**Hepatitis B and C virus in West-Central Turkey: Seroprevalence in healthy individuals admitted to a university hospital for routine health checks**

Bir üniversite hastanesine rutin sağlık kontrolü için başvuranlarda hepatit B ve C virus seroprevalanlığı

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Departments of 'Infectious Diseases and Clinical Microbiology, 'Family Medicine and 'Microbiology, Afyon Kocatepe University School of Medicine, Afyon

**Background/aims:** The aim was to determine the seroprevalence rates for hepatitis B virus and hepatitis C virus and the frequency of hepatitis B core antibody seropositivity alone in residents of a city in Turkey. The subjects visited the Family Medicine Outpatient Clinics of a university hospital in Afyon for routine health checks between January 2002 and January 2004. **Methods:** A single serum sample was collected from subjects examined. The sera were analyzed by ELISA for hepatitis B surface antigen, hepatitis B surface antibody, anti-HBcIgG and anti-hepatitis C virus. Samples that were HBsAg-positive were also screened for hepatitis B early antigen and antibody. Subjects who were positive for anti-HBcIgG alone were considered to have "isolated anti-HBcIgG positivity". **Results:** 1320 persons were screened. The rates of positivity for HBsAg, anti-HBs, isolated anti-HBcIgG, and anti-hepatitis C virus were 6.6%, 27.2%, 6.1%, and 2.2%, respectively. Eight percent of the 87 HBsAg-positive individuals were HBcAg-positive. Comparison of marker detection rates according to sex and age (younger than 50 years vs 50 years or older) revealed a significantly higher prevalence of HBsAg positivity in males than in females (p=0.02), and a significantly higher prevalence of anti-hepatitis C virus and anti-HBcIgG positivity in the older than in the younger group (p=0.001 and p=0.001, respectively). **Conclusion:** According to our results, the rates of hepatitis B virus and hepatitis C virus seropositivities in our region are similar to those reported in recent studies from other parts of Turkey. But these results cannot be extrapolated to all residents of the Afyon area because a random sampling method was not used for statistical analysis. However, they provide a good reference for future studies because of the large number of cases investigated.

**Key words:** Hepatitis B virus, hepatitis C virus, seroprevalence

**INTRODUCTION**

Hepatitis B virus (HBV) and hepatitis C virus (HCV) infections are still major public health problems around the globe. These diseases cause chronic hepatitis, cirrhosis, and hepatocellular carcinoma, all of which are serious public health concerns (1). Recent estimates indicate that there
are currently 2 billion people infected with HBV and 170 million infected with HCV worldwide (2, 3).

Many researchers have investigated prevalence rates of HBV and HCV infections in various groups (blood donors, health care workers, hemodialysis patients, and others) from different parts of Turkey. The data indicate that infection rates differ according to region. According to the findings, the rates of hepatitis B surface antigen (HBsAg) positivity and anti-HCV positivity in our country range from 1%-14.3% and 0.2%-2.4%, respectively (4, 5).

Anti-hepatitis B core antibody (anti-HBcIgG) is another serological marker for HBV infection. This antibody is frequently detected in combination with HBsAg or hepatitis B surface antibody (anti-HBs). Positivity for anti-HBcIgG alone may indicate past infection with HBV, but this finding is rare; patients with this marker are almost always positive for HBsAg or anti-HBs. Anti-HBcIgG positivity alone can also signal occult HBV infection, and this finding demands special consideration (6).

In this study, we assessed seroprevalence rates of HBV and HCV and the frequency of anti-HBcIgG seropositivity alone in healthy people who visited a university hospital in west-central Turkey for routine health checks.

MATERIALS AND METHODS

This prospective study involved patients who visited the Family Medicine Outpatient Clinics of Kocatepe University Hospital in Afyon, Turkey (population 130,000) for routine health checks between January 2002 and January 2004. None of the individuals had specific complaints; all visits were regular check-ups. Each person underwent a full physical examination and routine laboratory testing. Since hepatitis is endemic in Turkey, the latter included HBV and HCV serology. Individuals who had been previously diagnosed as carriers of HBV or HCV, those with history of jaundice, and those younger than 15 or older than 70 years of age were excluded.

Each subject included in the study was screened for the serological markers HBsAg, anti-HBs, anti-HBcIgG and anti-HCV using commercial kits based on a micro-ELISA method (Biomerieux, France). Rates of positivity for HBsAg, anti-HCV, and anti-HBs were analyzed according to age and sex. Patients who exhibited anti-HBcIgG positivity alone (i.e., those who were positive for anti-HBcIgG but negative for HBsAg and anti-HBs) were considered to have “isolated anti-HBcIgG positivity.” Rates of isolated anti-HBcIgG were also evaluated according to age and sex.

