Changing trends of seropositivity of HBsAg and HCV in blood donors

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Abstract

Introduction: The term “Viral hepatitis” is reserved for infection of liver caused by a group of viruses having a particular affinity for liver. Liver disease due to Hepatitis B Virus is a huge problem globally with an estimated worldwide carrier rate of 25% of the total infected cases; every year there are over 4 million acute clinical cases of HBV infection.

Method: All blood units donated by the voluntary donors are tested by serological method for the presence of various antigens or antibodies to detect the seropositive status of the donor before transfusion of that blood unit to any patient.

Result: In the year 2013 prevalence of HBsAg was 0.72% and that of HCV was 0.11% whereas in the year 2014 prevalence of HBsAg was 0.788% and that of HCV was 0.218% respectively.

Keywords: Trend of HBsAg and HCV prevalence

1. Introduction

The term “Viral hepatitis” is reserved for infection of liver caused by a group of viruses having a particular affinity for liver[1]. Types of Hepatitis viruses are: A, B, C, D, E and G respectively.

HBV is transmitted through percutaneous or parenteral contact with infected blood, body fluids and by sexual intercourse. Susceptibility to HBV is generally only to those who have not been vaccinated successfully or those who have developed anti-HBs antibodies after infection are immune to it. In highly endemic areas, HBV is most commonly spread from mother to child at birth or from person to person in early childhood[3,4,5].

Hepatitis C virus is a major cause of liver disease worldwide. HCV is parenterally transmitted especially in transfusion recipients; up to 1% of blood units might contain the responsible virus. Transmission occurs by percutaneous exposure to contaminated blood and plasma derivatives.

2. Materials and Methods

Hepatitis viruses are most commonly transmitted by blood and blood products, so it is very important as well as mandatory to screen the voluntary blood donors and test for the seropositivity thereafter, every unit of blood is to be tested for its seropositive status before being actually transfused.

In 2013, there were 10781 blood donors and in 2014 there were 14221 blood donors respectively. All of the blood bags were tested for HBsAg, HCV, HIV, VDRL and MP, which are mandatory pre-transfusion tests. For HBsAg screening, Microscreen 3rd generation kit (by Span Diagnostics) is used and for HCV screening, QUALISA 3rd generation kit was used. All the bags that are seropositive in step 1 ELISA are tested again by ELISA in step 2 and by Rapid card test in step 3.

2.1 For HBsAg screening: all the samples and all the reagents except Colour reagent and conjugate, are brought to room temperature before starting the procedure.

- 100 microlitre of sample diluents is added to required number of wells except blank well i.e. 1A.
- 100 microlitre Negative control is added to wells 1B, 1C, 1D & 100 microlitre Positive control to 1E & 1F.
- 100 microlitre of sample is added to the rest of the wells.
Then it is incubated for 60 minutes at 37°C.
Then washing procedure is done with diluted Washing Buffer.
Then 50 microlitre of Conjugate stabiliser is added followed by 100 microlitre of Conjugate reagent 2 to each well except 1A.
Again it is incubated for 30 minutes at 37 degree Celsius.
Washing procedure is again repeated.
Then 100 microlitre of Colour reagent is added except 1A.
Again it is incubated for 30 minutes at 37 degree Celsius.
Washing procedure is again repeated.
Then 100 microlitre of Colour reagent is added to all the wells including Blank wells. Again its is incubated for 30 minutes at room temperature in dark.
Then 100 microlitre of stopping solution is added to each well and the contents are mixed well.
The absorbance is read within 30 minutes with the microwell plate reader using primary filter at 450 nanometre and reference filter at 600-700 nanometre after blanking with 1A well and then result is calculated.

2.2 For HCV screening:

ELISA protocol sheet indicating the location of controls and specimen was prepared.
Controls are used in duplicate.
200 microlitre of sample diluents is added to separate wells.
Then 10 microlitre of control or sample is added in the wells.
The plate is then gently shaken to mix the contents.
Then it is incubated for 30 minutes at 22-28°C.
Washing procedure is done by a Washing Buffer.
Then 100 microlitre of diluted conjugate is added in each well and incubated for 30 minutes at 22-28°C.
Then washing procedure is repeated.
Then 100 microlitre substrate is added and incubated at 22-28 degree Celsius away from light for 30 minutes.
Lastly, 100 microlitre of Stop solution is added and the absorbance is read at 450 nanometre with 600-700 nanometre as reference within 30 minutes of stopping the reaction.

*In this study only the frankly reactive cases are considered as positive, i.e., out of the 3 steps (2 steps of ELISA and 3rd step of Rapid card test), if any 2 steps are positive, the blood sample is considered seropositive.

3. Results
The above data shows the change in the prevalence of seropositivity status of the blood donors for the Hepatotropic viruses.

