Single Theme Consensus Conference on “Hepatitis B and C: Carrier to Cancer”

ABSTRACTS

Hepatitis B

Spectrum of hepatitis B infection in family contacts. S Nijhawan, RR Rai, R Pokharna, S Nepalia, P Puri, R Singh. SMS Medical College, Jaipur

Introduction: Hepatitis B is known to spread by percutaneous, sexual and perinatal routes. There are other unknown nonpercutaneous suspected routes of spread responsible for familial aggregation of hepatitis B.

Aim: To study the spectrum of HBV infection in families having more than one case.

Patients: The families of 18 subjects of HBV infection who had two or more cases of HBV infection in the family were studied. All members of the family were subjected to clinical examination, AST/ALT levels and viral markers of HBV infection (HBsAg, IgM anti HBC, and HBeAg).

Results: The results are shown in the following Tables.

A) Families Subject screened screened Naxed HBsAg+ incidence among families
18 10 50 45.45%

There were 8 families each of 2 cases and 3 cases, 1 family each of 4 cases and 6 cases.

B) Suspected mode of spread

HWS 14 2 1 9 20
WV 4 5 0 1 15
Stage of disease: Carrier [8] [1] [0] [5] [15]
Chronic Hepatitis [3] [0] [1] [3] [2]
Acute Hepatitis [3] [2] [0] [1] [2]
Cirrhosis [0] [1] [1] [1] [1]

H- Husband, W- Wife, S- Sibling, M- Mother, F- Father.

Conclusion: Suspected sexual contact is responsible in 36% of the cases, suspected perinatal transmission is responsible in 20% patients. Unknown mode of spread is suspected in 44% patients. 58%, 18%, & 8%, were carriers, chronic hepatitis, acute viral hepatitis and cirrhosis, respectively.

HBsAg positivity rate among voluntary and replacement donors in the IRCB blood bank. SK Chaudhuri, Director, Blood Bank, IRCB, New Delhi

This study was undertaken with a view to know the HBsAg carrier status among the blood donors in the Red Cross Blood Bank as a part of augmenting the blood safety standards.

The study presents a comparative year-wise study of hepatitis B positivity rate of voluntary and replacement donors, by using recorded data, in the IRCB Blood Bank. The positivity rate among voluntary donors in 1996 was 2.11% whereas the same among replacement donors was 2.74%, in 1997 and 1998 (till 30.9.98) the rates are 1.87% against 2.08% and 0.99% as against 1.41%, respectively. The main observation is that positivity rate among replacement donors, is more compared to voluntary donors, thereby giving an impression that professional blood sellers in the garb of replacement donors are being bled.

The overall positivity rate is also on the increase during the last 3 years which needs attention from blood safety point of view. A stringent donor screening has been advocated to eliminate professional blood sellers from the blood bank.

Epidemiology of HBV infection in the general population: impact of rural - urban difference and socio-economic factors. Abhijit Chowdhury, Amal Santra, Sujit Chaudhuri, Prabir Banerjee and DN Guha Mazumder. Department of Gastroenterology, Institute of Post Graduate Medical Education and Research, Calcutta - 700 020

Objectives: Rural - urban and socioeconomic differences are important variables in the epidemiology of HBV infection for planning the preventive strategy. As part of an ongoing community based study, we have tried to look into the impact of these factors on the HBV carrier rate.

Methods: The rural sample (total - 2266, male - 1229 and female - 1037, age group 25 days to 89 years) was drawn from 7 villages in 7 different CD Blocks of one district in West Bengal. The urban sample (total - 586, male - 348 and female - 238, age - 2 months to 76 years) was drawn from two different housing estates in North & East Calcutta. Sera was analyzed for HBV markers and a questionnaire regarding risk factors was administered.

Results: Overall carrier rate was 5.1% (male 5.5%, female 4.7%). While the carrier prevalence rate was 6.22% in rural areas, it was only 1.02% in urban areas. Age less than 20 years, male sex, poor socio economic status, illiteracy and history of injections were the significant associations of the higher HBV carriage in rural areas. All the carriers were clinically asymptomatic.

Conclusion: Rural poor are the main reservoirs of HBV carriers and preventive strategies including health education on sterile injection need to be stressed in these areas.

Pre-core mutants of HBV and the clinical outcome in chronic liver disease. Sujit Chaudhuri, Bhaskar Bikash Pal, Abhijit Chowdhury, Amal Santra, Prabir Banerjee and DN Guha Mazumder. Department of Gastroenterology, Institute of Post Graduate Medical Education and Research, Calcutta - 700 020

Infection by precore mutants of HBV are known to be associated with a worsened prognosis and altered clinical outcome as compared to wild type of virus. The present study was done to assess the prevalence of the precore mutant HBV infection in chronic liver disease and to see whether their clinical presentation was different from that of infection with the wild type.

Methods: 64 consecutive patients of chronic HBV infection were studied. Precore mutants were defined by HBsAg positivity, HBV-DNA positivity, HBeAg absence and Anti-HBe positivity.

Results: 9/64 (14%) patients of chronic HBV infection
had the pre-core mutation. No significant difference was observed in respect of age, sex, clinical presentation, severity of liver illness and prognosis.

**Conclusion:** The course of liver disease is found to be similar in both precore mutants and wild type of chronic HBV infection.

**Antibody response to hepatitis B vaccine in thalassemic children.** Harjeev Singh, Mandakini Pradhan* and Sita Naik. Departments of Immunology and Genetics*, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow, Uttar Pradesh

Multiple transfused thalassemic children have high risk of acquiring blood borne infections such as hepatitis B Virus (HBV), HIV. These children are vaccinated to check against HBV infection. The present study was undertaken to assess the prevalence of markers of hepatitis B Virus immunity and infection in thalassemic children.

Sixty-one thalassemic children (median age 5.8 yr.) studied were given prophylactic HBV vaccination (Engerix-B) at 0, 1 and 6 months interval. Of the 61, 55 (90.16%) showed response to the vaccine and their median anti-HBs titer was 85 IU/L. Six of the 61 (9.84%) children had anti-HBs levels below the protective level, 10 IU/L. Also, 3 of the 55 (5.45%) vaccine responders and one of the 6 (16.66%) non-responders were HBsAg positive.

Therefore the findings suggest exposure to HBV infection in 4 children despite vaccination. Of these 3 were in the vaccine responder group, suggesting the presence of mutant viruses in these cases. These observations have implications for developing vaccination strategies against HBV infection.

**Modulation of tumor necrosis factor-α production by hepatitis B virus in human monocytic cell line, THP-1.** R. Govindarajan and Sita Naik. Department of Immunology, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow, Uttar Pradesh

Viruses can modulate the immune response by coding for cytokines, chemokines (virokines), cytokine receptors (viraceptors), MHC class I homologues and many other molecules of immunological significance. Viruses can alter the state of activation/death of cells. Certain viruses can directly (LTRs of retro viruses) or indirectly (transactivating proteins of HBV, HIV) regulate the gene expression in the host cells.

Extra-hepatic infection by hepatitis B virus which belongs to the hepadna viridae family, is well documented. Since HBx protein can transactivate viral and host cellular genes and the recent observation that PMBCs can be infected by HBV, we investigated the possibility that HBV genes may transactivate expression of TNF-α in infected cells. As a model, human monocytic cell line THP-1 was transfected with plasmids expressing X and S1S2S genes of HBV. Spontaneous and LPS-induced TNF-α production by these cells was quantified by ELISA. HBx transfected THP-1 cells produced 25 to 95% more TNF than the HBs1S2S transfected cells and controls.

The results indicate that X gene of HBV upregulates TNF-α production by THP-1. This may be relevant to the pathogenesis of HBV-induced chronic liver disease.

**Prevalence of hepatitis B virus markers in high risk health care workers. R Polithama, RR Rai, S Nijhawan. Department of Gastroenterology, SMS Hospital, Jaipur**

**Background:** Health care workers (HCW) are at high risk for acquiring hepatitis B virus (HBV) infection, due to frequent exposure to blood and potentially infectious body fluids.

**Aims:** To find prevalence of HBV markers in HCW.

**Method:** Two groups, first consisting for surgeon practicing for more than 10 yrs and second group consisting of laboratory workers dealing with blood were studied.

**Results:**

<table>
<thead>
<tr>
<th>No.</th>
<th>Age (yr)</th>
<th>HBsAg (yes)</th>
<th>Anti HBs (mIU/ml)</th>
<th>non vaccinated HCW</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A (Surgeons)</td>
<td>141</td>
<td>38.8</td>
<td>4:1</td>
<td>2 (1.41%)</td>
</tr>
<tr>
<td>Group B (Lab. workers)</td>
<td>50</td>
<td>32</td>
<td>3:1:1</td>
<td>12%</td>
</tr>
</tbody>
</table>

**Conclusion:** HBV carrier rate in high risk HCW is not different from general population and it may be cost effective for HCW to have prevaccination anti HBs screening.

**Familial clustering of HBV infection in India. R Chakraborty, A Chowdhury, S Chaudhuri, PN De, S Chatterjee, D Chattopadhyay, and MS Chakraborty. I.C.M.R. Virus Unit, Calcutta and I.P.G.M.E. & R., Calcutta**

Family members of HBV infected individuals are known to be at increased risk of infection. The magnitude of intrafamilial transmission of HBV in our country has not been addressed in detail. This is important for planning of preventive strategies against the virus.

**Methods:** HBV seromarkers (HBsAg, anti HBc and/or anti HBs, HBV DNA) were studied in 264 household contacts of 90 HBsAg positive index cases (42 asymptomatic carriers, 20 acute hepatitis, 25 chronic liver disease and 3 hepatocellular carcinoma).

**Results:** 61 family contacts were positive for HBsAg. Anti HBc and/or anti HBs were positive in 119 out of 189 contacts tested. HBV transmission amongst family members of carrier, acute hepatitis, chronic hepatitis were not significantly different. Prevalence of HBV markers was commonest in siblings (80% of them were infected) of the index case. Exposure was commoner in mother than father when a child was the index case. There was no correlation with family size or socioeconomic status and the observed
familial clustering of HBV.

**Conclusion:** Intra family transmission is an important way of the viral spread, this is independent of the clinical status of the index case and sibling and parent children transmission being particularly common. Thus vaccination coverage is particularly relevant in the family setting.

**The prevalence of hepatitis D in hepatitis B patients a hospital based study.** UK Chattopadhyay, D Pal. All India Institute of Hygiene & Public Health, Calcutta-73

Patients with symptoms of hepatitis admitted to Medical College Hospital, Calcutta were investigated. The serum samples from the selected group of 257 jaundiced patients were tested by ELISA technique (Abbott Laboratory Diagnostic Kits) for the presence of HAV-IgM, HBsAg and HBcIgM. The HBV positive sera were further investigated for antidualt IgG antibody by ELISA method (Abbott). Out of total 257 jaundiced patients, 168 were positive for markers of Hepatitis B, cases of acute hepatitis were 75 and another 93 were patients of chronic liver disease. All the 168 patients were tested for antidualt (IgG) antibody and 55 were found to be positive (32.73%). The high association between HDV and HBV infections in the present study could be due to the fact that only hospitalised patients of hepatitis were studied. This could explain the prolonged course of the disease in most of these individuals. A detailed follow up study on a larger scale is necessary to investigate the incidence of HDV in the Indian population.

**HBSAg carrier rate amongst (15-45 years) married women in an urban slum of Delhi. Preliminary communication from RTI project.** Suneela Garg,* Nandini Sharma,* R Saha,* Preema Bhalia,** Uma Saran,** Departments of *PSM, **Microbiology, and ***Obstetrics & Gynecology, Maulana Azad Medical College, New Delhi

The HBsAg carrier rate was studied as a part to assess overall load of reproductive tract infection (RTIs) / sexually transmitted infections (STIs) amongst married women in the reproductive age group (15-45 years) in an urban slum of Delhi. The total population of the slum is 3676 and the total eligible women (15-45 years) are 485 in number. Out of 184 women screened so far, 12 (6.8%) have been found to be positive using latex agglutination test. Out of positive cases, 4 (33.3%) were pregnant. Observations about few of the associated risk factors were higher risk with increasing age and gravidity. Husbands of majority of the positive cases were either rickshaw pullers, casual laborers or petty shopkeepers. An exploration of the other associated risk factors is being undertaken and also the need-based intervention is being done for the community.

**Hepatitis B carrier – a study of possible routes of acquiring the infection.** SK Thakur, RM Gupta, MKK Rao, SK Dham. Army Hospital RR, Delhi Cantt and Armed Forces Medical College, Pune

The HBV carrier rate in India varies from 2-5%. The infection is acquired through parenteral exposure – both percutaneous and non-percutaneous. With a view to analyze the route of acquiring the infection among asymptomatic healthy carriers we studied the epidemiological data of 232 subjects. All were positive for HBsAg by ELISA technique, were asymptomatic and with no clinical or biochemical evidence of hepatocellular necrosis.

Meticulous history taking was resorted to such that all possible modes of disease transmission were covered. Among the 232 subjects analyzed 60 (25%) gave a past history of jaundice, 19 (8%) gave a history of exposure to commercial sex workers, 12 (5%) had received previous blood transfusions, 4 (1.7%) had previous prolonged hospitalization, 4 (1.7%) who had undergone surgical procedures in the past, 12 (5.7%) had a positive family contact. In the remaining 121 (52%) no history for a possible route of transmission was forthcoming. These results show that in more than half of the asymptomatic healthy HBV carriers the route of disease transmission is not apparent.

