EDITORIAL

BEYOND HEPATITIS A AND B

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INTRODUCTION

Viral hepatitis is not a new disease. It was described by the ancient Greeks,[1] Hippocrates was thought to have written the first record. Since then there have been numerous accounts, particularly during wars. In the 1940s and 1950s[2] there were two major types of viral hepatitis termed infectious and serum hepatitis. With the discovery of HBsAg in the 60s and hepatitis A virus in the 70s, viral hepatitis was classified as hepatitis A, hepatitis B and non-A, non-B hepatitis. Then delta antigen, hepatitis D virus was identified and in 1989,[3] successful cloning of an antigenic component of the hepatitis C virus (HCV) was reported and quickly came the realisation that beyond hepatitis A and B there were hepatitis, C, D, E, and possibly hepatitis F and G.

HEPATITIS C

Hepatitis C virus (HCV) accounts for about 90% of post-transfusion chronic hepatitis[4] and 50% of sporadic non-A, non-B hepatitis in the Western world. It is a frequent cause of chronic hepatitis in the United States, Japan, Australia, and many parts in southern Europe. Some 500 million people worldwide are believed to have been infected with HCV.

Hepatitis C is an enveloped RNA virus of 30-80 nm in size. It is single stranded and the complete 9400 basepair nucleotide sequence of the HCV genome has been fully sequenced. The genome contains a single open reading frame that is cleaved into 3 putative structural proteins and six non-structural proteins. HCV mutates at a rapid rate thus there is genomic sequence variability among different HCV isolates. On the basis of the nucleotide sequence of the genome, HCV has been classified into distinct genotypes. Several systems are in use but a nomenclature system proposed by an international panel[5] is likely to being universally accepted. In this system genotypes are designated 1-6 with subtypes a, b, c, etc. There is marked geographic variation in dominant genotype distribution. Different HCV genotypes have been reported to be associated with differences in levels of viraemia, pathogenicity, and responsiveness to interferon therapy. Most studies found that genotype 1b is associated with higher HCV RNA levels, more active or advanced liver disease and poorer response to interferon therapy[6,7].

The original antibody test used was a first generation enzyme immunoassay(EIA) directed against serum antiC 100-3, an antigen from the nonstructural region of the virus. There were high false positive and false negative rates (both 20%) when used with low risk populations such as blood donors. The second generation assays detect antibodies to a combination of structural and non-structural antigens. Sensitivity and specificity of these tests are about 99%[8]. These assays detect IgG antibody and cannot differentiate between recent and longstanding infection. Assays to detect IgM antibodies are being investigated in some research laboratories. Supplemental assays to confirm a positive EIA test as reflecting true hepatitis C infection include immunoblot assays (eg RIBA-2) and dual bead assays (eg Abbot supplemental Assay). They comprise similar antigens to those incorporated in EIAs but presented in a different form. Direct viral detection and quantitation is possible using molecular biological techniques. The most common is polymerase chain reaction (PCR). Serum HCV RNA is of value in early diagnosis of acute HCV infection, being detectable between 1 to 3 weeks after clinical onset whereas with first generation tests for HCV, antibodies appear between 4 to 24 weeks (mean 15 weeks). A new assay, the branched-chain DNA assay or b-DNA assay, measures viral "signal" or probe rather than viral nucleic acid. Values are expressed as viral equivalents per millilitre. While PCR has some advantage over b-DNA assay, the b-DNA is automated and is easier to perform.

The prevalence of positive HCV antibodies in blood donors vary worldwide. In New York City anti-HCV was found in 0.9% to 1.4% [9], in the UK it was 0.5%, in Japan 1%, in Italy 0.9%, in France 0.7%, and 0.4% in Germany[10]. In Singapore, it was found in 0.3% of blood donors[11]. About 500 million people in the world are believed to have been infected with HCV. Only about 25% give a history of blood transfusion, and in 50% the mode of infection is uncertain[4]. Others had histories of intravenous drug abuse. A history of tattoos is an independent risk factor for hepatitis C infection[12]. The practice of various folk remedies in particular, acupuncture and the use of a non-sterile knife in the popular "SUIDAMA" therapy were reported with an increased risk of hepatitis C and B[13]. Occupational exposure to blood accounts for a small number of cases. About 1.7% of New York dentists were anti-HCV positive, compared with 9.3% of oral surgeons and 0.14% controls suggesting occupational exposure to blood as a risk factor of infection[14]. The risk of HCV transmission from a needle-stick accident is 10% if there is HCV RNA blood on the needle. Sexual transmission is rare. The risk is clearly increased by co-infection with human immunodeficiency virus (HIV). Five female partners of 231 repeatedly transfused haemophiliacs were found to be anti-HCV positive but their partners were co-infected with both HCV and HIV[15]. Nonetheless, having multiple sexual partners has been identified as a risk factor of hepatitis C infection and homosexuals have an increased risk of hepatitis C (4%-8%) compared with controls[4]. Vertical transmission from mothers to baby is rare unless the mother is also co-infected with HIV[18].

