

## A Correlative Study Between Biochemical and Hematological Parameters and Hepatitis C Prevalence in the Premises of Faisalabad

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**Abstract:** Hepatitis C virus (HCV) is an infectious agent which is responsible for liver diseases, cirrhosis and hepatocellular carcinoma. The present work was designed to find out the correlation between biochemical and hematological parameters and its prevalence. ALT and AST values were higher than normal in HCV positive patients (119.34 U/L), (108.21 U/L) had the significance correlation with HCV. ALP values were higher and hemoglobin values were lower but in normal range (242.28 U/L), (11.34 g/dL). Hemoglobin is decreased in chronic cases when there is lower liver efficiency. Platelet count was low in some cases as this parameter is affected with progress of disease (136.69<sup>3</sup>  $\mu$ /L) and ESR is raised more than its normal values (48.48 mm/hour). There is no so significant correlation of other parameters as bear ALT and AST. So ALT is more sensitive indicator of any liver abnormality and most commonly used as the primary indicator of liver function. In conclusion, the prevalence of HCV infections is substantially higher in Faisalabad city and the Microparticle Enzyme Immuno Assay (MEIA) is a useful method for epidemiology of viral Hepatitis C.

**Key word:** Hepatitis C • ALP • ALT • AST • MEIA

### INTRODUCTION

Many infectious diseases afflicting human beings are caused by viruses. Some viral diseases are very important because they are frequently fatal; including “Viral Hepatitis” [1]. The Hepatitis C virus (HCV) is the member of the *hepacivirus* genus in the family *Flaviviridae*. Hepatitis C is a blood-borne infectious disease that is caused by the hepatitis C virus HCV, affecting the liver [2]. Asymptomatic infection occurred initially, but chronic hepatitis developed which lead to inflammation, signs and symptoms may appear that are generally caused by either decreased liver function or increased pressure in the liver circulation [3]. The mechanisms responsible for the HCV life cycle in the liver of infected individuals are only partially understood because it has not been possible to infect normal human hepatocytes in culture with naturally occurring HCV obtained from HCV-infected patients and because HCV is known to infect only humans and chimpanzees [4]. The symptoms of acute hepatitis C infection include decreased appetite, fatigue, abdominal pain, jaundice,

itching and flu-like symptoms [5]. Approximately 15-40% of persons infected with HCV clear the virus from their bodies during the acute phase as shown by normalization in liver function tests (LFTs) such as alanine transaminase (ALT) and aspartate transaminase (AST) normalization, as well as plasma HCV-RNA clearance. The remaining 60-85% of patients infected with HCV develops chronic hepatitis C [6]. HCV transmission by in apparent percutaneous exposures has been caused by cross-contamination from reused syringes, needle, multiple-use medication vials, infusion bags and injecting-drug use paraphernalia [7].

The present research work will provide update data regarding hepatitis C prevalence, its correlation with biochemical and hematological parameters in the population of Faisalabad (Pakistan).

### MATERIALS AND METHODS

Research work was planned to find out the prevalence of Hepatitis C and its epidemiology from different regions of Faisalabad.

**Place of Work:** All the serological and biochemical analyses were performed in Bio-care Lab., Aziz Fatimah Hospital, Institute of Molecular Biology and Biotechnology, The University of Lahore and Enzyme Biotechnology Lab., University of Agriculture, Faisalabad.

**Patient Selection:** Sample collection was done from selected 100 individuals randomly from different regions of Faisalabad. Patients were considered to have hepatitis C infection if their ELISA was positive and had raised alanine aminotransferase (ALT).

**Biochemical and Hematological Analysis:** All samples were tested on AXSYM fully automated immune-analyzer from Abbott Diagnostics USA, obeying the Micro particle Enzyme Immune Assay (MEIA) principle, by the 3rd generation technology [6]. Alanine aminotransferase (ALT), Aspartate aminotransferase (AST) and Alkaline phosphatase (ALP) were estimated on Hitachi 902 fully automated chemistry analyzer by Roche Diagnostics Germany. A kinetic method for the determination of ALT activity according to recommendations of IFCC without pyridoxalphosphate activation [8]. Hematology parameters Hb, PLT were carried out on Sysmex XS800i (5-part Differential fully automated hematology analyzer) and ESR was done on MicroSed system by vital Diagnostics.

