SUMMARY

Hepatitis B virus (HBV) and hepatitis C virus (HCV) infections account for a substantial proportion of liver diseases worldwide. The aim of this study was to determine the prevalence of HBV and HCV serological markers among children and adolescents and verify the epidemiology of the HBV infection over than a decade of the introduction of vaccination program. Serologic markers to HBsAg, total anti-HBc and anti-HCV had been tested in 393 samples. The seropositivity for HBsAg was 0.76% and for total anti-HBc was 1.02%. Copositivity between HBsAg and total anti-HBc was verified in 0.76% of the analyzed samples. There was no seropositivity for anti-HCV marker. The seroprevalence of HBV infection markers among children and adolescents in the southern Brazilian region is high compared to that reported in other countries. Preventive measures, such as educational activities in addition to the universal childhood HBV vaccination, should be initiated in order to reduce the morbimortality and the economic burden associated with the disease.

KEYWORDS: Adolescents; Hepatitis B; Hepatitis C.

INTRODUCTION

Hepatitis B virus (HBV) and hepatitis C virus (HCV) infections account for a substantial proportion of liver diseases worldwide. An estimated amount of two billion people have been infected with the hepatitis B virus and over 350 million have chronic liver infections. About 25% of adults who become chronically infected during childhood die from HBV-related liver cancer or cirrhosis. HCV infections are also common worldwide. It is estimated that about 3% of the world’s population has been infected with HCV and that some 170 million are chronic carriers at risk of developing liver cirrhosis and/or liver cancer. These chronic carriers represent a reservoir sufficiently large for HCV to persist.

The hepatitis B virus carrier rate variation is 1-20% worldwide. This variation is related to differences in the route of transmission and the patient’s age at infection. In the areas of low prevalence (rate of 0.1-2%), the sexual and percutaneous transmission during adulthood are the main ways of transmission. In areas of intermediate prevalence (rate of 3-5%), the sexual, percutaneous and transmission during delivery are the major routes. In areas of high prevalence (rate of 10-20%), the predominant mode of transmission is perinatal, and the disease is transmitted vertically during early childhood from the mother to the infant.

Although HCV infection has both acute and chronic forms, most of the morbidity associated with infection is realized through the development of chronic liver disease in a subset of infected people, years after initial acquisition of the infection. Thus, a major determinant of the future burden of disease is the past and present incidence of infection. However, establishing the incidence of HCV infection is difficult because most infections are initially asymptomatic and available assays do not distinguish acute from chronic or resolved infection. Acute disease reporting systems can underestimate the incidence of HCV infection, even in countries with well-established surveillance systems.

In Brazil, the HBV vaccine was included in the National Immunization Program in 1996, but was implemented in the State of Santa Catarina in 1993 for children under five years of age and for professionals at risk. The hepatitis B immunization schedule for newborns consists of three doses (0, 1, 6 month schedule). Besides, the program includes the prevention of perinatal infection, through maternal screening and prophylaxis of newborns, HBV vaccination for all children, to prevent the infection in childhood and older, vaccination of adolescents who were not protected and individuals belonging to risk groups. One of the goals of the Brazilian Health Ministry is the immunization of young people under 19 years of age.

The HBV universal vaccination has demonstrated reduction in rates of infection. However, after the implementation of the vaccination routine, it seems useful to verify the impact of routine immunization on the epidemiology of the infection by analyzing the trend and the change of several indicators, through serum-epidemiological studies. Thus, after over than a decade of the introduction of vaccination program against
HBV in Brazil, it is necessary to verify the impact of this practice in the epidemiology of infection in the country.

Epidemiological surveillance for hepatitis C is also essential, even if there is no vaccine available. Identification of HCV-infected people is required to initiate prevention activities and reduce the risks for HCV transmission and chronic liver disease. Anti-HCV-positive individuals require further evaluation for chronic HCV infection and liver disease, and those with chronic hepatitis C require evaluation for possible antiviral therapy and the need for further medical management.

In this study, the aim is to determine the prevalence of HBV and HCV serological markers among children and adolescents and verify the epidemiology of the HBV infection over a decade of the introduction of vaccination program.