The majority of patients infected with hepatitis C virus have mild flu-like symptoms or subclinical infection. Only 15%-20% become symptomatic with jaundice. The mean incubation period is 7.8 weeks[17] with 80%-90% of patients developing within 6-12 weeks of a transfusion. In the acute stage, serum ALT is up to 800IU in 50% of cases and greater than 2000IU in only 10% of cases. At least 50 (and probably 80%) fail to clear the virus[18]. Those failing to eradicate the virus may be normal or have a

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typical fluctuant (yo-yo) course over many years. Monophasic or biphasic elevations in transaminase levels have also been noted. There do seem to be some "healthy carriers" of HCV with extremely low levels of viral replication and normal liver histology but most individuals will have some hepatic inflammatory changes. The course of chronic hepatitis C is usually one of slow progression; older age, concomitant alcohol use, and concurrent HBV or HIV infection may be important aggravating co-factors. From a study reported by Bisceglie it appears that about 20% of patients with chronic hepatitis develop cirrhosis after 10 or more years of infection[19]. However, Seef found only a slight increase in liver-related deaths in subjects with non-A, non-B hepatitis (1.9%) compared to controls after an average follow-up of 18 years[20]. Much work needs to be done to identify the factors that determine the rate of progression of HCV related liver disease.

There is now substantial epidemiologic evidence linking chronic HCV infection and the development of HCC. A high prevalence of anti-HCV in patients with HCC has been identified around the world. The role of HCV becomes important particularly in area where the prevalence of HBV is low. Thus, in Japan, 76.2% of 105 patients with HCC had anti-HCV and in France anti-HCV was present in 58.2% of patients with HCC, compared with 29% in South Africa and 30% in Taiwan[4]. Hepatoma takes many years to develop. Kiyosawa[21] found the mean interval of transfusion to identification of HCC to be 29 years. In a prospective study Ikeda[22] found the appearance of HCC in 10.4% by 3 years, 21.5% by 5years, 72.5% by 15 years. Apart from these sequelae, there is a growing number of reported extra-hepatic manifestations of chronic hepatitis C infection. In 98% of patients with essential mixed cryoglobulinaemia[23], a condition characterised by purpura, arthralgia and renal impairment, anti-HCV can be demonstrated. Polyarteritis nodosa, membrano-proliferative glomerulonephritis[24] and Sjogren's syndrome have been reported to be associated with hepatitis C. First generation anti-HCV tests produced false positive results in patients with rheumatoid factor, but HCV infection has not been reported in association with clinical rheumatoid arthritis[4]. Aplastic anaemia, a well recognised complication of non-A, non-B hepatitis is currently believed not to be related to HCV[4].

There are conflicting reports regarding the occurrence of hepatitis C antibodies in patients with autoimmune liver diease. The ELISA assay for anti-HCV is prone to false positive results in these patients with high immunoglobulins in their serum. These false anti-HCV results are usually in low titre and, when HCV RNA test is used, the results prove to be negative. The anti-HCV may actually disappear with immunosuppressant treatment as globulin levels decrease[25]. However, there is a small subgroup of patients who have anti-HCV positivity and autoantibodies who respond poorly to prednisolone but may benefit from interferon therapy. It is possible that some patients with type 1 autoimmune hepatitis have chronic hepatitis C infection. Alternatively, autoantibodies in autoimmune hepatitis patients may cross react with HCV related antigens to produce the anti-HCV positivity. In addition, some patients with autoantibodies found in type 2 autoimmune hepatitis (anti-liver kidney microsomal, Anti-LKM1 positive) are anti-HCV positive. These patients respond to alpha interferon and not to immunosuppressant therapy, suggesting a viral aetiology. The low titre anti-LKM-1 appears to be an epiphenomenon in patients with chronic HCV infection and is not a reflection of an ongoing autoimmune process. This association between HCV and autoimmune hepatitis is important to clinicians as treatment of autoimmune hepatitis relies on immunosuppressives, whereas chronic hepatitis C is treated with interferon. When interferon is given to autoimmune hepatitis patients it could cause clinical deterioration. Currently, confirmation of HCV infection by PCR appears to be the only reliable course.

Hepatitis C infection as determined by HCV RNA can be found in 11%-46%[26,27] of patients with alcoholic liver disease. HCV infection lowers the threshold for severe liver injury in alcoholics and vice versa. The fact that clinically advanced alcoholic liver disease occurs at a substantially younger age in patients with HCV RNA than in patients without HCV RNA, also argues for synergy between HCV RNA and alcohol.

