

Liver biopsy prognostication – the historical perspective
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Introduction

Needle puncture of the liver was performed by a number of pioneers of medical science including Erlich and Lucatello in the 1880s. Although this was largely used for therapeutic drainage of cystic lesions including abscesses there was at least some appreciation that obtaining tissue through this route might provide invaluable diagnostic information. Needle biopsy procedures for the investigation of diseases of solid organs took a further 50 years or more to become established and it was not until the end of the 1940s and early 1950s²⁻⁵ that diagnostic percutaneous liver biopsies began to be used in clinical practice. This was initially used primarily for the differential diagnosis of obstructive jaundice (or what Movitt referred to as regurgitation jaundice². It became increasingly applied however to the assessment of infective disorders, in particular tuberculosis and in the Middle East in the diagnosis of schistosomiasis⁶. Others appreciated the potential role of needle biopsy of the liver in the work up of patients with malignant diseases⁷. The introduction of the Mengini needle in the late 1950s was a further boost to this diagnostic modality as was the subsequent development of other biopsy devices such as the Tru-Cut biopsy.

Obtaining a primary diagnosis in a patient with abnormal liver function remains the principal indication for percutaneous liver biopsy but its role has evolved over time (Table 1), thus information obtained from histological examination of liver tissue can offer important prognostic features which can influence therapeutic options and map out the likely clinical course. This presentation will chart the “rise and fall” of the liver biopsy in prognostication but will argue that the oft-predicted “fall” is not only slow in coming but unlikely to be precipitous.

Table 1. Indications for liver biopsy: a 21st century perspective

- Establishing diagnosis in patients with abnormal liver function tests
 - Investigation of pyrexia of unknown origin
 - Diagnosis of systemic disorders
 - Investigation of cholestatic disorders
 - Diagnosis of chronic hepatitis
 - Assessment of grade and stage of chronic hepatitis – important for prognostication and therapy
 - Assessment of therapeutic effects in chronic hepatitis
 - Diagnosis of fatty liver disease
 - Assessment of grade and stage of fatty liver disease
 - Assessment of efficacy of therapy for fatty liver disease
 - Diagnosis of post-transplant complications
 - Diagnosis of intrahepatic tumours
 - Assessment of grade of intrahepatic malignant neoplasms
 - Assessment of non-tumourous liver in patients referred for resection
 - Pre-transplant assessment (living related donors, cadaveric donors, recipients)
 - Investigation of hepatomegaly
 - Diagnosis of inherited metabolic defect (iron overload, Wilson disease etc.)
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The rise of liver biopsy in prognostication

Before considering how liver biopsy interpretation became an increasing part of predicting outcome in patients with liver disease, it is important to stress that with liver disease a clear separation between diagnosis and prognosis is somewhat artificial. Establishing the underlying pattern of liver injury and establishing a firm diagnosis continues to play a crucial role in predicting likely outcome but it has been the increasing use of semi-quantitative analyses to assess grade and stage in both non-neoplastic and neoplastic disease which has increased its position within the prognostic armamentarium of the clinical hepatologist⁸⁻¹⁰.

Chronic viral hepatitis

It is thought that in excess of 350 million people worldwide are infected by hepatitis B while some 180 million individuals are chronic carriers for hepatitis C. Both of these viral hepatitises are associated with chronic disease including the development of cirrhosis and the complication of hepatocellular carcinoma. Chronic infection with both viruses is generally asymptomatic and progression is highly variable. Therapeutic intervention tends to require long courses of expensive anti-viral agents many of which are associated with significant side effects and with hepatitis C the response rates are still sub-optimal in spite of recent advances in drug development. As a consequence, particularly with hepatitis C, the decision of whether or not to treat has to be informed by some judgement of the likely rate of progression. Routine liver function tests correlate poorly with the true activity of the disease and cannot be reliably used as prognostic factors¹¹.