**Natural history of chronic hepatitis B – a follow up of untreated cases.** SK Thakur, PS Reddy, H Subramanya. Departments of Gastroenterology and Pathology, Army Hospital Research and Referral, Delhi Cantt 10

**Objectives:** To assess histologically the natural history of untreated mild chronic hepatitis B (CLH/CPH).

**Subjects:** 18 patients with biopsy-proven mild chronic hepatitis B. 14 had CLH and 4 had CPH.

**Method:** All patients had evidence of active viral replication. They were followed up without treatment. Biopsy specimens were obtained at entry and after 12-18 months follow-up. Knowell's histological activity index was used to assess the grade and stage of disease.

**Results:** At follow up 6/18 (4 CLH and 2 CPH) cases had progressed to moderate/severe chronic hepatitis (CAH).

**Conclusion:** One-third cases of mild CH-B (CLH/CPH) progress to moderate/severe CH (CAH) if left untreated.

**Chronic hepatitis B: a 7 years histological follow up of interferon treated patients.** SK Thakur, PS Reddy, H Subramanya, SK Dham. Departments of Gastroenterology and Pathology, Army Hospital Research and Referral, Delhi Cantt 10

**Objectives:** To assess the short and long term sequential changes of liver pathology after therapy with interferon alpha 2b in patients with chronic hepatitis B.

**Subjects:** 30 patients with biopsy-proven chronic hepatitis B.

**Method:** All patients had evidence of active viral replication. They received interferon alpha 2b 6 MU TIW S/C for 16 weeks. Biopsy specimens were obtained prior to treat-
ment and after completing treatment. Short-term follow up was done at 1-3 years and where possible (12/30) long-term follow up was done at 3-7 years.

Results: There was significant improvement in the HAI (p<0.05). While portal inflammation and intralobular necrosis regressed there was no improvement in the index of fibrosis. Short and long term follow up showed either persistence or further improvement of histological regression.

Conclusion: Interferon alpha 2b in a dose of 6 MU TIW SIC for 16 weeks is effective in inducing sustained histological improvement in CH-B.

Effect of Jigrine in HBV infected acute viral hepatitis patients: M M Hussain, SP Singh, SJ Khudmiri, N Farooq, MN Khaga and CM Habibullah. Centre For Liver Diseases, Owaisi Hospital and Research Centre, Hyderabad 500 058

HBV infection is a worldwide health problem. Recent HBV infection in adults generally leads to acute viral hepatitis. 90% of these patients clear the virus on its own. The remaining 10% of patients may become chronic carriers. There is no effective treatment for viral hepatitis B except alpha-interferon. However, the cost of treatment is very high. In the present study we evaluated the effect of a herbal drug Jigrine in acute viral hepatitis B patients. Jigrine has earlier been shown to be hepatoprotective in CCl4 and alcohol treated rats. Further, it has also been shown to be effective against WHBV. Forty acute viral hepatitis B patients were enrolled into the study after informed consent and were followed for 4 months. Their HBV profile was done before and after treatment. Clinical and biochemical evaluation was carried out every week during the follow up period. Our results show that 33 patients became negative for HBsAg; out of 17 HBeAg positive patients 15 became negative, 16 patients seroconverted to anti HBe while 21 patients developed antibodies to surface antigen. Out of 16 HBV DNA positive patients 12 became negative during follow up. The clinical and biochemical parameters will be discussed.

A multicentric open labelled evaluation of immunogenicity (seroconversion rate) and reactogenicity of a recombinant DNA hepatitis B vaccine of Cuban origin when administered in a two dose schedule. Kauskal Madan, Anil Jain, P Jandwani, RK Gupta, P Kar, US Mathur. Department of Medicine, Maulana Azad Medical College, New Delhi & SMS Medical College and Hospital, Jaipur

Introduction: Hepatitis B infection is an important public health problem all over the world. As no specific treatment is available, greatest emphasis is placed on prevention through immunization.

Objective: The present study was carried out with an aim to evaluate the immunogenicity (seroconversion rate) and reactogenicity of a novel recombinant DNA hepatitis B vaccine of Cuban origin (EnVac HB) when administered in a two dose schedule.

Population and Methods: The study was conducted at two independent centres (New Delhi and Jaipur) and 111 healthy volunteers without any symptomatic or serological evidence of HBV infection were administered the Cuban vaccine intramuscularly in a dose of 20 mg at day 0 and day 30. Blood samples were collected for evaluation of seroconversion at days 30, 60 and 90 at both the centres and, in addition, at day 120 at New Delhi. All subjects were evaluated for any adverse event for 120 hours subsequent to each dose of the vaccine.

Results: The mean age of the vaccinees was 23.94 years and the male:female ratio was 61:50. An overall seroconversion rate of 24.3% was obtained at day 30, 68.5% at day 60, 94.5% at day 90 and 99.1% by day 120. The vaccine was well tolerated with no serious adverse reactions. Minor side effects such as injection site tenderness, erythema and/ or low grade fever were observed in 4.5% of the subjects.

Conclusions: The recombinant DNA hepatitis B vaccine of Cuban origin is safe, well-tolerated and highly immunogenic and only two doses of this vaccine given one month apart can provide a acceptably good seroconversion rate vis-a-vis the conventional three-dose regimen.

Efficacy of the 1st indigenously developed recombinant vaccine against hepatitis B virus in Indian subjects. Navana Joshi, Ajit Kumar, Nagaraja Rao, M Girish Narayan, Nagarjuna Kumar YR and Sethu Babu. Dept. of Gastroenterology, Nizam’s Institute of Medical Sciences, Owaisi Medical Research Centre, Osmania General Hospital, CDR Hospitals, Hyderabad

The first genetically engineered hepatitis B vaccine produced in our country (Shanvac-B) has been clinically evaluated in different age groups and with different schedules. The anti HBs titers were determined using Abbott kits. The clinical trials showed 100% seroconversion in neonates and children with geometric mean titers (GMT) of 1741.63 and 1761.35 mIU/ml after the 3rd dose, following 0, 1, 2 and 0, 1, 6 months schedule respectively. Comparison of the indigenous vaccine with the commercially available vaccine has shown comparable seroconversion with high GMT. Comparison of 0, 1, 2 and 0, 1, 6 months schedules in adults showed 100% seroconversion with antibody titres of 749.12 and 6275 mIU/ml respectively. Low dose vaccine with 10 mg in young volunteers has also shown 100% seroconversion and GMT of 1674.28 mIU/ml after 3rd dose. Follow-up results in healthy adults at the end of 1 year before booster have revealed a GMT of 421.9 mIU/ml, while it was 622.6 mIU/ml after 3rd dose. In renal failure patients vaccine combined with different doses of leucovorin (colony growth stimulating factor) has shown 100% seroconversion compared to 50% observed in control group without leucovorin. The safety of vaccine was established in all the studies. Thus
Hepatitis B and C: carrier to cancer — Abstracts

Hepatitis B virus DNA in asymptomatic carriers: relationship with HBsAg/anti-HBs and HBeAg status. R Hari, KV Mohan, KG Murugavel, SP Thyagarajan. Department of Microbiology, Dr. ALM PGIBMS, Taramani, Madras 600 013

Objectives: To study the HBsAg status among HBsAg carriers and to correlate the HBV-DNA status with the 'e' Ag positivity and the HBsAg titer.

Design: 330 asymptomatic carriers of hepatitis B virus attending the Department of Microbiology, Dr. ALM PGIBMS, Taramani, Madras were selected and evaluated for their HBsAg status, anti-HBe status, HBV-DNA status and the titer of HBsAg.

Methods: HBsAg, anti-HBe and anti-HBc were tested using commercially available ELISAs. HBV-DNA was tested by the PCR technique. The titer of HBsAg was determined by titration using the Auszyme Macro ELISA (Abbott, USA).

Results: Of the 330 carriers tested, 57 carriers (17.27%) were HBsAg positive and 270 (81.8%) carriers were anti-HBe positive. 49 (86.1%) of the 57 'e' Ag positive carriers were positive for HBV-DNA, while 48 (17.7%) of the 270 anti-HBe carriers were positive for HBV-DNA. Titration of these 'e' Ag positive carriers revealed a titer of >1:10^9 and above while lower titers of 1:10^2, 1:10^3 and 1:10^4 were seen in the anti-HBe positive carrier group. In two HBsAg positive patients HBV-DNA was negative initially and later showed positivity on follow-up.

Conclusion: Higher titers of HBsAg were seen in 'e' Ag positive group (>1:10^9) when compared with anti-HBe positive group. Moreover the presence of HBV-DNA in the non-replicating carrier group reveals that they may also be infective. In view of the HBV-DNA positivity in 17.7% of anti-HBe positive carriers, the possibility of pre-core mutants is to be further analysed.

Evaluation of risk factors in HBV infected chronic hepatitis patients. MN Khaja, SJ Khudnir, MM Hussain, N Farees, DN Reddy, CM Habibullah. Centre for Liver Diseases, Osmania Hospital and Research Centre, Hyderabad; *Asian Institute of Gastroenterology, Medina Nova Hospital, Hyderabad

Viral hepatitis is an important health problem throughout the world, especially in India. HBV has worldwide distribution and the infection rates in some parts of the world like Hong Kong, Taiwan and South China ranges between 15-20% while in Korea and north China infection rates are above 10%. India has an intermediate range of HBV endemicity with carrier rate of about 4.7% and contributes 9% of the total HBV carriers. HBV is transmitted mainly by blood transfusion, IVDA and sexual contacts. In the present study we have evaluated the risk factors associated with HBV infection in 197 chronic hepatitis patients. We have also studied the markers of HBV infection in these patients. Our results show that 47 patients were positive for HBsAg, HBcAg and HBV DNA. 63 patients were positive for HBsAg and HBV DNA while 74 patients were positive for the surface antigen only. 2 patients had mutant virus infection while 4 were positive for surface and envelope antigens only. 4 patients were positive for HBV DNA only and 1 patient was infected with envelope mutant virus. 2 patients were negative for all markers. Among the risk factors evaluated prior surgery in the last 6 months and injury at the saloons were found to be the major cause of HBV infection. Following these blood transfusion, visit to a dentist and sexual transmission were common. 23 patients did not have any risk factors evaluated. The results will be discussed.

The work was partially supported by Fulford (I) Ltd.

Incidence of HBsAg positivity among various population subsets in the Armed Forces. M Jaiprakash, SK Thakur, MKK Rao. Departments of Gastroenterology and Armed Forces Transfusion Centre, Army Hospital R & R, Delhi Cantt-10

Hepatitis B is a major public health problem. We in India are in the intermediate zone of HBsAg positivity (2-5%).

Objectives: To study the prevalence of hepatitis B virus infection among various population subgroups in the Armed Forces.

Subjects: Armed Forces personnel — serving and retired and their close family contacts were screened for HBsAg positivity. These included healthy voluntary blood donors, health care workers, CRF patients on dialysis, patients at the malignant diseases treatment centre and multiply transfused patients.

Methods: Screening was done using ELISA technique.

Results: Prevalence of HBsAg positivity was as follows. Healthy voluntary blood donors 468/17,591 (2.66%), health care workers 36/332 (11%), CRF on dialysis 26/160 (16%), patients with malignancies 126/700 (18%) and multiply transfused patients 101/676 (15%).

Conclusion: This study shows that the prevalence of HBV infection in the Armed Forces is similar to that reported in the general population.

Immunohistochemistry for core and surface antigens in chronic hepatitis. RR Sharma, RK Vashist, RK Dhiman, Y Chawla. Departments of Pathology and Hepatology, Postgraduate Institute of Medical Education & Research, Chandigarh

Background: Hepatitis B virus infection constitutes a significant proportion of patients presenting to liver clinic with chronic hepatitis.

Aim: To study immunohistochemical staining of liver tissue for HBsAg and HbcAg in patients of chronic hepatitis

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and correlate it with histological activity index.

Methods: 100 cases of chronic hepatitis (65 M and 35 F) were selected. Exclusions were alcoholic liver disease, cystic fibrosis, drug induced chronic hepatitis and biliary cirrhosis. Histological scoring of liver biopsies was done using Knodell's numerical scoring system. Immunohistochemical staining was done by indirect immunoperoxidase technique using goat polyclonal anti-HBsAg and rabbit polyclonal anti-HBeAg. HBsAg in the serum was detected by ELISA.

Results: (1) Out of 100 chronic hepatitis cases, serum HBsAg was positive in only 40 patients whereas tissue HBsAg was positive in 48 patients. Out of 48 tissue HBsAg +ve cases, 13 patients also showed HBeAg on immunohistochemical studies. (2) Patients with higher grades of histological activity index (HAI) score had higher values of serum bilirubin and prothrombin time as compared to the patients with low HAI score. Significantly higher levels of serum transaminases (AST/ALT) were observed in patients who were positive for both HBsAg and HBeAg when compared with HBeAg positive patients or HBsAg negative patients. (3) 83.3% of HBsAg positive patients showed mixed pattern of surface antigen expression whereas expression of core antigen was predominantly nuclear (77%). (4) No significant correlation was observed between the pattern of antigen expression and HAI score.

Conclusion: Immunohistochemistry is a good modality to evaluate necroinflammation and in determining the etiology of chronic hepatitis cases.

Poor response to low dose interferon therapy in chronic liver disease due to hepatitis B virus infection in Indian patients. G Choudhuri, VA Saraswat, S Lakhtakia, SK Dedhich, P Mehrotra, SR Naik. Department of Gastroenterology, SGPGIMS, Lucknow

Background: The reported literature on treatment of chronic hepatitis B virus infection in Asians has shown conflicting results.

Aim: To assess virological and clinical response of low dose interferon therapy in patients of established hepatitis B related chronic liver disease in north India.