In addition to age and sex categories, the anti-HBs-positive subgroup was divided according to vaccination status (natural versus vaccinal HBV immunity). Subjects who tested positive for both anti-HBs and anti-HBcIgG were considered to have natural immunity to HBV infection. Those who were positive for anti-HBs alone were considered to have vaccine-induced immunity.

All subjects who were HBsAg-positive were further screened for hepatitis B e antigen (HBeAg) and antibody (anti-HBe). HBeAg and anti-HBe seropositivity were determined by micro-ELISA method using commercial kits (Organon Teknika Biomerieux, France).

The frequencies of positivity for HBsAg, anti-HBs, isolated anti-HBcIgG, and anti-HCV in the different categories that were analyzed were all compared using Fisher’s exact test.

RESULTS

A total of 1320 individuals (age range, 15-70 years) were seen for regular health checks during the study period, and all were included in the study. The total comprised 714 (54%) females and 606 (46%) males. The mean age of the females was 40.7±13.9 years and that of males 41.2±14.5 years. For analysis, the entire group was also divided into a younger and an older age group (15-49 years and 50-70 years, respectively).

Of the 1320 total subjects, 87 (6.6%) were HBsAg-positive, 359 (27.2%) were anti-HBs-positive, 81 (6.1%) were isolated anti-HBcIgG-positive, and 29 (2.2%) were anti-HCV-positive. None of the subjects was simultaneously positive for HBsAg and anti-HCV. The rates of HBsAg positivity in females and males were 4.6% and 9%, respectively. The corresponding rates for anti-HCV positivity were 2% and 2.5% and for anti-HBs positivity were 27% and 27.4%. The rates of isolated anti-HBcIgG positivity in females and males were 5.9% and 6.4%, respectively. Comparison of rates for the two sexes revealed a significantly higher rate of HBsAg positivity in males than in females (p=0.02), but no statistically significant sex differences were detected for anti-HCV, anti-HBs, or isolated anti-
HBcIgG (p>0.05, p>0.05, and p>0.05, respectively).

In the younger age group, the rates of HBsAg, anti-HBs, isolated anti-HBcIgG and anti-HCV seropositivity were 7%, 26.3%, 3.6% and 1.3%, respectively. The corresponding rates in the older age group were 5.6%, 29.5%, 12.5% and 4.5%. The older group had significantly higher frequencies of isolated anti-HBcIgG positivity and anti-HCV than the younger group (p=0.0001 and p=0.001, respectively), but there were no significant differences between the rates of HBsAg and anti-HBs positivity in these two groups (p>0.05 and p>0.05, respectively). Table 1 shows the findings in the 1320 subjects grouped according to age and sex.

Of the subgroup with natural immunity, 135 (49%) were males and 138 (51%) females, and 171 (63%) were in the younger group and 102 (37%) in the older group. Statistical comparison showed that younger subjects accounted for a significantly larger proportion of the two immunity-type subgroups than older individuals (p = 0.0001 for the vaccinal immunity group, p = 0.004 for the natural immunity group). Table 2 shows the distributions of anti-HBs-positive subjects according to sex, age group, and HBV immunity status.

Seven (9%) of the 87 HBsAg seropositive subjects were identified as HBeAg-positive. The other 80 subjects were identified as HBeAg-negative, anti-

### Table 1. Distribution of subjects according to sex and age

<table>
<thead>
<tr>
<th>Serological marker</th>
<th>Female (n=714)</th>
<th>Male (n=606)</th>
<th>Younger age group (n=944)</th>
<th>Older age group (n=376)</th>
<th>Total (n=1320)</th>
</tr>
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<tbody>
<tr>
<td>HBsAg</td>
<td>33 (4.6%)</td>
<td>54 (9%)</td>
<td>66 (7%)</td>
<td>21 (5.6%)</td>
<td>87 (6.6%)</td>
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<tr>
<td>Anti-HBs</td>
<td>193 (27%)</td>
<td>166 (27.4%)</td>
<td>248 (26.3%)</td>
<td>111 (29.5%)</td>
<td>359 (27.2%)</td>
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<td>Isolated anti-HBcIgG</td>
<td>42 (5.9%)</td>
<td>39 (6.4%)</td>
<td>34 (3.6%)</td>
<td>47 (12.5%)</td>
<td>81 (6.1%)</td>
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<tr>
<td>Anti-HCV</td>
<td>14 (2.2%)</td>
<td>15 (2.5%)</td>
<td>12 (1.3%)</td>
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### Table 2. Distributions of the anti-HBs-positive subjects by sex and age according to HBV immunity status

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<th>Subject grouping</th>
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| Male             | 31 (36%)
| Female           | 55 (64%)
| Younger age group | 77 (90%)
| Older age group  | 9 (10%)
| Total            | 86 (24%)