Although there are clinical algorithms (see below) that can help in predicting outcome, liver biopsy investigation has been central to the diagnostic work up of patients with HCV since the virus was first identified in 1989. Over the last 25 years a number of key publications have demonstrated that the grade and stage of disease on biopsy can predict the likely development of cirrhosis^{9, 12}. Recent studies have highlighted that the degree of steatosis is also a risk factor for progression of fibrosis in hepatitis C¹³.

In hepatitis B, biopsy findings appear to provide less prognostic information than with hepatitis C. In the early days following discovery of the "Australia antigen" biopsy interpretation was important in establishing a diagnosis. It became clear that the so-called ground glass hepatocytes were a feature on biopsy of at least a proportion of patients that were found to have circulating Australia antigen and this remained an important marker of the disease before serum tests and molecular virology were properly established¹⁴. As new anti-viral agents were being developed for the treatment of hepatitis B, biopsy interpretation was used in stratifying patients for therapeutic options but in the early 21st century it is rare for the treatment options in hepatitis B to be dependent on histological findings. The likelihood of response to therapy can generally be identified by assessing biochemical results, HBV DNA levels and HBV serological studies. The efficacy of biopsy findings in predicting the likelihood of progression to cirrhosis is controversial with contradictory findings being observed in two key prospective studies^{15, 16}. There is some evidence that changes on biopsy may be important in predicting response to novel anti-virals in hepatitis B. This was supported by a meta-analysis of clinical trials of lamivudine in the US¹⁷. Furthermore other studies have underlined the importance of monitoring the outcomes following therapy which again have important prognostic messages¹⁸.

Fatty liver disease

It has long been established that only a proportion of chronic alcoholics will develop progressive disease ultimately leading to micronodular cirrhosis. A sizeable number will not progress beyond a simple steatosis but others develop steatohepatitis, the key precursor lesion for the

development of cirrhosis. The long term prognosis depends on the severity of hepatic injury but the degree of liver function test abnormalities correlate poorly with the extent of hepatic injury particularly in those who continue to imbibe. A series of key papers by a Veterans Administration Co-operative Study Group in the 1980s and early 90s demonstrated that the worst prognosis in alcoholic liver disease was in those with alcoholic hepatitis superimposed on cirrhosis¹⁹. The group also identified other prognostic features most notably the degree of cholestasis.

Biopsy findings are of even greater importance in mapping the likely outcome in non-alcoholic fatty liver disease (NAFLD). Again there is a poor correlation between liver function test abnormalities and degree of injury. A number of retrospective and prospective studies have demonstrated that simple steatosis without inflammation carries a benign prognosis while those with marked a necro-inflammatory component to their NAFLD are likely to progress to advanced fibrosis and cirrhosis^{21, 22}. From this point of view, there has been a great deal of effort made in most centres to distinguish between those who have simple steatosis and those who have steatohepatitis but more recent evidence will be presented which suggests that those with some features of steatohepatitis, including ballooning degeneration, but without established fibrosis or marked necro-inflammatory activity may also run a benign course. As noted below there are strenuous efforts being made to identify surrogate markers that predict stage and likely outcome in NAFLD but none of these has yet been fully validated.

Liver tumours and precursor lesions

For a number of decades, liver biopsy (generally through interoperative wedge biopsy sampling) played a central role in the staging of a number of malignant neoplasms, in particular Hodgkin's disease and non-Hodgkin's lymphoma. With advances in imaging modalities this is now only very rarely undertaken and cannot be regarded as being part of the standard staging protocol in such disorders. The role of biopsy in the management of individuals with suspected hepatocellular carcinoma is highly controversial²³⁻²⁵. Many centres have now moved to using fine needle aspiration biopsies for the detection of hepatocellular carcinoma where the sensitivity is around 90% or even greater for lesions over 3cm. However, there has been increasing concern of the risk of needle track seeding following biopsy²⁴. At the turn of the century consensus guidelines were drawn up by EASL which reduced the protocolised indications for biopsy in patients with suspected hepatocellular carcinoma²⁵. The role of tissue interpretation being considered for transplantation remains a matter of debate^{23, 24}. What is clear however is that assessment of needle biopsy material is unlikely to provide useful prognostic information based on tumour grade given the heterogeneous nature of many HCCs. Identification of changes that are thought to represent precursor lesions for hepatocellular carcinoma, pose difficult problems for the diagnostic hepatopathologist. There is now firm evidence that the small cell dysplasia may indeed be a precursor lesion but in large cell change the cells are incapable of further division and therefore would not in themselves lead to tumourogenesis²⁶. Nevertheless both appear to be useful surrogate markers for an increased risk of the development of hepatocellular carcinoma and patients who have such changes on biopsy need to be more closely monitored for the development of neoplasms. Fleming et al²⁷ have stressed the importance of identifying dysplasia in bile duct epithelium in patients with primary sclerosing cholangitis as a marker of likely progression to cholangiocarcinoma.