Patients & Methods: 19 consecutive patients of chronic active hepatitis without (14 cirrhosis, with elevated transaminases (ALT > 1.5 normal) and presence of HBeAg and/or HBV DNA in the serum were offered treatment with IFN a 2b ITW (or 3 MU when 5 MU was not tolerated) for 24 weeks. Therapy has been completed in 18 and is ongoing in one. Of 2 patients with cirrhosis who also received lamivudine 150 mg OD for 24 weeks, one has completed treatment.

Results: 12 patients completed 24 weeks of therapy. Initial biochemical remission occurred in 9 (75%) with breakthrough in 3 (25%). End of treatment virological response was observed in 7 (58%) with absence of HBV DNA in serum by PCR; however there was biochemical relapse with increase in ALT in 2 patients within 3 months of stopping treatment. Sustained virological response occurred at 6 months or more after end of therapy in 5 (42%), 2 of whom also lost the HBeAg at 6-18 months.

Conclusion: The eradication rate of hepatitis B with low dose IFN therapy is 42%. Probably higher doses or combination regimes with nucleoside analogues need to be tried to obtain better results.

Effect of Phyllanthus amarus, an Indian medicinal plant on healthy carriers of hepatitis B virus: results of six clinical trials - 1990-96. Thyagarajan SR,1 Jayaram S,1 Pannecorvam A,1 Valliammai T,1 Benjamin Samuel,2 Sheila Cameron,3 and Eric Walker,3 1Dept. of Microbiology, Dr. ALM PGIBMS, University of Madras, Chennai, India; 2Dept. of Nephrology, CMC Hospital, Vellore, India; and 3Scottish Centre for Infection & Environmental Health & Ruchill Hospital, Glasgow, UK

Background to study: Phyllanthus amarus, an Indian medicinal plant, has been traditionally used in Ayurveda and Siddha systems of medicine for the treatment of clinical jaundice. It has been reported to exhibit marked antihepatitis B virus and hepatoprotective properties besides safety. The reported clinical trials revealed inconsistency.

Objectives: To evaluate the clinical efficacy and reproducibility of Phyllanthus amarus treatment in clearing HBV from Indian and Scottish carriers.

Design: 214 healthy longterm HBV carriers were included in two double blind and four open clinical trials based on standard inclusion/exclusion criteria. Phyllanthus amarus and placebo were given in capsules of 250/500 mgs thrice daily for 3 to 6 months; HBsAg clearance, HBeAg seroconversion, HBV-DNA clearance were assessed during treatment and followup.

Results: The results of all the above trials have revealed that P. amarus treatment has a) brought about 20-25% HBsAg clearance Indian carriers and 13.0% HBsAg clearance in Scottish carriers, irrespective of whether they were originally HBeAg positive or negative. b) induced 25-57% HBeAg seroconversion in Indian carriers (mean: 35.4) and 33% in Scottish carriers. c) When HBsAg clearance rate was analysed in HBsAg and HBeAg positive groups and HBsAg positive and HBeAg negative groups, the observed outcome was: (i) In Indian carriers, HBsAg clearance in HBsAg positive group was 12.5-20% (mean = 18.5%). The same in HBeAg negative group was 36.1-100% (mean=40%). In Scottish carriers, the same was 0% in the HBeAg positive group and 21.3% in the HBeAg negative group. Significant reduction in HBV-DNA levels in HBeAg positive carriers was observed even though HBsAg persisted in them.

Conclusion: These results unambiguously demonstrated antihepatitis B viral drug potentials of Phyllanthus amarus. Chemical and biological standardisation of the P. amarus product, besides the nature of the soil are proposed as pivotal for the reproducibility of bioefficacy.
Hepatitis B and C: carrier to cancer — Abstracts

Retrospective analysis of children below the age of 2 years and pregnant women for HBsAg positivity. Chaddha MS, Shalik MS, Arankalle VA. National Institute of Virology (Indian Council of Medical Research), Pune

Objectives: To assess the extent of HBV infection in children below the age of 2 years and to evaluate the importance of vertical route in the transmission of HBV.

Methods: Serum samples from 293 children and 850 pregnant women were screened for HBsAg and all HBsAg positives were tested for anti-HBc IgM antibodies employing ELISA.

Results: HBsAg positivity was found to be 1.4% (1/69, up to 6 months), 3.2% (7/216, 7-12 months) and 2.5% (1-2 years) in children and 2.6% in pregnant women. Based on anti-HBc IgM positivity among HBsAg positives, probable vertical and horizontal transmissions in young children were evaluated.

Conclusions: The study emphasizes importance of perinatal and horizontal transmission of HBV.

Prevalence of pre-core HBV mutants among HBsAg carriers. Gandhe SS, Chaddha MS, Arankalle VA. Hepatitis Division, National Institute of Virology (Indian Council of Medical Research), Pune

Objectives: To evaluate the extent of presence of pre-C mutants in HBsAg carriers.

Methods: One hundred and one HBsAg carriers, 6 acute HBV patients and 5 fulminant hepatic failure (FHF) patients were examined for evidence of infection with wild and/or mutant HBV. All the samples were screened in nested PCR for HBV DNA in pre-core region. Restriction fragment length polymorphism (RFLP) analysis examining the characteristic single point mutation at nucleotide 1896 of the HBV genome was performed on all the HBV DNA positive samples.

Results: Approximately 59% HBsAg carriers were found to be positive for HBV DNA in pre-core region. Among the HBV DNA positive HBsAg carriers, 21.56% showed the presence of pre-C mutants, 60.78% showed wild type HBV whereas 17.64% had mixture of wild HBV and mutant HBV. One of the 6 acute HBV patients was infected with mutant HBV whereas 1/5 FHF patients examined was positive for both wild and mutant HBV.

Conclusions: Though substantial proportion of HBsAg carriers circulate pre-C mutants, the same was, at least not immediately, associated with abnormal liver functions.

HBsAg positivity among urban and rural populations of Pune (1982-1988). Arankalle VA, Chaddha MS and Gandhe SS. Hepatitis Division, National Institute of Virology (Indian Council of Medical Research), Pune

Objectives: To evaluate HBsAg prevalence among urban and rural populations over a period of 15 years.

Methods: A total of 6565 serum samples representing urban (Pune) and rural (Bhor) areas were evaluated for the presence of HBsAg employing ELISA. Urban samples were collected in 1982, 1992 and 1998 whereas rural populations were bled in 1983, 1987, 1995 and 1998. People living in the same area were bled each time.

Results: HBsAg positivity among urban children (<15 years) bled in 1982 (17/902, 1.8%) and 1992 (28/1290, 2.1%) was not different. However, a significant decline (p<0.05) was noted in 1998 (9/881, 1.0%). Among rural children, no difference in HBsAg positivity was noted in 1983 (8/195, 4.1%) and 1987 (11/359, 3.04%) followed by a significant drop in 1995 (4/458, 0.87%, p<0.001) which remained unchanged in 1998 (6/976, 0.61%). HBsAg positivity among urban and rural adults (>15 years) did not change with time. Age-related HBsAg positivity among rural and urban populations was not different.

Conclusions: The results show probable decline in HBV circulation in the areas under investigation.

Five year follow up of HBsAg carriers. Arankalle VA, Chaddha MS and Chobe LP. Hepatitis Division, National Institute of Virology (Indian Council of Medical Research), Pune

Objectives: To determine prognosis of Indian HBsAg carriers identified during blood donation. Methods: Seventy five HBsAg carriers identified during voluntary blood donation were clinically, biochemically and serologically followed at three month interval for two years. 34/75 carriers were followed for additional three years.

Results: At the time of first blood sample collection, the mean age was 30.4 (18-52 years); 8/75 (10.6%) carrier were females. None of the carriers gave a history of intravenous drug abuse. History of jaundice was given by 16% whereas 5.3% had received blood transfusion. HBsAg positivity was 9.3%. Majority (~80%) of the carriers were anti-HBe positive. During the 2 year follow up, serum ALT levels remained within normal limits and none of the carriers complained of symptoms indicative of hepatocellular failure. None developed anti-delta or anti-HCV antibodies. HBsAg titres in ELISA remained unchanged. Elavation of alpha-fetoprotein levels was not observed. Except for one HBeAg positive carrier seroconverting to anti-HBe with concomitant serum ALT rise and followed subsequently by lowering of HBsAg titre, parameters for other carriers remained unchanged.

Conclusions: The results suggest that HBsAg carriers identified during blood donation have a good prognosis.

Correlation of hepatitis B markers with serum enzymes and histopathology in various liver diseases. SM Vasenwala, A Malik, M Manjula, MU Rabbani. Dept of Pathology, *Microbiology and Medicine, JN Medical College, AMU, Aligarh

90 cases of liver diseases were studied. Investigations
performed were routine, serology for HBV markers, serum enzymes, prothrombin time (PT) and liver biopsy. 53/50 cases were positive for HBV markers as 23 (25.3%) for HBsAg, 40 (44%) for anti HBc and 28 (30.8%) for anti HBe. Serum bilirubin was raised in 40 (72%) AST in 34 (61.2%), ALT 38 (68.4%) and PT in 35 (63%) cases. On the basis of laboratory tests the 53 cases were categorized as acute hepatitis (27), 3 in window period, 7 in acute stage and 17 in convalescence; fulminant hepatitis (9), 3 in window period, 6 in acute stage; chronic hepatitis (4), 2 in chronic persistent hepatitis and 2 in chronic active hepatitis; cirrhosis (12) and hepatocellular carcinoma (1) case.

The HBV markers and serum enzymes help in diagnosis and prognosis of acute hepatitis while liver biopsy with HBV markers is a useful tool in detecting reactivation of disease in chronic hepatitis.

**Prevalence of hepatitis B virus in acute hepatitis in south India. Mohan Prasad, Ruby Thomas, Devaraj. Viral Hepatitis Research Laboratory, KG Hospital, Coimbatore**

**Objective:** This study was undertaken to determine the occurrence of hepatitis B among all cases of acute hepatitis in south India.

**Definition:** Acute hepatitis was defined as an acute onset of jaundice of less than 6 weeks in duration.

**Material:** 686 consecutive samples over a 2 year period from September '96 to September '98 were studied prospectively for the prevalence of hepatitis A to E viruses. Cases with history of alcoholism, drug intake were excluded, using a detailed questionnaire. Ultrasonography of abdomen was done to rule out obstructive causes of jaundice.

**Methods:** III generation ELISA kits of Sorin Biomedica were used to analyse HBsAg and IgM anti core antibodies in all samples.

**Results:** Hepatitis B virus was seen alone in 98 samples (14.28%). It was found in combination with hepatitis A and or hepatitis E viruses in 10 samples (1.45%). HBsAg alone or in combination with other viruses was seen in 27 samples (3.9%) wherein IgM core antibodies were negative, indicating that these cases were because of acute hepatitis due to other viral agents superimposed on HBsAg carrier state.

**Conclusion:** Hepatitis B is the cause of acute hepatitis in 15.73% of 686 consecutive cases analysed for hepatitis A to E viruses in south India.

**Prevalence of hepatitis “B” virus in voluntary blood donors in Coimbatore. Mohan Prasad, Ruby Thomas, Devaraj. KG Hospital, Coimbatore**

**Objective:** This study was undertaken to define the prevalence of HBsAg in healthy voluntary blood donors in and around Coimbatore in south India.

**Material:** Blood donors were both from blood donation camp conducted outside Coimbatore as well as from those who came to the hospital to donate blood. 14,240 such donors were screened from September '96 to August '98.

**Methods:** III Generation ELISA kit from Sorin Biomedica was used to detect HBsAg in all samples.

**Result:** HBsAg was detected in 124 samples out of 14,240 giving the result at 0.87%.

**Conclusion:** Hepatitis B prevalence in Coimbatore, south India is much lower that the national average, the prevalence being 0.87% in 14,240 voluntary blood donors.

**Therapy of chronic hepatitis “B” by combination of nucleoside analogues with interferon. VG Mohan Prasad. KG Hospital, Coimbatore**

**Objective:** Interferon (IFN) monotherapy in chronic hepatitis B leads to 0 to 43% complete response in most trials (Lok et al). This study was undertaken to determine if the response could be improved by combining either ribavirin or lamivudine with interferon, thereby also lowering the 5 mg dose of IFN to 3 mg thrice weekly.

**Study:** 12 males in the age range 28 to 70 years with a mean of 52 years, 2 with CAH and 10 with cirrhosis were enrolled in 4 groups. Patients of all 4 groups received *Phyllanthus neri* 3 grams/day.

**Results:** Group A: 3 patients (1 CAH + 2 cirrhosis); controls only *Phyllanthus neri* 3 grams/day; 0/3 cleared the virus or normalised ALT. Group B: 2 cases (1 CAH + 1 cirrhosis); IFN 3 mg thrice weekly x 4 months; 0/2 cleared the virus or normalised ALT. Group C: 7 cases (2 CAH + 5 cirrhosis); IFN 3 mg thrice weekly with ribavirin 200 mg qid for 4 months, 5/7 became negative for HBV DNA, HBsAg and HBsAg. 1/7 who did not comply with ribavirin dosage converted HBsAg to negativity, normalised ALT, but continued to be HBV DNA positive 1 year posttherapy. 1/7 who had envelope-mutant strain did not respond. Group D: 2 cases (both with envelope mutant HBV cirrhosis) IFN 3 mg thrice weekly with lamivudine 150 mg qid for 4 months; naive patient responded. Patient who failed again.

**Conclusion:** Combining ribavirin with interferon resulted in 100% (6/6) complete response if HBsAg is taken as end-point in wild type virus induced cirrhosis or CAH and 83% response (5/6) if HBV DNA was the end point. There was 50% CR in with envelope-mutant cirrhosis (1/2).