**Blood Samples Treatment:** 6-8 ml blood was drawn by using sterilized syringes by vein puncture technique, divided it into two tubes, 4-5ml in plain sterilized tube and tri-Potassium EDTA coated tubes. Plain tube was allowed to left more than half hour at room temperature for clot, then finally centrifuge at 4000 rpm for 15 minutes, separated serum fractions were transferred in autoclaved aliquotes and placed in AXSYM.

## RESULTS

The age groups of 10-20 years contains 10% individuals out of which 80% were found negative and 20% cases were shown result as positive for hepatitis C. In negative cases the results revealed that serum ALT, AST and ALP levels were  $26.38 \pm 5.78$ ,  $23.25 \pm 4.95$  and  $168.50 \pm 16.81$  U/L, respectively while positive cases revealed the value of serum ALT, AST and ALP  $73.50 \pm 12.02$ ,  $64.50 \pm 14.85$  and  $285.00 \pm 42.43$  U/L, respectively (Figs. 1,2,3). The hemoglobin concentration in negative and positive cases was  $13.94 \pm 1.47$  and  $11.15 \pm 1.63$  g/dL, respectively (Fig. 4). The platelet counts of negative cases were  $244.50 \pm 80.26$  and among positive

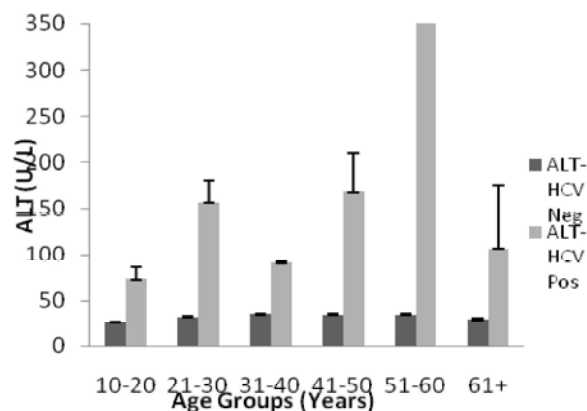


Fig. 1: Serum ALT levels in Hepatitis C among (-ve) and (+ve) cases in different age groups

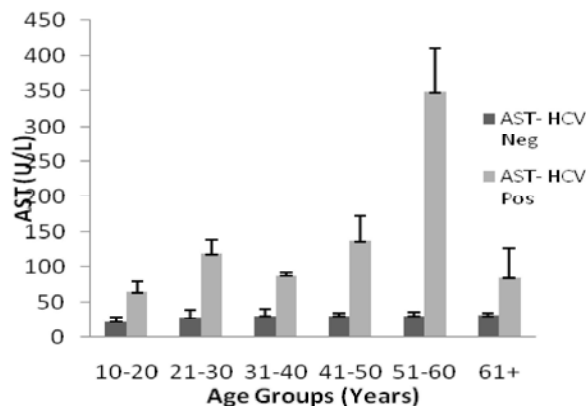


Fig. 2: Serum AST levels in Hepatitis C among (-ve) and (+ve) cases in different age groups.

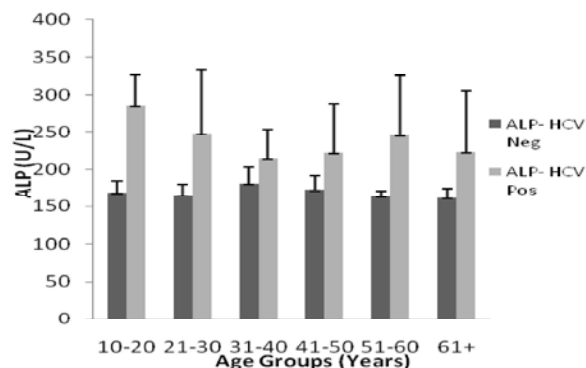


Fig. 3: Serum ALP levels in Hepatitis C among (-ve) and (+ve) cases in different age groups.

cases it was  $110.00 \pm 14.14$   $\mu$ L (Fig. 5). The ESR (erythrocyte sedimentation rates) of negative cases was  $7.75 \pm 5.85$  and the values of ESR find raised in the positive cases as  $47.50 \pm 10.61$  mm/h (Fig. 6).

The age groups of 21-30 years contains 60% individuals out of which 73.33% were found negative and 26.67% cases were declared as positive for hepatitis C.