A total of 359 subjects (27.2% of total; 166 (27.4%) of all males and 193 (27%) of all females) were found to be anti-HBs seropositive (Table 1). Eighty-six (24%) of these individuals had vaccine immunity against HBV infection and the other 273 subjects (76%) were naturally immune to HBV. Of the subgroup with vaccine-induced immunity, 31 (36%) were males and 55 (64%) females, and 77 (90%) were in the younger group and 9 (10%) in the older group. Of the subgroup with natural immunity, 135 (49%) were males and 138 (51%) females, and 171 (63%) were in the younger group and 102 (37%) in the older group. Statistical comparison showed that younger subjects accounted for a significantly larger proportion of the two immunity-type subgroups than older individuals (p = 0.0001 for the vaccinal immunity group, p = 0.004 for the natural immunity group). Table 2 shows the distributions of anti-HBs-positive subjects according to sex, age group, and HBV immunity status.

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Hepatitis B and C virus infections are important community health problems in Turkey, and different regions have different seroprevalence rates for these viruses (4). Many studies have examined HBV and HCV infection in Turkey, and the results indicate that the prevalence of HBsAg positivity ranges from 1% to 14.3%, depending on geographical area (4). Based on these reports, Turkey is considered a region with medium endemicity for HBV infection (4).

The current study is the first to have examined HBV and HCV seroprevalence in Afyon, a city in west-central Turkey. We sought to determine the seroprevalence of HBV and HCV in healthy people who visited our hospital for routine health checks. The data revealed 6.6% seropositivity for HBsAg and 2.2% seropositivity for anti-HCV. These results are similar to those reported in recent studies from other parts of Turkey (4, 5).

As noted, the population of Afyon is 130,000. Kocatepe University Hospital is the only university health center in the city, and people from all parts of the city and its outlying areas attend this hospital for routine health checks. Although our sampling method was not statistically rigorous enough to reflect the entire population of the Afyon area, the number of subjects was large (1320 subjects in total). In our opinion, the results provide a rough idea of the rates of HBV and HCV infection in the Afyon area.

Our study revealed a significantly higher rate of HBsAg seropositivity in males than in females (p=0.02), but no sex difference with respect to anti-HBs seropositivity (p>0.05). These findings are similar to those that Erden and coworkers reported for Istanbul (5). This higher frequency of HBsAg positivity in males is likely explained by the fact that all healthy Turkish men are required to complete at least one period of army service. During this time, they are exposed to potential blood transmission via instruments such as scissors, razor blades, straight razors, etc. during haircutting/shaving by barbers.

We found no significant difference between males and females with respect to seroprevalence of anti-HCV (p>0.05); however, the older age group had a higher rate of anti-HCV positivity than the younger group (p=0.001). Part of the reason for this difference may be increased public awareness about viral hepatitis throughout Turkey. Efforts are being made to inform the population about risks and prevention through regular public education programs. Also, the older group had had more years of possible exposure to HCV or HBV.

We revealed a significantly higher rate of HCV seropositivity in the older versus younger age group (p=0.001). According to this result, it may be considered that immunity is weakened in the elderly and that transmission possibility of HCV is low in the older age group. But this is not clear. Because we did not investigate HCV RNA in those patients, our subjects may have been inactive HCV carriers.

In the younger age group, the rate of subjects with natural immunity to HBV was two times greater than in the older age group (63% vs 37%), which may reveal decreased immunity to HBV in the elderly.

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In our study, the rate of isolated anti-HBcIgG positivity was 6.1%. Previous reports from Turkey and other parts of the world have noted corresponding prevalence rates ranging from 0.1% to 20% (7-9). Detection of anti-HBcIgG in the absence of HBsAg and anti-HBs can signify one of at least four different situations related to HBV infection: 1) it can be years after the infection has resolved and anti-HBs has fallen below detectable levels; 2) the finding can reflect passive transfer of anti-HBC from blood donation; 3) the individual may have occult HBV infection; or 4) the results for the other HBV markers may be false-negatives (10-13). The most important of these four situations health-wise is occult HBV infection. It has been documented that such infections may play a role in transmission of the virus, and that occult HBV may be a risk factor for development of hepatocellular carcinoma. Further testing of our subjects who exhibited isolated anti-HBcIgG positivity was beyond the scope of this study. However, such investigations