**Acute HBV hepatitis: an effective treatment plan. L Venkatagathy, Ajay Hospital, Centre for Digestive Diseases, Erode, Tamilnadu**

**Aim:** To evaluate the efficacy of a combination of ribavirin, UDCA, Phyllanthus and silymarin in the treatment of acute HBV hepatitis.

**Method:** 22 consecutive patients (13 males, 9 females: age ranging from 3 years to 68 years with median being
Hepatitis B and C: carrier to cancer — Abstracts

Introduction: Carrier state for HBV is a global health problem, the burden being approximately 350 million cases. Many of these subjects are asymptomatic and their natural history is poorly defined.

Aim: 1) To study the biochemical, serological and histological profile of incidentally detected asymptomatic HBsAg positive subjects. 2) To compare the biochemical and histological profile of HBeAg positive chronic carriers with those having anti-HBe positivity.

Materials and Methods: 240 subjects who were asymptomatic and detected incidentally to be HBsAg positive for >6 months were enrolled into the study. Of these 190 patients were negative for markers of HCV, HDV, HIV and had no significant history of alcohol or drug intake. Sixty eight patients consented for liver biopsy which was analysed by two histopathologists blinded to patients' clinical and laboratory data using the Knodell's score. The histological activity index (HAI) of <3 was considered as normal or nonspecific and a score of >3 was considered chronic hepatitis. Subjects' stored sera was analysed for ALT levels, HBeAg (ELISA), anti-HBe (ELISA) and HBV-DNA by PCR. Serum ALT level of >35 was considered as elevated ALT.

Results: Of 68 patients analysed 28 were HBeAg positive and 40 were anti-HBeAg positive. The biochemical and histological profile of subjects with HBeAg and anti-HBe positivity is depicted in the Table below.

<table>
<thead>
<tr>
<th>HAI score</th>
<th>N ALT (n=28)</th>
<th>^ ALT (n=22)</th>
<th>Anti-HBe (+) (n=40)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;3</td>
<td>4</td>
<td>6</td>
<td>8</td>
</tr>
<tr>
<td>&gt;3</td>
<td>2</td>
<td>16</td>
<td>6</td>
</tr>
</tbody>
</table>

In patients with anti-HBe positivity, HBV DNA was analysed in 28 patients. Nine were HBV-DNA positive and 19 were negative. Of the 9 HBV-DNA positive patients normal ALT was seen in 2 and increased ALT in 7. Only 1 of these 9 patients had normal biopsy. Of 19 DNA negative patients, ALT was normal in 7. Only 3/7 had normal histology. Therefore only 3/68 patients (HBsAg negative, normal ALT, HBV-DNA negative and normal histology) may possibly constitute true carriers.

Conclusions: 1) There is significant correlation of ALT levels (p=0.02) but no correlation of HBeAg and HBV-DNA positivity with the histological activity of disease in incidentally detected asymptomatic HBsAg positive subjects. 2) True HBV carrier state probably does not exist, if exists, is seen only in a small fraction of such subjects.

Biochemical and serological profile of incidentally detected asymptomatic HBsAg positive subjects. SR Agarwal, D Kapoor, R Chandra, V Thakur, SK Sarin. Department of Gastroenterology, G B Pant Hospital, New Delhi

Introduction: The HBV carrier state has been defined in literature as HBsAg positivity for period of >6 months.
This however, does not take into account the presence or absence of ongoing hepatic inflammation and infective potential of such subjects.

**Aims:** 1) To study the biochemical and serological profile of incidentally detected asymptomatic subjects positive for HBsAg antigen for >6 months. 2) To study the replicative status of HBV in such subjects.

**Materials & Methods:** 240 consecutive asymptomatic subjects referred to our centre (1996-1998) because of incidental detection of HBsAg were enrolled in this study. The HBsAg positivity for more than 6 months was established in all subjects. Patients were excluded (n=50) if found to be positive for HCV, HDV, HIV, alcohol abuse or significant drug intake. Thirteen patients were excluded because of non-availability of complete serological workup. Stored sera of these subjects were analysed for ALT (Normal = 35 IU/L), HBeAg (ELISA), anti-HBe (ELISA). In patients who were anti-HBe positive, HBV-DNA was tested using KR.

**Results:** In all, 177 subjects were analysed. The biochemical and serological profiles of these subjects are depicted in the Table below.

<table>
<thead>
<tr>
<th>ALT levels</th>
<th>HBsAg positive (n=177)</th>
<th>Anti-HBe positive (n=116)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>12 (20%)</td>
<td>49 (42%)</td>
</tr>
<tr>
<td>Elevated</td>
<td>49 (80%)</td>
<td>67 (56%)</td>
</tr>
<tr>
<td>&gt;2 x UNL</td>
<td>22 (37%)</td>
<td>27 (23%)</td>
</tr>
<tr>
<td>&gt;5 x UNL</td>
<td>7 (11%)</td>
<td>8 (7%)</td>
</tr>
</tbody>
</table>

*p=0.004; UNL - upper limit of normal

Of 116 anti-HBe positive subjects, HBV-DNA was analysed in 64 (55%) subjects. Of these 25 (39%) had normal ALT and 39 (61%) had increased ALT. In the former group 35/25 (100%) had normal ALT. The latter 11/39 (28%) had increased ALT. Conclusions: 1) In India, incidentally detected asymptomatic HBsAg positive subjects are about two times more likely to be anti-HBe positive. 2) Around 67% of such incidentally detected subjects have increased ALT. 3) Even when anti-HBe positive, 25% of these incidentally detected subjects have detectable viral DNA. 4) At least 49/177 (HBeAg +ve, increased ALT) of these incidentally detected subjects would readily qualify for treatment.

**Lamivudine in patients with HBV related decompensated liver disease.** D Kapoor, SR Aggarwal, RC Gupta, V Thakur, SK Sarin. Department of Gastroenterology, G B Pant Hospital, New Delhi.

**Introduction:** HBV related chronic liver disease patients in a decompensated state are not eligible for therapy with interferon. However, some form of antiviral therapy may benefit these patients.

**Aims:** 1) To study the effect of lamivudine on clinical, biological and serological profile of HBV related decompensated chronic liver disease patients. 2) To study the effect of lamivudine on HBV DNA suppression.

**Material and Methods:** 15 patients of HBV related decompensated chronic liver disease were enrolled. The patients were negative for markers of HCV, HDV, HIV and history of alcohol/drug intake. None of the patients received any therapy in the past. A baseline clinical examination was followed thereafter by monthly examination. In addition, the following parameters were tested at the times indicated: ALT, serum albumin, prothrombin time - 4 weeks; HBsAg - 0, 6, 12, 18, 24 mo; HBeAg and anti-HBe - 0, 3, 6, 12, 15, 18, 24 mo; HBV DNA (bDNA) - 0, 4, 8, 12 weeks, thereafter 3 months. In patients negative for HBV DNA by bDNA assay, the serum was tested with PCR. Patients received lamivudine at the dose of 150 mg/day after giving written informed consent. They were screened for adverse effects of drug. They continued to take diuretics and antibiotics as prophylaxis against SBP.

**Results:** 7 patients were HBe + and 8 anti-HBe +. Mean follow up 12.1 months (range 4-26). The patients' pre and post treatment biochemical and clinical parameters are depicted in the Table.

<table>
<thead>
<tr>
<th></th>
<th>ALT (IU/L)</th>
<th>S. albumin (g/dL)</th>
<th>S. bilirubin (g/dL)</th>
<th>Child-Pugh score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre Tt</td>
<td>111.5</td>
<td>3.33</td>
<td>1.73</td>
<td>8.2</td>
</tr>
<tr>
<td>Post Tt</td>
<td>57.6*</td>
<td>3.57*</td>
<td>1.65*</td>
<td>6.7*</td>
</tr>
</tbody>
</table>

*P=0.001. All values expressed as mean

The HBV DNA was suppressed in 9 patients after 4 weeks (by PCR). 5/7 (43%) HBe + patients seroconverted to anti-HBe over 3-12 mo and remained so during the remainder of follow up. In 9 patients control of ascites was made easier. Only 1 patient had worsening of PSE during follow up. None developed any adverse effect to the drug.

**Conclusions:** 1) Lamivudine is extremely effective in suppression of HBV DNA. 2) In about 43% of HBe + patients, seroconversion was achieved. 3) There is a significant improvement in Child-Pugh score of these patients. 4) The therapy has few, if any, side effects.

**Cost of hepatitis B virus related disease in India — a pilot study.** Kishore Chaudhry, RAJIV GUPTA, NS MITRA, RS GOSAI, SK SARIN. ICOR, Dept. of Gastroenterology, G B Pant Hospital, New Delhi.

**Background:** India has about 40 million HBV carrier and 4 million patients with HBV related chronic liver diseases. Economics of HBV disease would help in initiation of control measures. Few studies undertaken elsewhere have not measured the exact expenditure by patients or their relatives.

**Objectives:** The study estimated direct and indirect expenditure, by HBV carriers and patients of chronic liver disease due to HBV, and identified factors resulting in variation of expenditure.

**Methodology:** Sixty six consecutive HBsAg+ patients were interviewed by medical-social worker, to collect data on expenditure related to their disease, under various heads, namely, consultation, investigations, therapy, hospitalization, extra expenditure on food, lodging, transport charges.
Hepatitis B and C: carrier to cancer — Abstracts

loss of income, and any expenditure by relatives/friends. Carrier was defined as a person positive for HBsAg for more than 6 months. Any expenditure on conditions not related to hepatitis was excluded. Data was analyzed to identify demographic and disease related factors, resulting in higher expenses, by univariate as well as multiple regression analysis by software package EPI INFO.

Results: Average expenditure by a carrier of HBV during the last six months was Rupees 4,122 (range 20 to 28,600). Investigations accounted for about 57% of the expenditure. Only history of hospitalization resulted in statistically significant higher expenditure. Patients visited the hospital an average of 3 times. A patient of chronic liver disease spent an average of Rupees 23,561 during last six months (range 60 to 111,395). Major expenditure heads included drugs (56%), investigations, loss of income, transport, extra money on food, etc. Univariate analysis revealed higher expenditure by patients staying outside Delhi, staying far from hospital (for those living outside Delhi), using costlier modes of transport (for Delhi residents), education (no trend visible) and with history of hospitalization. Multivariate regression analysis revealed a significant model with hospitalization, residence outside Delhi, and higher family income, significantly resulting in higher expenditure. These patients visited the hospital an average of 4.8 times.

Conclusions: The methodology has been found to be feasible. The pilot data indicates an enormous expenditure by patients of HBV related diseases.

Comparative evaluation of histological features of chronic hepatitis B and chronic hepatitis C in an Indian population. D Rakeja, V Malhotra, R Gondal, P Saluja, SK Sarin,* M Sidhu. Department of Pathology and *Department of Gastroenterology, G B Pant Hospital, New Delhi

Large number of studies have indicated that steatosis, lymphoid aggregate/follicles and bile duct injury are histological features more consistently associated with chronic hepatitis C than chronic hepatitis B. We have examined liver biopsies of thirty patients of chronic hepatitis B with equal number of age-matched patients with chronic hepatitis C. Steatosis, lymphoid follicles/aggregates and bile duct injury were noted in 66.6%, 36.6% and 25.6% cases respectively of chronic hepatitis B as compared to 70%, 33.3% and 30% cases respectively of chronic hepatitis C. Thus none of the features were considered distinctive of HCV infection.

Higher immunogenic response of HBV vaccine in contacts of HBV related chronic liver disease patients. V Thakur, RC Gupta, N Tandon, MK Parvez, SN Kazim, RK Kaul, SK Sarin. Dept. of Gastroenterology, GB Pant Hospital, New Delhi

Background: Close family contacts of patients infected with hepatitis B virus (HBV) related chronic liver disease, constitute a high risk group which needs mandatory vaccination. Immunogenicity of a vaccine in contacts exposed (IgG anti HBc positive) versus unexposed (IgG anti HBc negative) to HBV infection was studied.

Material and Methods: Seventy nine family contacts, who were HBsAg negative and had persistently normal ALT and no clinical or ultrasonographic evidence of liver disease were vaccinated against HBV without knowing prior HBV exposure status, using a recombinant vaccine. A dose of 10 (pediatric) or 20 mg (adult) was injected at deltoid at 0, 6, and 6 months. The anti HBs (AHI) titres were measured using ELISA at baseline, 1, 2 and 7 mo. Serum IgG anti HBc (ELISA) was studied at baseline and at 7 mo. Subjects were divided into two groups. Gr I: Exposed, Gr II: Unexposed. AHI titres > 1 was defined as seroconversion and titres > 10 IU/L as seroprotection.

Results: Immunogenic response of HBV vaccine in exposed as well as non exposed contacts was comparable. 24 of 25 (96%) exposed and 51 of 54 (94%) non exposed contacts were seroprotected after the completion of the schedule. The seroprotective anti HBs response after the first dose (mo. 1) was significantly higher in exposed contacts. Moreover anti HBs titres were significantly higher in exposed contacts mo 1, 2 and 7 after every shot of vaccine irrespective of the sex. At mo 7 non responders were 1 of 25 and 3 of 54 in gp I and II, respectively (pens). Immunogenic response in contacts of the age above 40 years of age was significantly lower in unexposed contacts after the first dose.

Conclusions: 1. In exposed family contacts seroprotection occurred earlier and was stronger. It resembled an anamnestic reaction. 2. Anti HBs titres were significantly higher in exposed contacts at all times. Lowered immunogenic response was observed in non exposed contacts of the age above 40 years.