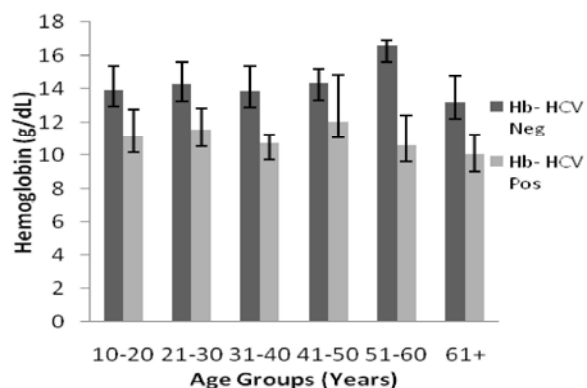


Fig. 4: Hb Concentration in Hepatitis C among (-ve) and (+ve) cases in different age groups.

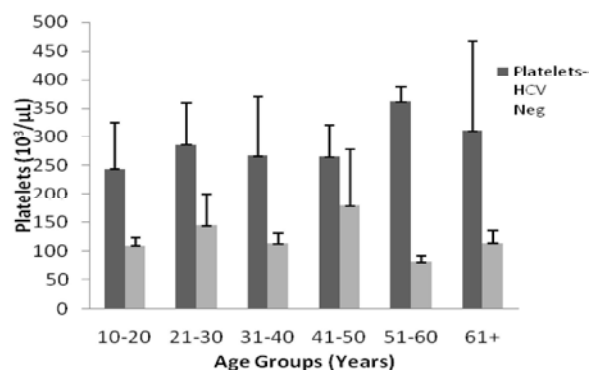


Fig. 5: Platelets counts in Hepatitis C among (-ve) and (+ve) cases in different age groups.

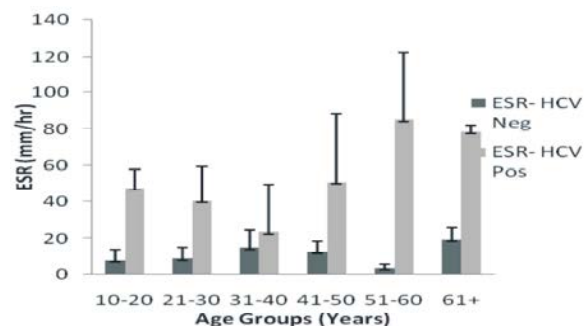


Fig. 6: ESR in Hepatitis C among (-ve) and (+ve) cases in different age groups.

In negative cases the results revealed that serum ALT, AST and ALP levels were  $32.85 \pm 12.37$ ,  $28.60 \pm 10.62$  and  $165.58 \pm 14.62$  U/L, respectively while positive cases revealed the value of serum ALT, AST and ALP  $98.19 \pm 23.58$ ,  $87.56 \pm 19.67$  and  $247.13 \pm 86.71$  U/L, respectively (Figs. 1,2,3). The hemoglobin concentration in negative and positive cases was  $14.29 \pm 1.34$  and  $11.56 \pm 1.27$  g/dL, respectively (Fig. 4). The platelet counts of negative cases were  $286.43 \pm 73.08$  and among positive cases it was  $145.50 \pm 53.91$   $10^3/\mu\text{L}$  (Fig. 5).

The ESR (erythrocyte sedimentation rates) of negative cases was  $8.85 \pm 5.98$  and the values of ESR find raised in the positive cases as  $40.38 \pm 19.41$  mm/h (Fig. 6).

The age groups of 31-40 years contains 11% individuals out of which 90.91% were found negative and 9.09% cases were declared as positive for hepatitis C. In negative cases the results revealed that serum ALT, AST and ALP levels were  $35.67 \pm 3.79$ ,  $30.44 \pm 10.15$  and  $181.56 \pm 21.82$  U/L, respectively while positive cases revealed the value of serum ALT, AST and ALP  $91.00 \pm 1.41$ ,  $87.50 \pm 3.54$  and  $214.50 \pm 38.89$  U/L, respectively (Figs. 1,2,3). The hemoglobin concentration in negative and positive cases was  $13.94 \pm 1.47$  and  $13.89 \pm 1.50$  g/dL, respectively (Fig. 4). The platelet counts of negative cases were  $266.78 \pm 103.96$  and among positive cases it was  $114.00 \pm 16.97$   $10^3/\mu\text{L}$  (Fig. 5). The ESR (erythrocyte sedimentation rates) of negative cases was  $14.89 \pm 9.61$  and the values of ESR find raised in the positive cases as  $23.50 \pm 26.16$  mm/h (Fig. 6).