The management of acute sporadic or transfusion related hepatitis is largely non-specific and supportive. Therapeutic trials of alpha interferon have been undertaken which have indicated that interferon may help to prevent the progression to chronic disease. Further studies are needed to substantiate these results. There is more data on the treatment of chronic hepatitis C with interferon. The dose of alpha interferon used is 3 million units thrice weekly for 6 months. This produces normalisation of ALT in approximately 50%[28] of patients. Failure of normalisation of ALT by 12 weeks of treatment is strongly predictive of non-response and should lead to cessation of therapy. Relapse occurs in about 50% of patients usually within 6 months of stopping interferon. Long-term response is thus approximately 20%-25%[28]. If patients remain in remission at 12 months, there is a high likelihood of maintaining remission for at least eight years[29]. The most important predictive of response to alpha interferon are absence of cirrhosis, younger age, level of HCV RNA and possibly viral genotype. It has been suggested that extending the treatment to 9 months or 12 months may reduce relapse but this is not a consistent finding. Side-effects include fever and chills, fatigue, alopecia, bone marrow suppression. Auto antibodies are detected transiently in up to 50% of patients treated and symptomatic thyroiditis may occur in 5% of patients. Other anti-viral therapy, such as ribavirin with interferon, and lymphoblastoid interferon, are currently being evaluated. In a study of patients who relapsed or failed to respond to interferon alone, the combination of ribavirin and interferon resulted in sustained response in 40% of patients and this was always accompanied by a loss of HCV RNA in serum[30]. This result needs to be substantiated.

Patients with decompensated hepatitis C cirrhosis have successfully undergone liver transplantation. Recurrence of hepatitis C infection is common and 90% of the transplanted patients will be HCV RNA positive again. In general, post transplant hepatitis is mild but progression to chronic active hepatitis occur in 28%[4] of recipients and cirrhosis develop in 3%[4]. Actuarial survival rates are 94%-95%, 89%-90%, and 87% at 1, 2, 3 years respectively in two large series[31,32]. A large, recent series from Pittsburg suggested an adverse effect of HCV infection on outcome when compared to non-viral liver disease[33].

Preventive measures are needed to limit the spread of this infection. Important public health measures include screening of blood donors for anti-HCV which is routine in most blood banks. Vaccination of high risk individuals would also be effective. However, there are many difficulties associated with the development of a vaccine. They are: the genomic heterogeneity of HCV, its high mutation rate, inability to culture HCV and the lack of cross-protection against heterologous and homologous strains.

HEPATITIS D

Hepatitis D Virus (HDV) initially known as delta antigen is a RNA virus. The virus exists only with hepatitis B virus as helper. In areas of high prevalence, spread is largely perinatal and intrafamilial, but intravenous drug abuse is a significant factor in areas of low prevalence. Sexual transmission has been reported from Taiwan. The diagnosis of acute HDV infection is made by

detecting HDV antibodies. These antibodies appear months after the onset and finally disappear with recovery. The best diagnostic method is by HDV RNA in serum and tissues but this is not routinely available. It is found within a month after onset of infection and is present in chronic hepatitis. Alpha interferon given for a 12-month period (5 million unit thrice for 4 months, followed by 3 million units for 8 months) results in reduction of ALT in 60% of patients but this was sustained in only 25% of cases[34]. After 12 months of therapy only 4% maintained a normal ALT. Results with a higher dose of interferon, 9 million units thrice for 12 months were more encouraging producing normalisation of ALT in 71% of patients and were sustained in 50% of patients[35].

HEPATITIS E

This RNA virus is enterically transmitted and resembles hepatitis A in its clinical course. It has been associated with water-borne epidemics in Asia and Africa and, more recently, Mexico. The peculiarity of this infection is the high mortality seen in pregnant women. Serological assays for the detection of IgM and IgG antibodies to hepatitis E virus (anti-HEV) are available. Although hepatitis E is a self-limiting infection, significant morbidity and mortality can occur, particularly during epidemics. There is thus a need for a vaccine to be developed.

HEPATITIS NON A-E

Several observations point to the existence of as yet unidentified hepatic virus(es). About 11%-40% of post-transfusion non-A, non-B hepatitis and 18% of community acquired acute non-A, non-B hepatitis are negative for HCV markers. More than 95% of cases of non-A, non-B fulminant hepatitis are not due to HCV or HEV. Hepatitis associated aplastic anaemia were shown to be negative for hepatitis C. If a single agent were responsible for these conditions, it will presumably be designated as hepatitis F. However, there is some evidence from Japan that some of these so-called hepatitis F are actually caused by a hepatitis B virus mutant[35]. Sequencing of HBV DNA revealed mutation in the X gene coding region. These mutations may lead to suppression of replication and expression of HBV DNA and serological markers. However, two groups have announced the identification of a virus[36] tentatively named hepatitis G virus (HGV), HGV has been shown to have partial homology to HCV and may be responsible for some cases of non-ABC chronic hepatitis. The next decade will witness the characterisation of these virus(es) using molecular biological techniques.

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