Significance of HBV-DNA seropositivity in anti HBc positive healthy carriers in India. MK Parvez, R Kaul, V Thakur, RC Gupta, N Tandon, SK Sarin. Department of Gastroenterology, G B Pant Hospital, New Delhi

Background: Presence of anti HBc generally indicates the cessation of HBV replication. However, HBV-DNA remains positive in a fair proportion in the asymptomatic anti HBc positive carriers. The correlation of HBV-DNA positivity with biochemical and histological status of liver disease in incidentally detected anti HBc positive carriers was studied.

Material and Methods: Sera of twenty six liver biopsy proven HBsAg carriers who were anti HBc positive (ELISA) were taken. The HBV-DNA was extracted by standard phenol-chloroform method and detected by PCR, using primers from the conserved region of HBV-genome. Further, histological activity index (HAI) and biochemical (AST and ALT) status were assessed.
Results: Of the 26 anti HBe positive carriers (age = 31±10 yrs), twelve sera (46%) were found positive while fourteen sera (54%) were found negative for the HBV-DNA (p=n). Serum ALT levels were higher in the DNA-negative group. Since the difference was not significant (Table), both the groups had slightly comparable histology.

Conclusions: 1. Both HBV DNA +ve as well as HBV DNA -ve carriers with anti HBe positivity showed comparable histological and biochemical features. 2. There was no correlation of histological and biochemical status with the presence of HBV-DNA in anti HBsAg carriers.

Table

<table>
<thead>
<tr>
<th>Parameters</th>
<th>HBV-DNA +ve (n=12)</th>
<th>HBV-DNA -ve (n=14)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HAI</td>
<td>4.34±1.8</td>
<td>4.12±2.63</td>
</tr>
<tr>
<td>AST (IU/L)</td>
<td>36.66±12.0</td>
<td>22.8±6.0</td>
</tr>
<tr>
<td>ALT (IU/L)</td>
<td>38.8±3.37</td>
<td>46.7±36.87</td>
</tr>
</tbody>
</table>

Novel mutations in the hepatitis B virus surface gene.
SN: Karij. Salma M Wakis, V Thakur, SE Nassen, SK Sarin. Department of Gastroenterology, G B Pant Hospital and Eukaryotic Genes Expression Laboratory, National Institute of Immunology, New Delhi

Objectives: HBV was considered to be genetically stable, but it is now clear that mutations occur frequently, probably because reverse transcription, essential in the replication cycle of HBV is error prone. The HBsAg mutant described most frequently, has an amino acid substitution at position 145. Other mutants have been identified with amino acid substitutions in the immunodominant 'a' determinant of HBsAg.

Materials and Methods: Sera from six biopsy proven chronic hepatitis patients, positive for HBsAg and HBV DNA by the diagnostic PCR, were collected for this study. HBsAg, HBeAg and anti HBe were done using commercial ELISA. Extracted DNA from sera samples were subjected to the PCR to amplify the fragment of the surface gene which includes the 'a' determinant region. Amplified PCR product was purified with Qiagen PCR purification kit. Subsequently it was sequenced and the complete 'a' determinant region along with its upstream and downstream sequence was read.

Results: In five patients, there was a complete wild type sequence for the whole 'a' determinant region. In one patient only, a novel point mutation was observed at aa 128 (A to V). In the same patient, mutations at aa 115 and aa 118 were also observed. At aa 118 threonine was replaced with valine and at aa 115 there was a silent mutation (T to T).

Conclusion: Alanine at position 128, which comes within the first loop of 'a' determinant region was substituted with valine may be having some clinical importance and needs to be studied. Although aa 115 and aa 118 do not come exactly in the range of 'a' determinant region, but their nearby positions could be of immense clinical significance.

Hepatitis C

Epidemiology of hepatitis C in the local and immigrant population of Saudi Arabia. S Risw Melodi. Sr Lecturer, Dept. of Pathology, J N Medical College, AMU, Aligarh

The hepatitis C virus (HCV) is an RNA virus, related to flaviviruses, and the cause of most diseases previously described as parenterally transmitted non A non B hepatitis. Of those most at risk are recipients of multiple blood transfusions. This study was conducted on healthy blood donors in KFSH Hospital, Buraidah, Saudi Arabia, from Feb 95 to Feb 97. In all, 11007 donors were screened for hepatitis C (anti HCV) beside HBsAg and other parameters. 403 (3.68%) were found positive for anti HCV which is a little higher to hepatitis B cases which comprised only 375 (3.4%). The highest incidence (8.4%) was seen in the fifth decade of life. The Egyptians topped the list, showing almost 8 times higher positivity for hepatitis C compared to hepatitis B. Among the local Saudis the trend was reverse, where anti HCV positivity was lower to HBsAg. The immigrants from the Indian subcontinent, India, Pakistan and Bangladesh were almost equally prone to hepatitis B and C, while the Philipinos, Syrians and Turkish had great affinity for hepatitis B and minuscule minority was positive for hepatitis C.

Prevalence of hepatitis C virus in acute hepatitis in South India. Mohan Prasad, Ruby Thomas, Devraj. Viral Hepatitis Research Laboratory, KG Hospital, Coimbatore

Objectives: This study was undertaken to determine the occurrence of hepatitis C among all cases of acute hepatitis in South India.

Definition: Acute hepatitis was defined as an acute onset of jaundice of less than 6 weeks in duration.

Material: 686 consecutive samples over a 2 year period from September '96 to September '98 were studied prospectively for the prevalence of hepatitis A to E viruses. Cases with history of alcoholism, drug-intake were excluded, using a detailed questionnaire. Ultrasonography was done to rule out obstructive causes of jaundice.

Methods: III generation ELISA kits of Sorin Biomedica were used to analyse anti-hepatitis C virus antibodies in all samples.

Results: Hepatitis C virus antibodies were detected alone in 18 samples (2.62%) and in combination with other viruses in 3 samples (0.43%).

Conclusion: Hepatitis C is the cause of acute hepatitis in 3% of 686 consecutive cases analysed for hepatitis A to E viruses in South India.

Therapy of chronic hepatitis C: combination of ribavirin and UDCA with Interferon. VG Mohan Prasad. Hepatology Unit, KG Hospital, Coimbatore

Objectives: Sustained response with interferon monotherapy in chronic hepatitis C is less than 25% (Davis et al). This
study was undertaken to find out if the efficacy of interferon could be improved by combining ribavirin and UDCA.

Study: 19 patients - 14 males and 5 females in the age range of 43 years to 57 years, 4 of them with CAH and 15 with cirrhosis were enrolled.

Methods: All patients had anti HCV antibodies by ELISA 3 of Sorin Biomedica and HCV RNA (qualitative).

Results: Group-A: 5 pts (1 CAH + 4 cirrhosis): Ifn 3 miu tiw x 6 months; 1 pt with CAH became HCV RNA negative. Group-B: 7 pts (1 CAH + 6 cirrhosis): Ifn 3 miu tiw with ribavirin 200 mg qid x 6 months; 1 pt with cirrhosis became HCV RNA negative. Group-C: 7 pts (2 CAH + 5 cirrhosis): Ifn 3 miu tiw + ribavirin 200 mg qid + UDCA 300 mg bid x 6 months; 2 pts with CAH and 2 pts with cirrhosis became HCV RNA negative.

Conclusion: Monotherapy with interferon and combination with ribavirin produced less than 20% sustained response in cirrhotics. Combining UDCA improved the rate of response to 44%. Induction dosing with 5 miu of interferon with 2 antivirals probably would be the line of choice in cirrhosis due to hepatitis C virus.

Prevalence of hepatitis C virus: a report from Gujarat Cancer and Research Institute, Porajit N Goswami, Hitendra A Patel, Jayshree S Patel. Department of Microbiology, GCI, Ahmedabad 380 016

Hepatitis C virus (HCV) is one of the major causative agents of parenterally transmitted hepatitis, next to HBV. Enzyme immuno assays have been developed for detecting antibodies to HCV. We used third generation EIA kit. The prevalence of HCV antibodies was studied in blood donors, patients suffering with liver diseases and post transfusion cancer patients.

Out of total 14,325 blood donors sample, 45 (0.31%) were positive for anti-HCV. Total 262 patient samples have been screened for anti-HCV and 11 were positive, of which 1/22 (4.5%) with acute viral hepatitis, 3/23 (13%) with liver malignancies and 7/222 (3.1%) have other than liver malignancies. The significance will be discussed.

TGF-β levels in HCV related chronic liver disease as studied by semiquantitative RT-PCR. S Ray, S Broor, D Ghosh, Y Vaishnav, SL Broor, L Dar, P Seth. Department of Microbiology, AIIMS; Virology Section, ICGEB; Department of Gastroenterology, GB Pant Hospital, New Delhi

TGF-β family of cytokines is ubiquitous and multifunctional and play an important role in growth and development, inflammation and repair and host immunity. The mammalian TGF-β isoforms (TGF-β1, β2, β3) are secreted as latent precursors and have multiple cell surface receptors of which at least two - β1 and β2 - mediate signal transduction. It has been reported that in cases of chronic liver disease TGF-β levels have been found to be upregulated.

In this study an attempt has been made to show the status of TGF-β in HCV related chronic liver disease. RT-PCR for TGF-β mRNA was done on liver biopsies. In 3 biopsy samples from control group TGF-β mRNA could not be detected. TGF-β mRNA was present in biopsies from 4 patients of chronic liver disease due to HCV. In 1 patient of fulminating hepatic failure also TGF-β mRNA was detected in biopsy. The levels of TGF-β have been quantitated by semi-quantitative PCR using TGF-β mimics.

An increase in TGF-β levels in HCV infection thus may have a role in pathogenesis of HCV related cirrhosis and hepatocellular carcinoma as is evident from clinical sample data.

In-vitro studies of the levels of TGF-beta mRNA in HepG2 cells transfected with sub-genomic fragment of HCV. S Ray, S Broor, D Ghosh, Y Vaishnav, SL Broor, L Dar, P Seth. AIIMS; Virology Section, ICGEB, New Delhi

TGF-β is a multifunctional group of cytokines that play an important role in cellular growth, development, differentiation and proliferation. TGF-β also mediates fibrogenesis, chondrogenesis and osteoporosis by inducing synthesis of extracellular proteins. It is synthesised by a number of cells and though not detected in the liver under normal conditions it is present in HepG2 (human hepatoblastoma) cells.

Studies have shown an upregulation in the levels of TGF-β mRNA in liver biopsies of patients with chronic liver disease. We have made an attempt to study the status of TGF-β in vitro using HepG2 cells transfected with a sub-genomic fragment of HCV consisting of the core, envelope (E1 and E2), and non structural regions NS2 and part of NS3 incorporated in a mammalian expression system. Post transfection processing of the cells by RT-PCR showed the presence of HCV and TGF-β mRNA. Quantitation of the levels of TGF-β by semiquantitative PCR using TGF-β mimics showed a ten fold increase in the levels of TGF-β in transfected cells compared to mock transfected or untransfected cells. This shows that any of the viral proteins incorporated in the HepG2 cells must be playing an important role in the upregulation of TGF-β which might have some correlation with the pathogenicity of HCV.

Incidence of anti-HCV positivity in extrahepatic syndromes. Lt Col M KK Rao, Lt Col SK Thakur, Lt Gen D Raghunath. Armed Forces Medical College, Pune & Army Hospital (R&R), Delhi

HCV infection has been reported to lead to many immunological abnormalities causing extrahepatic manifestations in the infected subjects. We studied anti-HCV positivity in few selected syndromes using RIBA-3 assay.
A total of 47 extrahepatic syndromes were studied between Oct 1994 to Mar 1995. These included polyarteritis nodosa (n=14), membranous glomerulopathy (n=31) and suspected porphyria cutanea tarda (n=2). The anti-HCV positivity in these cases was found in 04, 07, and 01 cases respectively. Out of these 12 cases testing positive for anti-HCV, there was cryoglobulinemia in 04 cases. This finding instigated us to look for cryoglobulinemia in the 67 anti-HCV positive biopsy proved CAH cases and 19 (28.35%) of these CAH cases tested positive for cryoglobulins. Surprisingly, the cryoglobulins were lost in 17 out of the 19 cases following interferon therapy. It is concluded that extrahepatic syndromes such as those highlighted above must be investigated for anti-HCV in addition to looking for markers of HBV.

Prevalence of chronic hepatitis C in multitransfused patients in eastern India. Ajay Kumar Sinha, Assistant Director, Immunology and Vaccine Development, National Institute of Cholera & Enteric Diseases, Calcutta 10; Subhas Chandra Hazra, Reader in Medicine, and Utpal Chaudhuri, Reader in Haematology, Department of Medicine, Medical College, Calcutta

The current incidence of chronic hepatitis C (HCV) infection with or without cirrhosis was recorded in Calcutta, Eastern India. This region appears to be endemic, particularly in multitransfused hematopoietic disorders. Presence of HCV infection was investigated on 230 multitransfused cases by using ELA and confirmed by HCV RNA testing. Antibodies to HCV alone were found to be present in 57 (24.8%) cases and dual infection with HBV and HCV was noted in 17 (7.4%) patients with cirrhosis of the liver. The results indicate the higher prevalence of chronic liver disease due to hepatitis C among multitransfused patients in eastern India and outcome of this dual infection seems to be more harmful.