MATERIALS AND METHODS

Study design: We conducted a cross-sectional study to evaluate the prevalence of serological markers for HBV and HCV among children and adolescents.

Studied population: The amostral size was calculated based on the statistical formula \( n = \frac{4z^2_p q}{(2ME)^2} \), being \( z \) value of the normal curve in the (usually bicaudal), \( p \) = initial estimate of the proportion, \( q \) = \( 1-p \) complement (1-p) and \( ME \) = margin of error on the maximum tolerable parameter. Considering 0.5 the initial estimate of the proportion and the complement of \( p \) equal to 0.5, with prevalence of HBV and HCV markers of 50% with a 95% confidence interval and 0.05 alpha error, it was demonstrated to be necessary 384 participants. An amostral plan was designed, aiming to reproduce the distribution of the population of children and adolescents attended by the Programa de Saúde da Família in health posts in the city of Blumenau. It was evaluated about 10 to 30 volunteers in each health unit visited, being those 17 out of 40. The sample was composed of 393 children and adolescents attended at these health institutions in the period between October 2007 and August 2008. The participants were included by randomized selection.

Data processing and analysis: Inclusion criteria were age between 10 to 15 years and presentation of an informed consent signed by parents or responsible ones.

Blood samples were collected by venipuncture in the health institutions selected for the survey, to determine the serologic markers HBsAg, total anti-HBc and anti-HCV. After separation of the serum, the samples were stored at -20 °C to perform the serological tests. The analysis of blood samples was performed in the Municipal Laboratory of the city of Blumenau, Santa Catarina State, Brazil.

Vaccination data of the participants who had presented hepatitis B virus infection markers had been verified in the vaccination document. All participants who were positive for HBV infection markers had presented three doses of vaccine in the vaccination certificate.

Serologic testing: HBsAg, total anti-HBc and anti-HCV were detected by Microparticle Enzyme Immunoassay (MEIA) with commercial kits AxSym® (Abbott Diagnostics, Chicago, Illinois, USA). The MEIA is a variation of the principle of Enzyme Immunoassay (EIA) and the solid phase comprises microparticles that increase the sensitivity of the method. This solid phase EIA uses the antigens and/or antibody adsorbed on the surface for the binding of complementary analytes. The bound analyte is detected by a number of antibody-antigen reactions. The EIA that qualitatively detects the surface antigen of hepatitis B virus (HBsAg) in human serum or plasma uses microparticles coated with anti-HB monoclonal antibodies. In the same way, those who qualitatively detect the total anti-HBc and anti-HCV recombinant antigens have adsorbed the microparticles in the solid phase.

Ethical approval: This study was approved by the Ethics Committee in Human Research at the Federal University of Santa Catarina, in the Protocol n. 238/07, and approved by the Health Secretary in Blumenau.

RESULTS

The population was composed of 393 children and adolescents. Among the participants, 210 were female (53.44%) and 183 were male (46.56%). The age range was 10 - 15 years, and the mean age was 12.5 years (± 1.7). The most frequent age group was between 12 and 13 years (40.20%). Table 1 refers to the general characterization of the sample, with the population distribution according to sex and age.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Population</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>210 (53.44%)</td>
</tr>
<tr>
<td>Male</td>
<td>183 (46.56%)</td>
</tr>
<tr>
<td>Age (years)</td>
<td></td>
</tr>
<tr>
<td>10-11</td>
<td>105 (26.72%)</td>
</tr>
<tr>
<td>12-13</td>
<td>158 (40.20%)</td>
</tr>
<tr>
<td>14-15</td>
<td>130 (33.08%)</td>
</tr>
</tbody>
</table>

The seropositivity for HBsAg was found in 0.76% of the samples (3/393) and total anti-HBc was 1.02% (4/393). Copositivity between HBsAg and total anti-HBc was detected in 0.76% (3/393). The seropositivity for HBV, which means positive samples that reacted with one or two tested markers (HBsAg and total anti-HBc), was 1.02% (4/393) (Table 2). In the evaluation of the 393 samples for anti-HCV, no case of positivity for the antibody was found.