Liver transplantation

The lack of correlation between liver function test abnormalities and tissue injury has already been referred to and it is no less important in the post-transplant patient. Liver biopsy emerged in the early days of orthotopic transplantation as an invaluable tool in assessing not only the presence but the severity of acute cellular rejection and there has been increasing awareness that early post allograft changes may predict longer term graft survival; in particular the presence of perivenular necrosis has been recognised to be a signal for increased risk of subsequent chronic rejection²⁸. Biopsy findings are also invaluable in predicting the likely rate of progression in

recurrent hepatitis C infection and in other recurrent disorders including primary biliary cirrhosis.

Drug-induced injury

Liver biopsies can be used to determine likely outcome in several forms of drug-induced injury; perhaps this is best seen with agents that produce a steatohepatitis eg. amiodarone and methotrexate which is used widely in the treatment of psoriasis and rheumatoid arthritis. Abnormal liver function tests documented in the face of long term amiodarone therapy cannot be used to predict the grade and stage of drug induced steatohepatitis and the likely progression to advanced disease with cirrhosis in this disorder can only be achieved through histological examination. Historically dermatologists have obtained base line biopsies in patients to be commenced on methotrexate; rheumatologists have only generally done this if there were other features to suggest possible pre-existing chronic liver disease. Repeat biopsies have regular intervals have also been used to monitor any progressive fibrosis, a recognised complication of long term methotrexate injury and significant fibrosis is seen as a contra-indication to continued therapy. A recent study from our centre has suggested that current protocols may lead to over biopsying in patients on long term methotrexate²⁹.

The fall of liver biopsy in prognostication?

Inter- and intraobserver variations

The histological assessment of severity in chronic hepatitis was put on a firm footing by the deliberations of an International Hepatopathology Study Group in the late 1960s³⁰. From this emerged a qualitative approach where biopsy findings were categorised into chronic persistent, chronic active or chronic lobular hepatitis. Knodell and colleagues³¹ in the late 1970s developed a semi-quantitative scoring system for assessing the severity of injury; a number of individual histological features were assigned a score and the sum of such scores provided the histological activity index. Others subsequently argued for the separation of assessment of necro-inflammation from that of fibrosis (in other words grade and stage), this is a central feature of the two most widely used systems in current practice, Ishak's modified HAI³² and the METAVIR score³³. There has been an unfortunate tendency for clinicians to consider such semi-quantitative scores as being hard data which forms part of a linear relationship with severity of disease. Although in the Ishak HAI individual components of the necro-inflammatory score are separately identified, summation is often used and this is of questionable biological validity. Hepatopathologists have been scrupulously open in flagging up that the semi-quantitative approach is open to both inter and intra-observer variations and this has been seized upon by those who would wish to see biopsies "fall from grace". In reality however providing those involved in scoring meet regularly to cross-validate and quality control the Kappa values are highly satisfactory. Similar arguments have been raised about the value of biopsy interpretation in fatty liver disease. Early indications are that the recently developed NASH activity score can provide equally acceptable Kappa values.