Evaluation of risk factors in HCV infected chronic hepatitis patients. SJ Khundmiri, DN Reddy, MN Khaja, N Farooq, MM Husain, CM Habibullah. Center For Liver Diseases, Owaasi Hospital and Research Centre, Hyderabad, & Asian Institute of Gastroenterology, Medinova Hospital, Hyderabad

HCV is the major cause of 90% of PTH cases. However, after the development of diagnostic tests for HCV, the risk of infection has declined considerably. In India the estimated HCV prevalence in voluntary blood donors ranges between 1.85% (Delhi) to 3% (Hyderabad). HCV is transmitted primarily by parenteral route and sources of infection include injection drug use, needle stick accidents and transfusion of blood and blood products. However, transmission by sexual routes is also common. In the present study we have evaluated the risk factors associated with HCV infection. Among the 184 patients reported to our Center for HCV RNA analysis 104 were positive for both anti HCV and HCV RNA. 57 patients were positive only for HCV RNA while 3 patients were positive for anti HCV only. 20 patients were negative for both anti HCV and HCV RNA. Among the risk factors evaluated renal transplant and/or hemodialysis and surgery 6 months prior to infection were the major routes of HCV transmission. Blood transfusion and sexual transmission followed hemodialysis and surgery as the risk factors. 23 patients did not have any risk factors evaluated. The results will be discussed.

The work was partially supported by Fulford (I) Ltd.

Prevalence of anti-HCV antibodies in various high risk surrgroups in India. Shalini Sharma, SKThakur, MKK Rao, M Jaiprakash. Army Hospital R R, Delhi Cantt-10 and Armed Forces Medical College, Pune

The prevalence of antibodies to hepatitis C virus (HCV) was investigated among voluntary blood donors and various high risk subgroups. The commercially available RIBA-3 assay kit was used. Antibodies to HCV were detected in 1.54% of the 16,000 healthy voluntary blood donors screened.

Among these with HIV seropositivity (N=234) the prevalence was 27.7%, acute viral hepatitis (N=736) 18.34%, chronic liver diseases (N=65) 36.9%, HBsAg carriers (N=260) 15%, cardiac patients (N=64) 4.76%, tuberculosis (N=308) 12.33% and those with sexually transmitted diseases (N=251) the prevalence was 26.29%. Prevalence of HBV markers in all these subgroups was higher as compared to anti-HCV prevalence. These results suggest that HCV infection is not a major problem in the healthy general population. It assumes significance in the immunocompromised and high risk for parenteral exposure.

Sero-prevalence of hepatitis C virus antibodies in thalassaemia population. KJ Prasad, C Wattal, A Sharma. Department of Clinical Microbiology, Sir Ganga Ram Hospital, New Delhi

Objectives: To find out the exact prevalence of HCV antibodies among different groups of blood recipients including thalassaemias and blood donors.

Methods: The data pertaining to detection of HCV antibodies by ELISA (third generation) during the years 1997 and 1998 at our hospital was analysed.

Results: Out of 2060 subjects screened for anti HCV antibodies, 201 were detected positive. The overall prevalence of seropositive cases was 9.75%. Among different groups of patients i.e., the thalassaemia patients, dialysis patients, liver diseases patients and blood donors, the seropositivity for HCV antibodies was found to be 30.47, 8,19, 10.53 and 1.36 percent, respectively. The highest seropositivity was among the thalassemia patients followed by liver disease cases. A declining trend in seropositivity was observed in the year 1998 as compared to
Hepatitis C virus infection in chronic liver diseases in India. S Broor,1 S Ray,1 SL Broor,2 S Rawat,2 D Kapoor,2 L Dar,1 P Seth.1 All India Institute of Medical Sciences1 and GB Pant Hospital,2 New Delhi

Hepatitis C virus (HCV) is associated with chronic liver disease. Different HCV genotypes cause varying severity of hepatitis and response to interferon therapy. Information on the role of HCV in chronic liver diseases and the prevalence of different genotypes is not available from India. This study was conducted on HCV infection in chronic liver disease patients in India by serology and detection of viral RNA by RT-PCR. One hundred and eighty-two patients of chronic liver disease attending GB Pant Hospital, were included in the study. Antibodies to HCV were detected by 4th generation ELISA (UBI 4.0 EIA). HCV RNA was extracted from plasma samples and RT-PCR was carried out with universal primers from the core region. For genotyping a nested PCR was done with a mixture of 4 type specific primers. Of the 182 samples tested for antibodies to HCV by ELISA, 23 (12.64%) were found to be positive. HCV RNA was detected in 27 samples by first PCR. There were 11 samples, which were ELISA positive but PCR negative, and there were 15 which were ELISA negative but PCR positive. Nested PCR was positive in all the 27 PCR positive samples and in additional 11 samples which were ELISA positive but first PCR negative. All the samples by nested PCR were genotyped as HCV type II (Okamoto et al, 1992). Thus in India HCV infection was detected in 38 (20.8%) out of 182 patients of chronic liver disease and all the strains were of HCV genotype II.

Hepatitis C virus (HCV) infection in children in North India. NK Arora, Prashant Mathur, Nitiika Arora, SK Panda.* Division of Paediatric Gastroenterology, Hepatology & Nutrition, Department of Paediatrics, *Department Of Pathology, All India Institute Of Medical Sciences, New Delhi

Background: Risk factors and prevalence of HCV infection in children are not clearly defined.

Objective: To determine the prevalence of anti-HCV antibodies in children with and without liver disease attending a tertiary care hospital in North India.

Setting: Tertiary care hospital and rural health dispensary.

Subjects: (i) 2176 children (6 months-10 years) attending paediatric outpatient of an urban tertiary hospital (n=1113) and a rural health dispensary (n=1063) for minor non-hepatic illness; (ii) 582 children with acute sporadic hepatitis and (iii) 390 children with chronic liver disease.

Methods: Enzyme immunosassay (EIA) developed in-house for detection of antibody against the prevailing HCV genotypes in India was used. Specific reactivity of the test was compared with commercial 2nd and 3rd generation EIA and RT-nested PCR.

Results:

<table>
<thead>
<tr>
<th>Study Group</th>
<th>N</th>
<th>Anti-HCV positive</th>
<th>H/o transfusion/injection in previous one year</th>
</tr>
</thead>
<tbody>
<tr>
<td>Children with no liver disease</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>urban 1113</td>
<td>58</td>
<td>5.2</td>
<td>2/55 (3.4%)</td>
</tr>
<tr>
<td>rural 1063</td>
<td>44</td>
<td>4.1</td>
<td>2/44 (4.5%)</td>
</tr>
<tr>
<td>Acute sporadic hepatitis</td>
<td>582</td>
<td>19</td>
<td>12/19 (63.1%)</td>
</tr>
<tr>
<td>Chronic liver disease</td>
<td>390</td>
<td>13</td>
<td>9/13 (69.2%)</td>
</tr>
</tbody>
</table>

Conclusions: Prevalence of anti-HCV antibodies in children with and without liver disease was similar although history of transfusion was higher in those with liver disease. This difference could be due to interviewer bias. The significance of anti-HCV antibody positivity and risk factors of HCV transmission in our paediatric population needs further work.

Hepatitis C in chronic renal failure and renal transplant recipients. Pooja Anand, SK Thakur, M Jaiprakash, UK Sharma, A Rajvanshi, HS Gill, AS Narula. Departments of Nephrology and Gastroenterology, Army Hospital RR, Delhi Cantt-10

The occurrence of hepatitis C virus (HCV) infection amongst chronic renal failure (CRF) patients and renal transplant recipients was investigated over a period of 4 years. A total of 252 patients were studied comprising 77 chronic hemodialysis (CHD) patients, 55 CRF patients not on dialysis and 140 renal transplant recipients. Patients were screened before and after a mean 10 session of hemodialysis and their baseline and post dialysis values of liver enzymes were determined. 12 (15.58%) of the 77 patients on CHD were HCV antibody positive. Similarly, among renal transplant recipients 12/140 (8.5%) were positive for HCV infection. The relative risk for hepatitis C was about 8 times greater for those with CRF compared with the normal controls which makes CRF an important risk factor. This risk further increases two-fold in those on CHD. The risk for HCV infection among immunocompromised renal transplant recipients is similar to that of CRF patients in general.
Hepatocellular carcinoma

Childhood hepatocellular carcinoma in India: a report of three cases. S Saigal, HP Nandeesh, V Malhotra, SK Sarin. Deptt of Gastroenterology, G B Pant Hospital, New Delhi

Hepatocellular carcinoma (HCC), a well documented malignancy in adults, is a relatively rare tumor in childhood. As in adults the oncogenic role of hepatitis B virus (HBV) seems very important in childhood HCC. We report three documented cases of childhood HCC seen during the years 1996-97. The clinical and virological profile of three cases is presented:

<table>
<thead>
<tr>
<th>Age</th>
<th>Presenting features</th>
<th>AFP</th>
<th>HBsAg</th>
<th>HBeAg</th>
<th>HBV DNA</th>
<th>Time of death</th>
</tr>
</thead>
<tbody>
<tr>
<td>1M</td>
<td>Abdominal pain, 8,75,000 + + NA 2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>9M</td>
<td>Abdominal pain 13,00,000 + + 6</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>14M</td>
<td>Abdominal pain 3,00,000 + + Lost to follow-up</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NA</td>
<td>not available</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

All the patients tested negative for anti HCV. Vertical transmission of HBV was documented in one patient. The tumors, being large and unrespectable in all three cases, had a very poor prognosis. In conclusion, HCC is not uncommon in children in India. As in adults, the commonest etiological agent is chronic HBV infection. Universal vaccination against HBV is likely to reduce the incidence of chronic liver disease and HCC in children in India and is strongly recommended.

Incidence of HBV and HCV Infection in Hepatocellular Carcinoma. MM Hussain, MN Khajji, N Parees, SJ Khundmiri, CM Habibullah. Centre For Liver Diseases, Owaisi Hospital and Research Center, Hyderabad

Chronic liver diseases are common in India. Hepatocellular carcinoma, an irreversible stage of CLD, is known to have several organic and environmental etiological factors. In the present study we have evaluated the occurrence and etiology of HCC. HCC was diagnosed ultrasonographically, by serum AFP and/or FNAC. Among the 78 patients evaluated, the incidence of HCC was predominant in 40-60 yrs age group. Males were found to be more affected than females. The main cause of HCC was observed to be due to chronic alcoholism. In the age group of 31-40 yrs alcoholism associated with viral infection was the predominant factor for augmenting HCC. However in the age group of 41-60 yrs incidence of HBV infection alone without alcoholism was also frequent. On the contrary HCC incidence due to HCV infection was found to be very low. The study suggests that chronic alcoholism in HBV or HCV infection may increase the progression of disease.

Clinico-epidemiological study on hepatocellular carcinoma with emphasis on role of hepatitis B and C. S Seema Sethi, Denise Frias, Binod Sinha. Department of Pathology, Lady Hardinge Medical College & S.K. Hospital, New Delhi and *Northwestern University Medical School & Hospital (NMH), Chicago, USA

Hepatitis B and C have been implicated in the causation of liver cancer. However more studies need to be done to evaluate their exact role in the pathogenetic evolution of liver cancer. A 5 year retrospective analysis of all liver cancers diagnosed at NMH was done. All available clinical and epidemiological data was analyzed and patients were followed up to the time of this study. A total of 239 cases were studied. Of these 186 cases were metastatic, 41 were hepatocellular carcinomas. 12 were other liver cancers. A history of hepatitis B was found in 7 cases and hepatitis C in 5 cases of hepatocellular carcinoma. Role of hepatitis B and C with the clinico-epidemiological data will be discussed.

Hepatocellular carcinoma at PGI Chandigarh. S Kapoor, SK Gupta, Sehgal, RK Dhiman, Y Chawla, Jyotsna. Departments of Pathology, Immunopathology and Hepatology, Postgraduate Institute of Medical Education & Research, Chandigarh

Introduction: Association of hepatocellular carcinoma (HCC) with hepatitis B and C virus is well recognised. The exact Indian trends regarding HCC are not known due to paucity of literature on this subject.

Aim: To study the morphologic spectrum of HCC and its correlation with alpha fetoprotein, hepatitis B and C virus markers.

Materials and Methods: Twenty (17 M:3 F) patients with HCC diagnosed by fine needle aspiration biopsy were included in the study. The serum of the patients was collected and analysed for alpha fetoprotein, hepatitis B surface antigen (HBsAg) and antibodies to hepatitis C virus (anti-HCV) by third generation ELISA.

Results: The most commonly complained at presentation was pain in abdomen (69.2%) followed by abdominal distension, jaundice and anorexia (38.5% each). Six patients gave a history of chronic alcoholic consumption. Investigations revealed a raised serum bilirubin in 41.7%, a raised alkaline phosphatase in 66.6% and an altered albumin globulin ratio in 90.9% of these patients. Imaging studies showed cirrhosis in 71.4%. There were multiple space occupying lesions in 46.6% and a solitary tumour mass in 46.6%. Splenomegaly was present in 57%, ascites in 40% and portal vein thrombosis in 45.7% of patients. Upper gastrointestinal endoscopy revealed varices in 62.5% of patients. HBsAg was positive in 70% and anti-HCV in 10%; dual infection was detected in one (5%) patient. Alpha fetoprotein levels were raised in 65%, the highest level recorded being 380 ng/ml (normal 0-15 ng/ml) which
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is forty times above normal limit. Most of the patients presented in preterminal stage. One patient underwent a hepatic lobectomy with uneventful postoperative period. Conclusion: Hepatitis B virus infection was a common accompaniment of HCC in our patients.

