hanson R, *et al.* The natural history of nonalcoholic steatohepatitis: a follow-up study of forty-two patients for up to 21 years. *Hepatology* 1990; 11: 74–80.
 - 50 Bacon BR, Faravash MJ, Janney CG, *et al.* Nonalcoholic steatohepatitis: an expanded clinical entity. *Gastroenterology* 1994; 107: 1103–9.
 - 51 Saadeh S, Younossi ZM, Remer EM, *et al.* The utility of radiological imaging in nonalcoholic fatty liver disease. *Gastroenterology* 2002; 123: 745–50.
 - 52 Lee RG. Nonalcoholic steatohepatitis: a study of 49 patients. *Hum Pathol* 1989; 20: 594–8.
 - 53 Ratziu V, Giral P, Charlotte F, *et al.* Liver fibrosis in overweight patients. *Gastroenterology* 2000; 118: 1117–23.

- 54 Matteoni CA, Younossi ZM, Gramlich T, *et al.* Nonalcoholic fatty liver disease: a spectrum of clinical and pathological severity. *Gastroenterology* 1999; 116: 1413–9.
- 55 Teli MR, James OF, Burt AD, *et al.* The natural history of nonalcoholic fatty liver: a follow-up study. *Hepatology* 1995; 22: 1714–9.
- 56 Sonsuz A, Basaranoglu M, Ozbay G. Relationship between aminotransferase levels and histopathological findings in patients with nonalcoholic steatohepatitis (letter). *Am J Gastroenterol* 2000; 95: 1370–1.
- 57 Angulo P, Keach JC, Batts KP, *et al.* Independent predictors of liver fibrosis in patients with nonalcoholic steatohepatitis. *Hepatology* 1999; 30: 1356–62.
- 58 Angulo P. Medical progress: nonalcoholic fatty liver disease. *N Engl J Med* 2002; 346: 1221–31.
- 59 Roenigk HH, Auerbach R, Maibach HI, *et al.* Methotrexate in psoriasis: revised guidelines. *J Am Acad Dermatol* 1988; 19: 145–56.
- 60 Kremer JM, Lee RG, Tolman KG. Liver histology in rheumatoid arthritis patients receiving long-term methotrexate therapy: a prospective study with baseline and sequential biopsy samples. *Arthritis Rheum* 1989; 32: 121–7.
- 61 Kremer JM, Alarcon GS, Lightfoot RW, *et al.* Methotrexate for rheumatoid arthritis. *Arthritis Rheum* 1994; 37: 316–28.
- 62 Kremer JM, Kaye GI, Ishak KG, *et al.* Light and electron microscopic analysis of sequential liver biopsy samples from rheumatoid arthritis patients receiving long-term methotrexate therapy. *Arthritis Rheum* 1995; 38: 1194–203.
- 63 Ros PR, Davis GL. The incidental focal liver lesion: photon, proton, or needle? *Hepatology* 1998; 27: 1183–90.
- 64 Reddy KR, Schiff ER. Approach to a liver mass. *Semin Liver Dis* 1993; 13: 423–35.
- 65 Torzilli G, Minagawa M, Takayama T, *et al.* Accurate preoperative evaluation of liver mass lesions without fine-needle biopsy. *Hepatology* 1999; 39: 889–93.
- 66 Huang GT, Sheu JC, Yang PM, *et al.* Ultrasound-guided cutting biopsy for the diagnosis of hepatocellular carcinoma – a study based on 420 patients. *J Hepatol* 1996; 25: 334–8.
- 67 Durand F, Regimbeau JM, Belghiti J, *et al.* Assessment of the benefits and risks of percutaneous biopsy before surgical resection of hepatocellular carcinoma. *J Hepatol* 2001; 35: 254–8.
- 68 Fornari F, Filice C, Rapaccini GL, *et al.* Small (< or >3 cm) hepatic lesions. Results of sonographically guided fine-needle biopsy in 385 patients. *Dig Dis Sci* 1994; 39: 2267–75.
- 69 Trevisani F, D’Intino PE, Morselli-Labate AM, *et al.* Serum alpha-fetoprotein for diagnosis of hepatocellular carcinoma in patients with chronic liver disease: influence of HbsAg and anti-HCV status. *J Hepatol* 2001; 34: 570–5.
- 70 Lim JH, Choi D, Kim SH, *et al.* Detection of hepatocellular carcinoma: value of adding delayed phase imaging to dual-phase helical CT. *AJR Am J Roentgenol* 2002; 179: 67–73.
- 71 Takamori R, Wong LL, Dang C, *et al.* Needle-tract implantation from hepatocellular cancer: is needle biopsy of the liver always necessary? *Liver Transplant* 2000; 6: 67–72.
- 72 Bruix J, Sherman M, Llovet JM, *et al.* Clinical management of hepatocellular carcinoma. Conclusions of the Barcelona-2000 EASL conference. *J Hepatol* 2001; 35: 421–30.