<table>
<thead>
<tr>
<th>Subject grouping</th>
<th>HBeAg-positive (n)</th>
<th>HBeAg-negative (n)</th>
<th>Rate of HBeAg positivity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>4</td>
<td>50</td>
<td>7.4%</td>
</tr>
<tr>
<td>Female</td>
<td>3</td>
<td>30</td>
<td>9.1%</td>
</tr>
<tr>
<td>Younger age group</td>
<td>4</td>
<td>62</td>
<td>6.1%</td>
</tr>
<tr>
<td>Older age group</td>
<td>3</td>
<td>18</td>
<td>14.3%</td>
</tr>
<tr>
<td>Total</td>
<td>7</td>
<td>80</td>
<td>9%</td>
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are necessary in this patient group for the reasons mentioned above (12). Our analysis revealed similar rates of isolated anti-HBcIgG positivity in males and females (p>0.05), but a significantly higher rate in the older than in the younger age group (p=0.0001). In some of the positive cases in the older group, it is possible that patients had had HBV infection at a younger age and that anti-HBs levels had dropped to undetectable levels with time. This might partially explain the difference in anti-HBcIgG findings between the two age groups. Another reason could be the above-mentioned improvements in community education about viral hepatitis and preventive methods in recent years. Furthermore, there has been a significant drop in the number of consanguineous marriages in Turkey over the past years. This could be reducing the number of HBV infections because it has been reducing transmission of the virus with sexual contact and via the horizontal route.

Although the rate of isolated anti-HBcIgG positivity in the older age group was higher than in the younger age group, the rates of anti-HBs positivity were similar in the two groups. As a result, we can consider that HBsAg positivity decreases and possibility of spontaneous HBsAg seroconversion increases with age.

Our testing revealed that 33.8% of the 1320 subjects in the study (6.6% HBsAg-positive plus 27.2% anti-HBs positive) were found seropositive for HBV. There has been an HBV vaccination program for neonates in Turkey since 1998, but this has had no impact on the rate of HBV infection in our country. There is a need to add routine adolescent vaccination to the strategy, as this will achieve more rapid reduction in HBV transmission (14,15). However, due to national economic problems, only risk groups of adolescents are currently being vaccinated, and the same holds for adults. In our study, only 24% of the anti-HBs-positive subjects were vaccinated. In both age groups of anti-HBs-positive individuals, the number of subjects with natural immunity to HBV was greater than the number of vaccinated cases (Table 2). The high rate of HBV infection we observed and these immunity findings underline the seriousness of the HBV problem in Afyon. Further, we found that younger individuals comprised significantly larger proportions of both the vaccinated subgroup (p=0.0001) and the naturally immune (p=0.004) subgroup of anti-HBs-positive cases than did individuals from the older age group. As noted above, only risk groups are vaccinated against HBV in our country, so these results indicate that even with better education programs in place, adolescents and younger adults in Turkey are exposed to this virus significantly more frequently than older adults. It is clear that the HBV vaccination programs for adolescents constitute the most effective strategy for preventing the spread of this disease.

Hepatitis B e antigen is another antigen associated with HBV infection, and is encoded by the precore sequence of the HBV genome. Positivity for HBeAg reflects ongoing replication and infectivity of the virus. Seronegative status for HBeAg in a case of confirmed HBV infection indicates that the patient is infected with an HBV mutant that does not express HBeAg. Carriers of HBsAg who are infected with mutant viruses that do not synthesize HBeAg will test positive for anti-HBe. Infection with mutant HBV strains is associated with negative response to treatment (1).

In our study, 9% of the HBsAg-positive subgroup (87 patients total) were seropositive for HBeAg. Bozdayi and colleagues (16) have reported that the HBeAg-negative phenotype in Turkish patients with chronic hepatitis is associated with mutations in the precore region of the HBV genome. The Mediterranean area and Middle East and Asian countries have the highest proportions worldwide of HBeAg-negative chronic HBsAg carriers. Recent data suggest that HBeAg-negative chronic hepatitis is more common than previously suspected, and that this condition occurs worldwide (17, 18). The precise seroprevalence of HBeAg in chronic HBsAg carriers in Turkey is unknown because no study to date has involved adequate numbers of such patients. A few investigators have looked at small numbers of chronic HBsAg carriers in Turkey, and the reported seroprevalence rates of HBeAg have ranged from 33% to 67% (16, 19, 20). The rate in our study was very low, but our total number of HBsAg-positive subjects was relatively small.

This study of HBV and HCV seropositivity and the rate of isolated anti-HBcIgG in healthy subjects (adolescents through geriatric age) was conducted at a Turkish university hospital and included a fairly large study sample. Our findings for the city of Afyon, located in west-central Turkey, were 33.8% seroprevalence for HBV and 2.2% seroprevalence for HCV. These figures are similar to corresponding results from other regions of Turkey. The
rate of isolated anti-HBcIgG positivity in this study was 6.1%. Our results cannot be considered reflective of all residents of Afyon and the surrounding area; however, the large number of subjects investigated gives it validity. We believe that this study is a good starting point that will create useful comparison for future HBV and HCV seroprevalence studies performed in our region.

REFERENCES