Sampling difficulties

In addition to problems of variation in interpretation, hepatopathologists have drawn attention to potential pit falls in relation to sampling^{34, 35}. A needle biopsy of liver represents at best 1/50,000th of the entire organ and many disorders, perhaps most notably the cholestatic conditions PBC and PSC are not evenly distributed throughout the liver. Recent studies have shown that this is also a feature of non-alcoholic fatty liver disease. Bedossa and colleagues³⁵ using computer modelling have indicated that accurate grading and staging in chronic hepatitis requires a biopsy of greater than 25mm, sadly an uncommon specimen in modern hepatological practice! Again these

limitations have been highlighted (perhaps over highlighted) by those who would prefer to see non-invasive methods replace liver biopsy in diagnosis and prognostication.

Development of clinical algorithms and proteomic/genomic approaches

Over the past decade there have been quite dramatic advances in evidence based practice in hepatology with the development of clinical algorithms; these will be dealt with in a later presentation in detail. For example Poynard and colleagues³⁶ have identified key clinical factors which are independently associated with rate of progression of fibrosis and hepatitis C and have developed the so-called Fibro Test based on simple biochemical measurements. Panels of surrogate markers in serum of liver fibrosis have also been described³⁷ and both the Fibro Test and serum marker studies have been shown to predict the stage of disease at two ends of the spectrum but remain of poor sensitivity for those with intermediate degrees of injury. Alternative approaches using proteomics or genomics in peripheral blood samples are also being actively pursued and again will be the subject of another presentation in this symposium.

Conclusion

Liver biopsies have remained as a core diagnostic tool in clinical hepatology over the past half century. In a number of areas information from the biopsy provides invaluable prognostic information. It has long been regarded as the “gold standard” for evaluation of liver diseases but increasing awareness of the sampling difficulties and problems of inter and intra-observer variation in grading and staging mean that we should probably regard it as being a “silver standard”. There is increasing attention being given to developing non-invasive alternative approaches to liver biopsy, particularly in prognostication but those who predicted the demise of the liver biopsy in the molecular era are yet to be vindicated.

References

1. van Leeuwen DJ, Wilson L, Crowe DR. Liver biopsy in the mid-1990s: questions and answers. *Semin Liver Dis* 1995;15:340-59.
2. Movitt ER. Differential diagnosis of regurgitation jaundice; the role of needle liver biopsy. *Ann Intern Med*, 1954;40:932-51.
3. Savonuzzi G, Trincas M, Remelli L. One hundred open air hepatic biopsies. *Arcisp S Anna Ferrara*, 1955;8:543-50.
4. Tyor MP, Cayer D. The clinical and experimental value of needle biopsy of the liver. *Gastroenterology*, 1952; 21:245-53.
5. Weisbrod FG, Schiff L et al. Needle biopsy of the liver; experiences in the differential diagnosis of jaundice. *Gastroenterology*, 1950; 14:56-72.
6. Ishak KG, Legolvan PC, Salib M et al. Needle biopsy of the liver and spleen in schistosomiasis; a histopathologic study. *Am J Clin Pathol* 1959; 31: 46-59
7. Bowden L, Kravitz S. Needle biopsy of the liver; a diagnostic aid in the treatment of cancer. *Cancer*, 1953; 6:1010-20.
8. Sheela H, Seela W, Caldwell C, Boyer JL, Jain D. Liver biopsy: Evolving role in the new millennium. *J Clin Gastroent* 2005; 39:603-610.
9. Campbell MS, Reddy KR. Review article: the evolving role of liver biopsy. *Aliment Pharmacol Ther* 2004; 20:249-259.
10. Bravo AA, Sheth SG, Chopra S. Liver biopsy. *N Engl J Med* 2001; 344:495.
11. Stanley AJ, Haydon GH, Piris J, Jarvis LM, Hayes PC. Assessment of liver histology in patients with hepatitis C and normal transaminase levels. *Eur J Gastroenterol Hepatol* 1996; 8:869-72.
12. Yano M, Jumada, H, Kage M, et al. The long-term pathological evolution of chronic hepatitis C. *Hepatology* 1996; 23:1334-40.
13. 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.
14. Scheur P J. Pathological aspects of viral hepatitis In: *Hepatology: A Festschrift for Hans Popper*. Eds Brunner H, Thaler H. Rave Press 1985; pp.109-117.
15. Fattovich G, Brollo L, Giustine G, et al. Natural history and prognostic factors for chronic hepatitis type B. *Gut* 1991; 32:294-8.
16. 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.
17. 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.
18. Dienstag JL, Goldin RD, Heathcote EJ et al. Histological outcome during long-term lamivudine therapy. *Gastroenterology* 2003; 124:105-17.