<table>
<thead>
<tr>
<th>Serologic marker</th>
<th>Positive (n)</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total anti-HBc</td>
<td>1</td>
<td>0.25%</td>
</tr>
<tr>
<td>Total anti-HBc + HBsAg</td>
<td>3</td>
<td>0.76%</td>
</tr>
<tr>
<td>No marker</td>
<td>389</td>
<td>98.98%</td>
</tr>
</tbody>
</table>

All volunteers that were positive for HBsAg marker received three doses of hepatitis B vaccine.
DISCUSSION

Chronic HBV and HCV infections are the most important causes of hepatocellular carcinoma (HCC), the fifth most common solid tumor in the world. The incidence and prevalence of HCC is rising in the United States and in Europe, and costs of care in HCC will continue to rise\(^1\). In the United States, the total economic burden of HCC in 2005 was estimated to be $454.5 million\(^2\). The development of more effective and cost-effective treatments for viral hepatitis and related complications is a challenge for the medical community\(^3\). Therefore, interventions to reduce the prevalence of HBV and HCV infections may yield substantial economic benefits.

The seroprevalence of anti-HCV among children and adolescents found in this study is in agreement with that observed in Spain\(^4\), and lower than that reported in the United States\(^5,6\), in Germany\(^7\) and in Pakistan\(^8\). The results of our study show a low circulation of the hepatitis C virus among the studied population, possibly due to the low exposure to risk factors for HCV infection. The risk factors most frequently cited as accounting for HCV transmission worldwide are injecting drug use and blood transfusions\(^9,10\).

There is a lack of data about HCV infection in pediatric and adolescent population in Brazil. Anti-HCV seroprevalence reports in Blumenau are available among people submitted to blood testing in a laboratory (1.6%)\(^11\) and blood donors (0.59%)\(^12\). Studies among blood donors revealed an overall confirmed anti-HCV seroprevalence of 0.35% in Santa Catarina\(^13\), 0.9% in Rio de Janeiro\(^14\) and 1.2% in Ribeirão Preto\(^15\). In a multi-center study of HCV seroprevalence among first time Brazilian blood donors, the frequency of anti-HCV was 0.97% in women and 0.38% in men\(^16\). The results of our study show a low circulation of the hepatitis C virus among the studied population, possibly due to the low exposure to risk factors for HCV infection. The risk factors most frequently cited as accounting for HCV transmission worldwide are injecting drug use and blood transfusions\(^9,11,12\).

The prevalence for HBsAg and for total anti-HBc reported here was higher than that found among adolescents in Portugal\(^17\) and among children and adolescents under 15 years of age in Saudi Arabia\(^18\). The prevalence of these markers reflects a higher exposure to risk factors for HBV infection in the studied population. Moreover, low educational level, low socio-economic status and perinatal transmission may also be associated with the higher prevalence. However, hepatitis B infection is more commonly transmitted in children through the horizontal route\(^1,13,19\).

Although the vaccination is the most effective strategy to prevent HBV infection, in our study half of non-infected participants showed antibodies to hepatitis B surface antigen below 10 mIU/mL (data not shown), and we found three cases of HBsAg in vaccinated individuals. These individuals may not have received the hepatitis B vaccine at birth and may have been exposed to the virus before vaccination. Furthermore, some other hypothesis such as mutations in the virus surface antigen\(^18\) and intraterine transmission of HBV from mothers to the fetus\(^20\) have been proposed to explain the HBV infection in vaccinated individuals. In this study, transmission of HBV to infants born from HBV carrier mothers cannot be ruled out. However, HBsAg status of mothers was not established. In addition, another limitation is the lack of virological data investigating possible vaccine escape mutants.