Correlation of hepatitis B and C viral markers with aflatoxin and AFP in HCC cases from Tamilnadu. KG Murugavel,* KVK Mohan,* NP Pramod,* P Rajasambandam,** Sunil Mathew,*** V Jayanthi,*** R Surendran, K Raghuram, RR Pranaliswamy, S Rajinikanth, and SP Thyagarajan.* **Department of Microbiology, Dr ALM Pitham, Madras 600 113; ***Department of Gastroenterology, Govt. General Hospital, Chennai 600 003; ***Department of Digestive Health and Diseases, Anna Nagar Peripheral Hospital, Chennai 600 040; Departments of Surgical Gastroenterology and Gastroenterology, Stanley Medical College, Chennai 600 010

Objectives: To study the prevalence of HBV and HCV among HCC cases in Tamilnadu and to correlate the AFP positivity with the viral and aflatoxin positivity.

Design: 170 cases of histopathologically-proven HCC cases were collected from the different Gastroenterology clinics of Chennai Govt. Hospitals.

Methods: Testing for HBV, HDV and AFP was performed using the commercially available ELISAs. Anti-HCV screening was done using the RIBA 3.0 (Chiron Corp., USA). HBV-DNA and HCV-HNA were detected using the PCR technique. Aflatoxin in serum and biopsies was detected by the in-house ELISA and immunoperoxidase technique.

Results: Of the 170 HCC cases, 100 (58.83%) were found to be positive for HBsAg and of these 25.87% of the cases were positive for HDV. 25 (14.7%) cases were found to be positive for anti-HCV. All the HCC cases were negative for serum aflatoxin. But, out of the 31 HCC biopsies analysed for aflatoxin, 18 (58%) were positive by the immunoperoxidase method.

Conclusions: The study suggests that HBV is the leading aetiological agent in HCC followed by HCV. The present study also reveals that biopsy samples may be useful for the detection of aflatoxin as the half-life of aflatoxin in serum is very low. The results of AFP levels in viral positive and negative groups will be presented and discussed.

Anticarcinogenic activity of d-limonene in N-nitrosodimethylamine-mediated hepatocarcinogenesis. RR Giri, BR Das. Molecular Biology Unit, Institute of Life Sciences, Bhubaneswar

The epidemiological studies have demonstrated that tobacco and dietary factors comprise approximately 60% of total cancer deaths. In both these cases N-nitroso compounds play an important role in the development of various kinds of cancer including liver cancer. N-nitrosodimethylamine (NDEA) is one such potent hepatic carcinogen. Our earlier experiments have demonstrated that NDEA can induce liver and lung tumor. Such carcinogenesis was found to be associated with elevation of two important nuclear oncogenes like c-jun and c-myc (Cancer Letters, 109, 121-127, 1996 and Cancer Letters, 112, 57-63, 1997). The present report demonstrates the cancer chemopreventive effect of d-limonene (a monoterpene found mainly in orange peel oil) in hepatocarcinogenesis induced by NDEA alone or along with phenobarbitone (NDEA-PB).

Hepatocarcinogenesis was induced by treating 35 days old AKR male mice continuously with 100 ppm of NDEA in drinking water or promoted by 500 ppm of phenobarbitone in 100 ppm of NDEA containing drinking water after 15 days of NDEA treatment alone. The tumor development as well as prevention was studied at morphological, histological and at gene expression level at various periods of treatment. The gene expression of c-jun and c-myc oncogene was studied by northern hybridization and immunohistochemistry. The results indicate that a clear progression of liver tumor is associated with overexpression of c-jun and c-myc oncogene at mRNA and protein level and with increase in duration of both the kinds of treatment. Interestingly such development of liver tumor and overexpression of protein as well as mRNA of c-jun and c-myc oncogene has been completely inhibited when supplemented with d-limonene. Thus we suggest that the inhibition of liver tumor development by d-limonene is mainly through blocking the expression of two important nuclear oncogenes like c-jun and c-myc. From the internet search it has also been found that no such work on the inhibition of c-jun and c-myc expression has been proposed by any other laboratory.

Thus the present report suggests strongly that d-limonene can be used as a potent cancer chemopreventive/therapeutic agent against liver carcinogenesis and tumors associated with overexpression of c-jun and c-myc.

Miscellaneous

Assessment of universal precautions among nursing staff of AIIMS in 1997 — a hospital-based study. SK Agarwal, MP Mohan, M Varghese. Deptt. of Nephrology and Nursing Services, AIIMS, New Delhi

Study was conducted to find out the awareness among the nursing staff about universal precautions (UP). An eleven-questions questionnaire was circulated to random 250 nurses working in various wards, including high risk wards also. They were asked not to sign the paper to hide the identity. 218 (87.2%) responded. Study shows that in AIIMS, 100% of nurses know that they are at a risk of blood-borne infection and majority of them have heard of UP; only 70% of them follow this in their day-to-day activity. More importantly, nearly half of them say that the hospital and
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the department where they are working has not done enough to make them aware about UP. Two-thirds agree that the hospital does not provide adequate facilities to follow these precautions. Almost all wish that the hospital should do more regarding this aspect of health care. While only 13.3% got adequate hepatitis vaccination, one-third have got some vaccination till the time study was done. Study concludes that universal precautions are not practised rigidly in our hospital due to various reasons. It may be an important factor in nosocomial spread of blood-borne infections.

Internet and surveillance. Sudhir Kumar Baner, Padam Singh. IRMS, ICMR, New Delhi

Each epidemic takes very large number of lives leaving behind morbid and disabled cases with huge economic loss. A timely action can prevent/reduce this avoidable catastrophe. For every timely action, there is a need of fast, efficient and cost-effective media.

In the present era, Internet is an emerging technology of communication. Information from one computer to another anywhere in the world can be communicated very fast, i.e., in a very short period of time. It can also fill the gap of disparity of knowledge between urban (metropolitan cities) and rural areas. It is estimated that by the year 2000 AD there will be around 400 million users all around the world. This technology was adopted by India in 1995 and in next ten years will reach into all villages of the country because of liberalised policy of India recently.

In the present paper the role of Internet in surveillance with reference to hepatitis in India in the near future will be presented.

Prevalence of HBV and HCV carrier state in healthy blood donors. Ali Elaissi, UP Dive, GD Coyaji. Jehangir Medical Research Institute, Pune

HBV infection is a global problem as it is endemic in many countries world-wide. It has been estimated that globally there are over 350 million carriers of HBV who are generally healthy and asymptomatic. The relationship between HBV and PTH has long been confirmed because of the presence of these carriers.

HCV infection is usually transmitted parenterally. It also accounts for up to 90% of PTH cases which are not caused by HBV. In almost 50% of patients, the mode of transmission of this virus is not known, and it has propensity to lead to chronic disease as well as the carrier state. Coinfection with HBV and HCV is also known.

During this study, samples were collected from the Blood Bank, Jehangir Hospital and Medical Centre, Pune, for the period 1995-1997. HBV carrier rate was found to be 1.05% amongst the 7,744 donor samples tested. HCV carrier rate was found to be 0.22% on the basis of anti HCV positivity amongst the 3,531 samples tested. Elevated ALT value (> 40 IU/L) and HCV-RNA were found to be present in only 25% of the repeatedly anti-HCV positive samples. 8 of the 82 repeatedly HBsAg positive samples (9.75%) were found to be HBeAg positive and had ALT values elevated over 40 IU/L. Of the 80 repeatedly HBsAg positive samples tested for anti-HBe and anti-HBc (total), 71.25% (57) and 57.5% (46) were found to be positive respectively. 2.43% (2) samples were found to be positive for IgM anti-HBc.

Coinfection of HBV and HCV in chronic hepatitis: evaluation of risk factors. CM Habibullah, SJ Khundmiri, MN Khaja, MM Hussain, N Parves, DN Reddy. Center For Liver Diseases, Owaisi Hospital and Research Center, Hyderabad and *Asian Institute of Gastroenterology, Medwina Hospital, Hyderabad

HBV and HCV coinfection has been reported to result in chronic carrier state and changes the progression of the disease causing high morbidity and mortality. HBV and HCV co-infection has been reported to be common especially in organ transplant patients. In the present study we have evaluated 33 patients with HBV and HCV coinfection. Our results show that 10 patients were positive for HBsAg, HBeAg, HBV DNA, anti-HCV and HCV RNA while 3 patients were positive for all markers except HBeAg. Two patients were positive for all the above markers except surface antigen, one patient was positive for HBV DNA and HCV RNA only. When the risk factors were evaluated, we found renal transplant and/or hemodialysis as the main cause of HBV and HCV coinfection. Three patients were found to have blood transfusion and three had multiple sexual contacts as the risk factors. The results will be discussed.

Association of hepatitis B and hepatitis C virus infection in the spectrum of chronic liver disease in northern India. P Kar, Paras Chandra, Gopal Krishna, BC Das. Dept. of Medicine, Maulana Azad Medical College and Division of Molecular Oncology, Institute of Cytology and Preventive Oncology (ICMR), New Delhi

Objective: To assess the role of hepatitis C virus (HCV) and hepatitis B virus (HBV) in the causation of chronic liver disease.

Setting: A large north Indian referral hospital.

Methods: Ninety four cases of chronic liver disease comprising of 62 cases of cirrhosis, 22 cases of chronic hepatitis (CH) and 10 cases of hepatocellular carcinoma (HCC) were evaluated for presence of viral markers for chronic liver disease including HBsAg (Eliasia Micro ELISA, Ranbaxy, England), IgM anti HBC (MHIBCHIA, Clonatec, France), IgM anti HCV (Innotest HCV AbII, Belgium). PCR for detection of HCV-RNA was done in all cases using primers from 5' non coding region.

Results: Out of 62 cases of cirrhosis, markers for HCV
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Medicine, Maulana Azad Medical College and *Blood Bank, LN Hospital, New Delhi 110 002

Background: Hepatitis B and hepatitis C are important hepatotropic agents that cause a significant proportion of acute and chronic liver diseases. Although the prevalence of these transfusion-transmissible agents in voluntary blood donors has been evaluated in the past, there is a paucity of large, extensively conducted studies.

Objectives: The present study was designed to evaluate the prevalence of markers of HBV exposure (HBsAg, IgG anti-HBc, HBeAg and anti-HBs) and HCV infection (anti-HCV) in the healthy voluntary blood donors of New Delhi.

Population and Methods: A total of 35,200 voluntary blood donors without any history or symptomatology suggestive of hepatobiliary disease or high risk behaviour were evaluated for the presence of HBsAg by a commercially available third generation ELISA kit. About 3000 random samples were tested for anti-HCV and 500 random samples were screened for other markers of HBV exposure (IgG anti-HBc, HBeAg and anti-HBs).

Results: The mean age of all the blood donors was 28.1 years and male:female ratio was 11.5:1. HBsAg was detected in 1387/35,200 (3.94%) donors and anti-HCV in 53/3000 (1.77%). The prevalence of other markers of HBV exposure was as follows: IgG anti-HBc (103/500; 20.6%), HBeAg (10/500; 2%) and anti-HBs (109/500; 21.8%). In 5 (1%) subjects, HBeAg was detectable despite the absence of HBsAg. Polymerase chain reaction for hepatitis B virus sequences in these samples could detect HBV DNA, suggesting that these subjects probably had surface mutant forms of the virus.

Conclusions: The prevalence of HBsAg amongst the voluntary blood donors of New Delhi is 3.94% and this is consistent with intermediate endemicity range (2-7%) reported for India. Other HBV related markers, especially IgG anti-HBc and anti-HBs, were present in a much higher number of donors, suggesting a very high HBV exposure rate. The prevalence of anti-HCV is 1.77% and this is comparable to that seen in the West.

Hepatitis B and C virus scenario in central India. SP Jaiswal, DS Chitnis, G Naik, A Jain, P Sauligia, A Sepaha. Dept. of Microbiology and Immunology, Gastroenterology and Nephrology, Chouhanam Hospital and Research Center, Indore (MP) 452 001

The present study describes the prevalence of HBV and anti-HCV antibodies in different populations in central India. Sera samples from healthy subjects, persons with liver diseases and persons receiving multiple blood transfusions were screened for HBsAg and anti-HCV antibodies by the latest generation ELISA kits. The HBsAg carrier rate among general population has been observed to be constant (around 3%), over the period of 10 years: HCV carrier rate was seen to be 1%. Among normal pregnant
females 12,911 (1.31%) were HBsAg reactive and 1,460 (0.21%) were anti-HCV antibodies reactive. In patients with liver diseases, HBV infection was detected in 39.4% of AVH cases, 40.19% CLDs cases, and 27.8% of hepatic failure cases. HCV antibodies were detected in 1.46% (7/478) AVH cases, 18.8% of CLDs cases and 8.19% among hepatic failure cases in the same group of patients. Among multitransfused CRF subjects, HBV and HCV prevalence was detected in 10% (60/600) and 27.5% (64/232) of the cases. In thalassemia subjects, HBV and HCV prevalence was found to be 3.84% and 21.5%, respectively. The findings suggest that HBV still remains major aetiological agent among sporadic cases of hepatitis while HCV has overtaken HBV in multitransfused subjects.

Hepatitis B and C in childhood chronic liver disease.
U Jhamb, V Malhotra, SK Mittal. Department of Paediatrics, MAMC, Delhi

84 cases of chronic liver disease in children have been studied for evidence of hepatitis B or C infection. Out of these 14 (16.6%) were found to be HBsAg positive with 11 (78.6%) males and 3 (21.4%) females. Anti-HCV antibody test was available in 45 cases and out of these 4 were positive. All 4 of these were males. Only one child had both hepatitis B and C infection. Detailed analysis of HBsAg positive cases revealed history of jaundice in 8 (57.1%), ascites in 2 (14.3%), encephalopathy in 2 (14.3%) and evidence of portal hypertension in 5 (35.7%) cases. Four patients were asymptomatic clinically and the disease was diagnosed on biopsy which was done because of persistent jaundice. Liver was firm and enlarged in 9 cases and both liver and spleen were normal in 4 (28.3%) cases. History of blood transfusion was positive in 4 (28.3%) cases. Multiple family members were involved in 4 cases. Serum transaminases were increased in 5 (35.7%) patients and normal in the rest. There was poor correlation between transaminase level and severity of histological picture. Liver biopsy was done in 8 cases, out of which 6 showed a picture of chronic hepatitis and 2 had cirrhosis.