19. Chedid A, Mendenhall CL, Gartside P et al. Prognostic factors in alcoholic liver disease. *Am J Gastroenterol* 1991; 86: 210-304
20. Nissenbaum M, Chedid A, Mendenhall C et al, Prognostic significance of cholestatic alcoholic hepatitis. *Dig Dis Sci* 1990; 35: 891-896
21. Matteoni CA, Younossi ZM, Gramlich T, et al. Nonalcoholic fatty liver tissue: a spectrum of clinical and pathological severity. *Gastroenterology* 1999; 116:1413-9.
22. Teli MR, James OF, Burt AD, et al. The natural history of non-alcoholic fatty liver: a follow-up study. *Hepatology* 1995; 22:1714-9.
23. Marsh JW, Dvorchik I. Should we biopsy each liver mass suspicious for hepatocellular carcinoma before liver transplantation? - yes. *J Hepatol* 2005; 43:558-62.
24. Stigliano R, Burroughs A K. Should we biopsy each liver mass suspicious for HCC before liver transplantation: -- No, please don' t. *J Hepatol* 2005; 43:563-568.
25. Bruix J, Sherman M, Llovet JM, et al. Clinical management of hepatocellular carcinoma. Conclusions of the Barcelona-2000 EASL conference. *J Hpatol* 2001; 35:421-30.
26. Ojangurren I, Castella E, Ariza A et al. Liver cell atypias: a comparative study in cirrhosis with and without hepatocellular carcinoma. *Histopathology* 1997; 30: 106-112
27. Fleming KA, Boberg KM, Glaumann H et al. Biliary dysplasia as a marker of cholangiocarcinoma in primary sclerosing cholangitis. *J Hepatol* 2001; 34: 360-365
28. Gomez R, Colina F, Moreno E et al. Etiopathogenesis and prognosis of centrilobular necrosis in hepatic grafts. *J Hepatol* 1994; 21: 441-446
29. Aithal GP, Haugk B, Das S et al Monitoring methotrexate-induced hepatic fibrosis in patients with psoriasis: are serial liver biopsies justified? *Aliment Pharmacol Ther* 2004; 19: 391-399
30. De Groote J, Desmet V, Gedigk P et al, A classification of chronic hepatitis. *Lancet* 1968; ii: 626-628
31. Knodell RG, Ishak KG, Black WC et al, Formulation and application of a numerical system for assessing histological activity in asymptomatic chronic hepatitis *Hepatology* 1981; 1: 431-435
32. Ishak K, Baptista A, Bianchi L et al. Histological grading and staging of chronic hepatitis *J Hepatol* 1995; 22: 696-699
33. Bedossa P, Poynard T et al. An algorithm for the grading of activity in chronic hepatitis C. *Hepatology* 1996; 24: 289-293
34. Scheuer PJ Liver biopsy size matters in chronic hepatitis: bigger is better. *Hepatology*. 2003; 38:1356-8.
35. Bedossa P, Dargere D, Paradis V Sampling variability of liver fibrosis in chronic hepatitis C *Hepatology* 2003; 38: 1356-1358
36. Poynard T, Bedossa P, Opolon P. Natural history of liver fibrosis progression in patients with chronic hepatitis C. The OBSVIRC, METAVIR, CLINIVIR and DISVIRC groups. *Lancet* 1997; 349:825-32.

37. Rosenberg WM, Voelker M, Thiel R et al, Serum markers detect the presence of liver fibrosis: a cohort study. *Gastroenterology* 2004; 127: 1704-1713