In the present study, the prevalence for anti-HBc alone was 0.25%. Worldwide, the prevalence of isolated anti-HBc in different populations ranged from 0.1 to 20%\(^3,17,26,41\). In Taiwan, one study with 1200 children aged seven years with complete HBV immunization in infancy showed that eleven children had new HBV infections with anti-HBc positivity as the only marker, and none became positive for HBsAg or had detectable HBV DNA by polymerase chain reaction. The percentage of protective anti-HBs in children without booster vaccination decreased from 71.1% at age seven years to 37.4% at age 12 years, but only one of 200 children who received a booster dose and two of 258 children who did not received had developed new anti-HBc positivity\(^42\). National vaccination program has decreased the spread of anti-HBc alone in this country\(^22\). In a 10-year follow-up study conducted in a community with a high spread of HBV infection, 0.8% had developed anti-HBc yet none showed a clinically significant breakthrough infection\(^43\). Another study showed that three out of 19,000 individuals who had developed HCC were HBsAg negative but were anti-HBc positive. This might suggest that there is a very small risk of HCC among those who have anti-HBc as the only indication of a previous infection\(^44\).

The present study showed that seropositivity for HBV infection among children and adolescents from Blumenau was lower than that recorded among people submitted to blood testing in a laboratory (38.5%)\(^21\) and blood donors (9.2%)\(^45\) in the same city. The overall prevalence rate of HBV infection in the studied population was also smaller than that found among blood donors in Santa Catarina (5.35%)\(^19\), Rio de Janeiro (3.68%)\(^22\), Acre (5.8%)\(^23\) and Ribeirão Preto (8.7%)\(^24\). Other authors have reported higher prevalence of HBV markers in older ages. In a rural area in the Amazon Region of Mato Grosso State, the overall seroprevalence of HBV markers was 40.0%\(^25\). The prevalence of serological markers of HBV infection was 11.1% in healthcare workers in Mato Grosso do Sul State\(^26\), and 24.1% among laboratory staff in Goiânia\(^27\). One study carried out among dentists from Campo Grande, Mato Grosso do Sul, showed 10.8% of seropositivity for HBV infection\(^28\).

The higher prevalence of HBV markers in older ages could be due to the greater number of years of potential exposure and the lack of awareness of HBV infection in earlier decades\(^11\). In addition, the vaccination may have contributed to this lower prevalence among children and adolescents aged 10-15 years. The low prevalence of HBV infection markers indicates the positive impact of routine infant hepatitis B vaccination, which was included in the National Immunization Program in 1996\(^29\), in the prevention of transmission in the studied population.

In conclusion, the prevalence of HBV infection was higher than HCV infection in the studied population. Until a safe and effective vaccine against HCV is available, information and educational campaigns regarding modes of transmission are the main preventive tools to reduce the burden of HCV infection and HCV-related disease. The data from our study show that the seroprevalence of HBV infection markers among children and adolescents in the southern Brazilian region, over than a decade of the introduction of vaccination program, is higher when compared to the other countries. This result reinforces the importance of HBV vaccination initiated at birth, to prevent perinatal HBV transmission, and the necessity of preventive measures including educational activities in addition to the universal childhood HBV vaccination, to further reduce the morbimortality and the economic impact associated with the disease.
RESUMO

Soroprevalência dos marcadores de infecção das hepatites B e C em crianças e adolescentes da região sul do Brasil

As infecções pelo vírus da hepatite B (HBV) e pelo vírus da hepatite C (HCV) representam a causa de uma substancial proporção das doenças hepáticas em todo o mundo. O objetivo deste estudo foi determinar a prevalência dos marcadores sorológicos do HBV e do HCV em crianças e adolescentes, e verificar a epidemiologia da infecção pelo HBV passada uma década desde a introdução do programa de vacinação. Os marcadores sorológicos HBsAg, anti-HBc total e anti-HCV foram avaliados em 393 amostras. A soropositividade para o HBsAg foi de 0,76% e para o anti-HBc total foi de 1,02%. Co-soropositividade entre HBsAg e anti-HBc total foi verificada em 0,76% das amostras analisadas. Não houve soropositividade para o marcador anti-HCV. A soroprevalência dos marcadores de infecção pelo HBV em crianças e adolescentes da região sul do Brasil é elevada em relação à reportada em outros países. Medidas preventivas, tais como atividades educativas além da vacinação infantil contra o HBV, devem ser iniciadas a fim de reduzir a morbimortalidade e o impacto econômico associados a doença.

ACKNOWLEDGMENTS

We thank all the volunteers who participated in this study, the Health Secretary of Blumenau and health workers.

REFERENCES


Received: 12 February 2010
Accepted: 27 October 2010