

## DETECTING OCCULT HEPATITIS B WHEN TESTING DONATED BLOOD

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Individuals carrying occult (latent) hepatitis B pose epidemiological threat. Testing donated blood donors for surface antigen HBsAg (hepatitis B virus, HBV) only does not allow to assume the blood safe from the point of view of infections, which can result in post-transfusion transmission of infection. Lack of confidence here is due to the fact that the virus is present in the body even when HBsAg is negative. The study analyzes data of 61,155 blood donors of the Republican Blood Center (Kazan), collected in 2010–2014. The tests applied were those aimed at detecting HBsAg, anti-HBc-total, anti-HBc IgM (enzyme immunoassay), and determining DNA of the virus in the blood by polymerase chain reaction in "real time". It was found that donors with occult hepatitis B are identified each year, but their numbers decrease gradually. To prevent the spread of the virus it is recommended to add the anti-HBc-total test to the standard set of diagnostic tests.

**Keywords:** occult hepatitis B, latent hepatitis B, HBV infection, donor, blood, enzyme immunoassay, PCR diagnostics, HBsAg, anti-HBc-total, anti-HBc IgM

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## ВЫЯВЛЕНИЕ ОККУЛЬТНОГО ГЕПАТИТА В ПРИ ТЕСТИРОВАНИИ ДОНОРСКОЙ КРОВИ

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Эпидемиологическую опасность представляют лица с оккультным (латентным) гепатитом В. Исследование крови доноров только на поверхностный антиген вируса (hepatitis B virus, HBV) HBsAg не может обеспечить полную инфекционную безопасность крови, следствием чего может быть посттрансфузионная передача инфекции. Это объясняется тем, что вирус присутствует в организме даже при отрицательном HBsAg. В работе проанализированы данные 61 155 доноров Республиканского центра крови (Казань), полученные в 2010–2014 гг. Анализировали результаты тестирования на HBsAg, anti-HBc-total, anti-HBc IgM (методом иммуноферментного анализа) и определения ДНК вируса в крови (методом полимеразной цепной реакции в «реальном времени»). Установлено, что доноры с оккультным гепатитом В выявляются ежегодно, хотя и отмечена тенденция к снижению их числа. Для предотвращения распространения вируса в популяции рекомендуется ввести в стандарт диагностики HBV-инфекции определение маркера anti-HBc-total.

**Ключевые слова:** оккультный гепатит В, латентный гепатит В, HBV-инфекция, донор, кровь, иммуноферментный анализ, ПЦР-диагностика, HBsAg, anti-HBc-total, anti-HBc IgM

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To avoid transmission of bloodborne infections from donor to recipient, it is imperative to ensure infectious safety of donated blood. Screening for hepatitis B (hepatitis B virus, HBV) helped to significantly reduce incidence of this infection [1]. However, despite availability of sensitive tests detecting HBV surface antigen (HBsAg), post-transfusion hepatitis B still occurs [2, 3]. This is due to the fact that the virus can lay dormant in liver tissue for a long time, and in some cases even in blood of HBsAg-negative patients [4, 5]. Therefore, occult (latent) hepatitis B is a risk factor in blood donation and organ transplantation [6]. There are documented cases of HBV DNA found in recipients that received Hbs-negative blood from donors who had occult

HBV. Thus, there is a real threat of spread of latent hepatitis B and its further activation in human body when the immunity is suppressed [5].

The goal of our study was detection of latent HBV at the stage of donated blood testing.

### METHODS

The study was conducted at the Republican Blood Center (RBC) of the city of Kazan. Data obtained from testing 61,155 donors in 2010–2014 were analyzed, namely: HBsAg tests, anti-HBc-total tests, anti-HBc IgM tests, HBV DNA tests.

HBsAg, anti-HBc-total and anti-HBc IgM were identified by ELISA using commercial reagents made by Vector-Best (Russia), sensitivity of 0,01 ME/ml. HBV DNA was detected by polymerase chain reaction in real time using COBAS S 201 (Roshe, Switzerland) device and a set of Sobas TaqScreen MPX Test reagents, v2.0 and CobasTaqScreen MPX ControlKit, v.2.0 (Roche).