The age groups of 41-50 years contains 10% individuals out of which 60% were found negative and 40% cases were declared as positive for hepatitis C. In negative cases the results revealed that serum ALT, AST and ALP levels were  $35.33 \pm 3.79$ ,  $30.67 \pm 3.21$  and  $173.00 \pm 19.52$  U/L, respectively while positive cases revealed the value of serum ALT, AST and ALP  $99.50 \pm 41.15$ ,  $90.00 \pm 34.73$  and  $222.00 \pm 66.39$  U/L, respectively (Figs. 1,2,3). The hemoglobin concentration in negative and positive cases was  $14.33 \pm 0.84$  and  $12.05 \pm 2.80$  g/dL, respectively (Fig. 4). The platelet counts of negative cases were  $265.33 \pm 80.26$  and among positive cases it was  $110.00 \pm 54.64$   $10^3/\mu\text{L}$  (Fig. 5). The ESR (erythrocyte sedimentation rates) of negative cases was  $12.67 \pm 5.77$  and the values of ESR find raised in the positive cases as  $51.25 \pm 37.05$  mm/h (Fig. 6).

The age groups of 51-60 years contains 5% individuals out of which 40% were found negative and 60% cases were declared as positive for hepatitis C. In negative cases the results revealed that serum ALT, AST and ALP levels were  $35.50 \pm 2.12$ ,  $23.25 \pm 4.95$  and  $164.50 \pm 6.36$  U/L, respectively while positive cases revealed the value of serum ALT, AST and ALP  $73.50 \pm 12.02$ ,  $64.50 \pm 14.85$  and  $246.00 \pm 80.07$  U/L, respectively (Figs. 1,2,3). The hemoglobin concentration in negative and positive cases was  $16.60 \pm 0.28$  and  $10.60 \pm 1.83$  g/dL respectively (Fig. 4). The platelet counts of negative cases were  $362.50 \pm 80.26$  and among positive cases it was  $81.00 \pm 10.54$   $10^3/\mu\text{L}$  (Fig. 5). The ESR (erythrocyte sedimentation rates) of negative cases was  $3.50 \pm 2.12$  and the values of ESR find raised in the positive cases as  $85.33 \pm 37.07$  mm/h (Fig. 6).

The age groups of 61+ years contains 4% individuals out of which 50% were found negative and 50% cases were declared as positive for hepatitis C. In negative cases the results revealed that serum ALT, AST and ALP levels were  $29.50 \pm 6.36$ ,  $31.50 \pm 3.54$  and  $161.50 \pm 13.44$  U/L, respectively, while positive cases revealed the value of serum ALT, AST and ALP  $140.50 \pm 68.59$ ,  $115.00 \pm 43.84$  and  $223.50 \pm 81.32$  U/L, respectively (Figs. 1,2,3). The hemoglobin concentration in negative and positive cases was  $13.20 \pm 1.56$  and  $10.05 \pm 1.20$  g/dL, respectively (Fig. 4). The platelet counts of negative cases were  $310.50 \pm 156.27$  and among positive cases it was  $114.50 \pm 21.92$   $\mu$ L (Fig. 5). The ESR (erythrocyte sedimentation rates) of negative cases was  $19.50 \pm 6.36$  and the values of ESR find raised in the positive cases as  $78.50 \pm 3.54$  mm/h (Fig. 6).

## DISCUSSION

Higher values of ALT in HCV positive patients were found in this present study while lower values in HCV negative patients that were in normal range. However an increased level of ALT is generally a result of liver disease associated with some degree of hepatic necrosis such as cirrhosis, carcinoma, viral or toxic hepatitis [9]. Normally ALT remained within the hepatocytes, but on damaging of hepatocyte, the enzyme escape out from hepatocytes and raised levels were found in the blood hence, there is significant correlation of ALT and HCV. As both ALT and AST are associated with liver diseases, liver damage, intraoperative hypotension, intraoperative blood loss, liver resection significantly correlated with liver enzyme elevations [9].