Table 1: The table below shows the monthly variation in the number of seropositive cases

<table>
<thead>
<tr>
<th>Year</th>
<th>Hepatitis virus</th>
<th>Jan</th>
<th>Feb</th>
<th>Mar</th>
<th>Apr</th>
<th>May</th>
<th>Jun</th>
<th>Jul</th>
<th>Aug</th>
<th>Sep</th>
<th>Oct</th>
<th>Nov</th>
<th>Dec</th>
</tr>
</thead>
<tbody>
<tr>
<td>2013</td>
<td>HBV</td>
<td>04</td>
<td>02</td>
<td>08</td>
<td>05</td>
<td>09</td>
<td>06</td>
<td>07</td>
<td>10</td>
<td>03</td>
<td>10</td>
<td>05</td>
<td>07</td>
</tr>
<tr>
<td></td>
<td>HCV</td>
<td>01</td>
<td>01</td>
<td>02</td>
<td>00</td>
<td>01</td>
<td>01</td>
<td>00</td>
<td>02</td>
<td>00</td>
<td>01</td>
<td>02</td>
<td>01</td>
</tr>
<tr>
<td>2014</td>
<td>HBV</td>
<td>01</td>
<td>04</td>
<td>11</td>
<td>05</td>
<td>08</td>
<td>06</td>
<td>10</td>
<td>15</td>
<td>15</td>
<td>11</td>
<td>11</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>HCV</td>
<td>01</td>
<td>01</td>
<td>02</td>
<td>01</td>
<td>03</td>
<td>04</td>
<td>02</td>
<td>10</td>
<td>03</td>
<td>02</td>
<td>01</td>
<td>01</td>
</tr>
</tbody>
</table>

The table and the chart below show the difference in the prevalence of various blood borne infections in the year 2013 & 2014:

Table 2: Difference in the prevalence of various blood borne infections in the year 2013 & 2014

<table>
<thead>
<tr>
<th></th>
<th>2013</th>
<th>2014</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Blood Donors</td>
<td>10781</td>
<td>14221</td>
</tr>
<tr>
<td>HIV reactive</td>
<td>(0.08%)</td>
<td>(0.098%)</td>
</tr>
<tr>
<td>HbsAg reactive</td>
<td>78 (0.72%)</td>
<td>112 (0.788%)</td>
</tr>
<tr>
<td>HCV reactive</td>
<td>12 (0.11%)</td>
<td>31 (0.218%)</td>
</tr>
<tr>
<td>VDRL reactive</td>
<td>06 (0.056%)</td>
<td>05 (0.035%)</td>
</tr>
<tr>
<td>MP positive</td>
<td>00 (0.0%)</td>
<td>00 (0.0%)</td>
</tr>
</tbody>
</table>

Figure 1: The figure below depicts the comparative increase in the number of HCV cases in the year 2014 as compared to the year 2013.
4. Discussion

The word Viral Hepatitis is used to describe infection of Liver caused by Hepatotropic viruses. Based on etiologic agent Viral Hepatitis is classified into 6 etiological types: Hepatitis A, Hepatitis B, Hepatitis C, Hepatitis D, Hepatitis E and Hepatitis G respectively.

Hepatitis A: also known as Infectious Hepatitis, responsible for 20-25% of clinical hepatitis in developing countries. It is usually spread by Faeco-oral route, parenteral transmission is extremely rare.

Hepatitis B: also known as Serum Hepatitis caused by HBV and is transmitted by recipients of blood and blood products, intravenous drug addicts, patients undergoing dialysis and Hospital workers. The outcomes of HBV infection are age-dependant and include symptomatic infection, acute hepatitis B, chronic HBV infection, cirrhosis and HCC[6].

Hepatitis D: Infection with Delta virus (HDV) in the hepatocyte nuclei of HBsAg-positive patients is termed Hepatitis D. HDV is a defective virus for which HBV is the helper. HDV infection and Hepatitis B may be simultaneous (Co-infection) or HDV may infect a chronic HBsAg carrier (Superinfection).

Hepatitis C: The diagnosis of this 3rd major category of hepatitis was made after exclusion of infection with other known viruses and was initially designated as non-A, non-B (NANB) hepatitis. However now this type has been characterised and now called Hepatitis C.

Hepatitis E: It is an enterically transmitted virus. The infection is generally acquired by contamination of water supplies such as monsoon flooding. It is not associated with chronic liver disease.

Hepatitis G: HGV infection has been found in blood donors, patients on hemodialysis and as co-infection with HIV.

HGV is cleared from plasma in majority of individuals and only a small percentage of cases have chronic infection, which do not develop hepatitis nor does infected blood require screening for HGV RNA.

5. Prevention

Prevention of chronic HBV infection has become a high priority in the global community. Routine screening of blood donors for HbsAg and HCV can efficiently exclude those donors who are persistent, low-level carriers and those in the window period of acute infection. For preventing HBV infection Vaccine is the most effective way. HBIG (immunoglobulin) protects by passive immunization if given shortly before or soon after exposure. It is also given in combination with HBV vaccines to newborns of HbsAg positive mothers. Integrating HB vaccine into childhood vaccination schedule has shown to interrupt HBV transmission. Universal precautions should be used while handling human blood and body fluids. Good personal hygiene can limit spread of hepatitis virus. Autoclaving and use of Ethylene oxide gas are accepted methods for disinfecting metal objects or equipments. In endemic areas Mass immunization campaigns are very effective.

6. Conclusion

In the year 2013 prevalence of HBsAg was 0.72% and that of HCV was 0.11% whereas in the year 2014 prevalence of HBsAg was 0.788% and that of HCV was 0.218%.

References