In chronic liver disease due to hepatitis B, serum enzymes correlate poorly with the histology and up to 1/3rd may have normal liver and spleen on examination and no features of portal hypertension. Only liver biopsy is helpful in diagnosing such cases. Also active screening of family members is beneficial and many asymptomatic cases may be picked up.

Exposure of tribal and island populations to HBV and HCV.
Anankalle VA,1 Padbidri VS,1 Mehandale SM,2 Gande SS,1 Umamathi UB,1 National Institute of Virology, ICGR Pune, 2National AIDS Research Institute (ICMR), Pune

Objectives: To assess the magnitude of HBV and HCV infections among tribal populations of Maharashtra and five major islands constituting the Andaman and Nicobar (A and N) group of islands.

Methods: Four tribes representing Bhawai (419), Dhule (500), Pune (272) and Thane (478) areas of the state of Maharashtra were investigated. Over 2000 serum samples from A and N islands were also screened retrospectively. ELISA and RIBA 3.0 were employed for HBsAg and anti-HCV testing, respectively.

Results: HBsAg positivity varied from 3.3% (14/419, Bhawai) to 5.5% (15/272, Pune) whereas prevalence of anti-HBs antibodies ranged from 25% (Bhawai) to 32% (Pune and Dhule). Exposure to HCV as evidenced by the presence of anti-HCV antibodies (RIBA 3.0) was infrequent. A pocket of high HBV endemicity (15.5% HBsAg carrier rate) was noted on one of the islands. Exposure to HCV varied from 0.15% (1/644) to 0.74% (1/134).

Conclusions: The study documents high exposure of the tribal populations to HBV and infrequent transmission of HCV.

Role of HIV in the suppression of HCV antibodies: A pilot study from India.
KVK Mohan, KG Murugavel, HB Fields, KR Balanisamy, S Rajanikanth, P Rajasambandam, K Raghuram, Sunil Mathews, V Jayanthi, SP Thyagarajan, Department of Microbiology, ALMPGIBM, Taramani, Madras 600 113; *Hepatitis Division, Centers for Disease Control, Atlanta, GA 30333, USA; **Department of Gastroenterology, Stanley Medical College and Hospital, Chennai 600 001; Department of Gastroenterology, Chennai Medical College, Chennai 600 003; #Department of Digestive Health and Diseases, Chennai 600 040

Objectives: To study i) the prevalence of HCV recombinant immunoblot assay (RIBA 3.0) indeterminates among the Indian population, ii) HCV-RNA and anti-HIV positivity among the RIBA indeterminates and iii) to analyse the role of HIV in the suppression of anti-HCV antibodies on follow-up.

Design: 62 RIBA indeterminate cases and 60 anti-HCV reactive cases from a total of 1,487 samples were tested for HCV-RNA and anti-HIV antibodies. The indeterminate cases were also followed up every month, for a period of 5 months to one year for HCV confirmation.

Methods: Anti-HCV was detected using the RIBA 3.0 (Chiron Corp, USA). HCV-RNA was tested by the RT-PCR technique using primers from the 5' noncoding region. Anti-HIV testing was performed using the commercially available ELISAs.

Results: Of the 1,487 samples screened by the RIBA 3.0, 62 (4.1%) were found to be RIBA indeterminates. Of these indeterminates, 33 (53.2%) had HCV-RNA in them. The RNA positivity was significantly higher in the c33 and c22 indeterminate groups than the c100 cases (p<0.001). Anti-HIV positivity was found in 15 (24.1%) of the RIBA indeterminate and 4 (6.6%) of the anti-HCV positive cases (p<0.001). Suppression of anti-HCV reactivity due to HIV
was significantly higher in the HCV RNA positive, RIBA indeterminate cases than the HCV-RNA negative, RIBA indeterminate cases (p<0.001). 12 indeterminate cases were followed-up, of which 4 were positive for anti-HIV and 8 were negative for anti-HIV. Of the 4 anti-HIV positive cases, only one (25%) seroconverted into a confirmed anti-HCV reactive status whereas, of the 8 anti-HIV negative cases, 5 (62.5%) seroconverted into a clear-cut anti-HCV reactive status (p<0.001).

Conclusions: The present study suggests that RIBA indeterminates have to be treated with caution, as a large number of the indeterminates samples may turn to be HCV-RNA positive and HIV seems to suppress the expression of anti-HCV antibodies among the HIBA indeterminate cases.

Random survey of hepatitis markers in patients and donors (a report from a private hospital). S Nagarajan, S Pareek, I Dhall. Batra Hospital and Medical Research Centre, New Delhi

At Batra Hospital and Medical Research Centre, screening of blood donors is undertaken for hepatitis B and C by two markers HBsAg and anti HCV, correspondingly these same markers were done on high risk patients who were clinically suspected to suffer from hepatitis, or were repeatedly transfused during dialysis, cardiac surgeries and other intensive care procedures. Our retrospective statistical analysis reveals that incidence of anti-HCV positivity in high risk patients has dropped from 23.15% to 6.09% from the year 1996 to 1998. This can categorically be attributed to the fact that from July '97 we are screening the blood for anti HCV. On the other hand the incidence of HBsAg has remained unchanged over the same period. This study reveals that of the 40 patients who were seropositive for HBsAg, 75% were also found positive for anti HBC.

Prevalence of co-infection of HGV in patients with HBV and HCV related cirrhosis in western India. Prabha Sawant, Pravin Rathe, Sheetal Bhadphale, Aamal Upadhyay, Viral Patrawala. Dept. of Gastroenterology, LTMMG and LTMMC, Sion, Mumbai

Objectives: To study the prevalence of hepatitis G virus (HGV) infection in patients of HBV and HCV related cirrhosis in western India.

Methods: A total of 99 patients with cirrhosis of liver proven by biopsy, laparoscopy, USG, liver scan, CT scan, clinical or biochemical profiles, combination of above modalities were studied. Sera collected from these patients were collected and stored at -80°C and were subjected to various tests. HBsAg was determined by passive hemagglutination, anti-HBe by hemagglutination inhibition. Anti-HCV was determined by the third generation ELISA (ELISA III, Ortho diagnostics) and HCV RNA by PCR with rested primers deduced from the 3'UTR, applied to those who tested positive for anti-HCV. HGV-RNA was tested by reverse transcription PCR.

Results: Detection of HGV-RNA in patients with cirrhosis in western India:

<table>
<thead>
<tr>
<th>HCV infection</th>
<th>HBV infection</th>
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</thead>
<tbody>
<tr>
<td>Anti-HCV</td>
<td>HCV-RNA</td>
</tr>
<tr>
<td>+ve</td>
<td>-ve</td>
</tr>
<tr>
<td>n</td>
<td>11</td>
</tr>
</tbody>
</table>

HGV-RNA 2 (18%) 1 (20%) 1 (17%) 1 (6%) 5 (20%) 12 (12%)

Conclusion: 1) HGV infection was found to be more common amongst patients with cirrhosis of liver due to viral infection HBV and/or HCV. 2) More so with HCV co-infection suggestive of similar mode of transmission. 3) Severity of liver disease was not affected by presence of co-infection by HBV or HCV infection.

Risk behaviour profile of blood donors at a tertiary care hospital. B Sood, SK Starin, R Saxena. Dept. of Preventive and Social Medicine, LTMG, Delhi, and Dept of Gastroenterology, GB Pant Hospital, Delhi

An essential component of donor screening prior to blood donation is risk behavior assessment. This is widely practiced in developed countries, but rarely completely in developing countries. In India, although the donor is asked few questions on certain disease conditions, questions regarding lifestyle and behavior pattern are rarely probed. This study was conducted at a tertiary care hospital, where 60% replacement donors were administered a confidential self-answering questionnaire to assess risk behavior. This study identified many risk behaviors in the donors, Premarital sex was experienced by 60% of unmarried donors and 21% of married donors. Also, one third of which in both groups was with a commercial sex worker. 21% of the donors had multiple sexual partners and more than two thirds of them did not practice safe sex. In this part of the country only two donors were found to be intravenous drug abusers. Blood transfusion had been received by only a single donor. These findings highlight that screening for risk behavior may exclude potentially unsafe donors, hence it may not only reduce the cost of collecting, storing and screening of blood, but also the resultant morbidity.

Seroprevalence of hepatitis B, hepatitis C and hepatitis E in multiple transfusion recipients. Dur MA, Yatthoo GN, Kamli S, Khuroo MS. Department of Gastroenterology, Sher-i-Kashmir Institute of Medical Sciences, Srinagar, Kashmir

Epidemiology and etiology of post transfusion hepatitis (PTH) has considerably changed during the last decade. The present study is aimed to determine the sero-prevalence of conventional PTH viruses (hepatitis B and C) and hepatitis
Hepatitis B and C: carrier to cancer — Abstracts

E virus in patients receiving blood and blood products in hepatitis E endemic area. The study was conducted in retrospective and prospective phases. In retrospective phase 145 patients having received the last transfusion at least 5 weeks prior to the date of sampling and 250 healthy non-transfused controls from the general population were enrolled. In prospective phase 25 patients required blood transfusion over a short period of 1 to 3 days, were enrolled. These patients were followed up for 2 months after transfusion. 1 pre-transfusion and 2 post-transfusion blood samples were taken at 0, 1 and 2 months respectively. Serum sample from the donor blood intended to be transfused was also stored. Liver function tests were done in patients, healthy controls and donors. In the patient population serological markers of hepatitis-B (HBsAg and IgM anti-HBc), hepatitis C (anti-HCV) and hepatitis-E (IgM and IgG anti-HEV) were assayed. In the control group only IgG anti-HEV was tested. The donor samples of only those patients who acquired hepatitis E were tested retrospectively. In the prospective group sero-prevalence of anti-HEV in multitransfused patients was 15% vs 4% in controls (p<0.001). In the prospective group of 25 transfusion recipients, 2 patients seroconverted to anti-HEV. One donor sample of one of these patients tested positive for IgM anti-HEV. Other two patients were positive for IgG anti-HEV prior to transfusion but had IgG loss at 2 months after transfusion. Sero-prevalence of anti-HCV in multi-transfused patients was 13.79%. 25% of patients with hepatitis C were also infected with HEV whereas only 13.6% of patients negative for HCV were positive for anti-HEV. Sero-prevalence of hepatitis B in multi-transfused patients was 11.7%. One patient was co-infected with HCV. The role of HCV and HBV in the PTH has been reestablished in this study. Considering the high endemicity of hepatitis E in Kashmir and high prevalence of hepatitis E in transfusion recipients, screening of donor blood for hepatitis E is recommended.

Iron profile in chronic liver disease (CLD) patients in India. N Tandon, V Thakur, RC Gupta, SK Sarin. Deptt. of Gastroenterology, GB Pant Hospital, New Delhi.

Objectives: Iron is an essential nutrient of the human body. Iron stores are primarily located in hepatic parenchyma. Hence a close association exists between iron and liver disorders. We initiated this study to assess the iron profile in liver disease patients in comparison to the healthy population.

Materials and Methods: One hundred and forty one patients with CLD (hepatitis B - 93, hepatitis C - 37, HCC - 11) and 78 HBV carriers consecutively seen were included. Ninety healthy individuals served as controls. Serum iron using colorimetric method and serum ferritin by ELISA were determined in fasting serum sample.

Results: Ninety one (65%) CLD patients, 47 (60%) HBV carriers and 56 (62%) controls were vegetarians. Mean values of serum iron, transferrin saturation index (TSI) and serum ferritin were well within the normal range in the study groups as well as controls (Table). They were however, significantly higher in HBV related CLD subjects compared to controls (p<0.05, p<0.001). Serum ferritin was significantly higher in all CLD groups, compared to controls (p<0.001). Iron parameters were in carriers compared to controls.

<table>
<thead>
<tr>
<th></th>
<th>HBV related</th>
<th>HCV related</th>
<th>HCC</th>
<th>HBV carriers</th>
<th>Controls</th>
</tr>
</thead>
<tbody>
<tr>
<td>B (μg/dL)</td>
<td>93</td>
<td>37</td>
<td>11</td>
<td>78</td>
<td>90</td>
</tr>
<tr>
<td>Age (yr)</td>
<td>40.4±11.8</td>
<td>43.5±13.4</td>
<td>56.8±14.1</td>
<td>33.5±10.5</td>
<td>37±11.3</td>
</tr>
<tr>
<td>S. iron (mg/dL)</td>
<td>105.3±68.6*</td>
<td>102.6±36.5*</td>
<td>88.2±34.1</td>
<td>96.8±46.7</td>
<td>86.7±43.7</td>
</tr>
<tr>
<td>TSI (%)</td>
<td>36.5±19.2**</td>
<td>36±15.3</td>
<td>41±21.4</td>
<td>33.6±18.7</td>
<td>30.7±15.8</td>
</tr>
<tr>
<td>S. ferritin (ng/mL)</td>
<td>110.7±8**</td>
<td>174.7±41.4**</td>
<td>119.5±34.5**</td>
<td>52.8±46.3</td>
<td>40.9±4.7</td>
</tr>
</tbody>
</table>

*p<0.05, **p<0.001 (significantly higher in CLD patients)

Conclusions: 1. The iron profile in liver disease patients is well within the normal range, though higher in comparison to controls. 2. The frequency of iron overload, very high in western countries, is much lower in India.