In HCV positive patients the higher values of AST were found as the raised values are found in liver cirrhosis, haemolysis, myocardial infarction, acute viral hepatitis, toxic hepatitis. As Very high values are also obtained in toxic hepatitis due to carbon tetrachloride poisoning and obstructive jaundice. ALT level is also found to be increased with age [5]. Increase in both transaminases is found in liver diseases with ALT much higher than AST. Serum ALT remains the most accessible test for monitoring the chronic hepatitis C viral infection [5]. AST/ALT ratio is highly specific and predictive (100%) of cirrhosis in patients with chronic HCV infection. The mean AST/ALT ratio in patients with chronic HCV infection with cirrhosis was significantly higher than in patients without cirrhosis. The ratio correlated positively with the stage of fibrosis [10]. The present study revealed higher values of ALP in HCV positive and lower values in

HCV negative individuals that were with in the normal range. It has been reported in literature that serum ALP level increased with increasing age, body mass index, C-reactive protein, monocyte count, serum uric acid, lead, cadmium, hyper cholesterolemia, diabetes, smoking, non-alcohol drinking, sex, age, liver diseases, lesion of liver and cardiovascular disease [11].

Our results indicated lower values of hemoglobin in HCV positive patients while values in HCV negative patients were within normal limits. Thus in advanced liver disease when albumin synthesis is impaired, the plasma oncotic pressure which prevents loss of fluid into tissue spaces is lost and generalized edema occurs. As in hepatitis the synthesis of albumin is impaired so lower values are found in hepatitis and liver cirrhosis. There is significance correlation of hemoglobiin and HCV as the values are slightly lower in HCV positive patients [12].

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## REFERENCES

1. Johnson, R., D. Gretch, H. Yamabe, J. Hart, C. Bacchi, P. Hartwell, W. Couser, L. Corey, M. Wener and C. Alpers, 1993. Membranoproliferative glomerulonephritis associated with hepatitis C virus infection. *J. Med.*, 28(7): 465-470.
2. Cox, A.L., D.M. Netski and T. Mosbrugger, 2005. Prospective evaluation of community acquired acute-phase hepatitis C virus infection. *Clinical Infectious Dis.*, 40(7): 951-958.
3. Wrong, J.B., R.S. Koff, F. Tine and S.G. Puker, 1995. Cost effectiveness of interferon alpha 2b for the treatment of chronic hepatitis B. *Annual International. Med.*, 122(9): 664-675.
4. Tews, B.A. and J. Dubuisson, 2009. Occludin, the final essential factor for HCV entry? *Future Virol.*, 4(4): 329-333.
5. Tsang, P.S., H. Trinh, R.T. Garcia, J.T. Phan, N.B. Ha, H. Nguyen, K. Nguyen, E.B. Keeffe and M.H. Nguyen, 2008. Significant prevalence of histologic disease in patients with chronic hepatitis B and mildly elevated serum alanine aminotransferase levels. *Clinical Gastroenterology and Hepatol.*, 6(5): 569-74.

6. Miyamura, T., I. Saito, T. Katayama, S. Kikuchi, A. Tateda, M. Houghton, Q.L. Choo and G. Kuo, 1990. Detection of antibody against antigen expressed by molecularly cloned hepatitis C virus cDNA: application to diagnostic and blood screening for post transfusion hepatitis. *Proceeding National Academy of Science USA*, 87: 983-987.
7. Alter, M.J., 2007. Epidemiology of hepatitis C virus infection. *World J. Gastroenterol.*, 13(17): 2436-2441.
8. IFCC (International Federation of Clinical Chemistry), 1986. Methods for the measurement of catalytic concentration of enzymes. *Journal of Clinical Chemistry and Clinical Biochemistry*, 24: 481.
9. Hsu, S.H., C.Y. Chan, T.N. Tam, S.H. Lin, K.C. Tang and S.D. Lee, 1996. The liver biochemical tests and serological markers of hepatitis B virus in the very old-aged population in Taiwan. *Zhonghua Yi Xue Za Zhi (Taipei)*. 57(1): 16-21.
10. Sunil, G., M.D. Sheth, L. Steven, M.D. Flamm, D. Fredric, M.D. Gordon and S. Chopra. 1998. AST/ALT Ratio Predicts Cirrhosis in Patients with Chronic Hepatitis C Virus Infection. *American Journal of Gastroenterol.*, 93: 44-48.
11. Zhonghua, Y.I., S.H. Hsu, C.Y. Chan, T. N. Tam, S.H. Lin, K.C. Tang, S.D. Lee and X.Z. Zhi. 1996. The liver biochemical tests and serological markers of hepatitis B virus in the very old-aged population in Taiwan Jan., 57(1): 16-21.
12. Dufour, D.R., J.A. Lott, F.S. Nolte, D.R. Gretch, R.S. Koff and L.B. Seeff, 2000. Diagnosis and Monitoring of Hepatic Injury and Performance Characteristics of Laboratory Tests. *Clinical Chemistry*, 46(12): 2027-2049.