## RESULTS

HBsAg is the screening marker for HBV-infection. Detection of this marker when testing donated blood confirms presence of active infection. Individuals with HBsAg are dismissed as donors and receive applicable treatment. In 2010–2014, Kazan's RBC registered 6 to 25 cases of marker detection per year (Fig. 1), the trend is downward.

Anti-HBc-total tests returned positive in 3.5–6.9 % of cases a year, which means that blood from 400 to 900 donors could not be used (Fig. 2). When this marker is detected, the blood is subject to anti-HBc IgM. In 2010–2014, M-class immunoglobulins were detected in 2.5–9.9 % of donated blood samples, with signs of latent HBV in donors' bodies (anti-HBc-total positive samples) (Fig. 3). Anti-HBc IgM is the viral activity market that signals the need for detailed medical examination of the person.

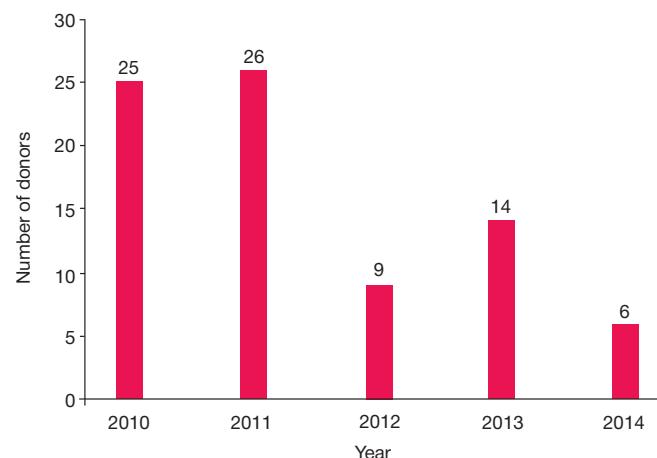


Fig. 1. Positive screening tests (HBsAg) statistics, 2010–2014

HBV DNA in blood serum is the main indicator of viral replication. Approximately 20 % of cases require solely DNA diagnostics to reveal latent HBV since no other tests are capable of detecting it [7]. At RBC, PCR is performed in real time to confirm absence of infection in donors that showed negative results for classical markers. This extra blood test also helps to detect active forms of HBV in cases of latent hepatitis B (Fig. 4).

## DISCUSSION

In Russia, far from all medical institutions test donated blood for antibodies to hepatitis B (using anti-HBc-total or anti-HBc IgM tests), which hinders timely detection of latent forms of HBV. Our findings allow concluding that absence of HBsAg does not guarantee infectious safety of donated blood, since a positive anti-HBc-total test indicates previous exposure to the virus and a positive anti-HBc IgM test signals an active infection.

HBsAg-negative phase can show low level of virus DNA in the body: it is detected in liver and rarely in serum [8]. That is why this phase does not mean full recovery, since a relapse can occur any moment [9, 10]. Patients with latent HBV were found to maintain effective immunological response (anti-HBc total), which can lead to reactivation of HBV and HBsAg reappearance [11].

The complications of detection of occult hepatitis B arise from non-mandatory character of tests for markers of latent HBV

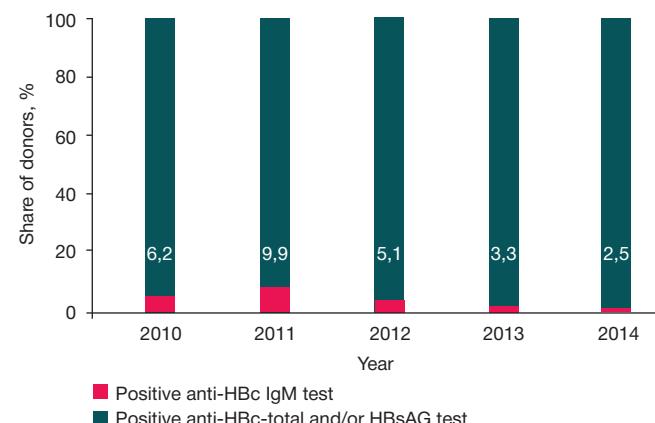


Fig. 2. Positive anti-HBc-total tests, 2010–2014

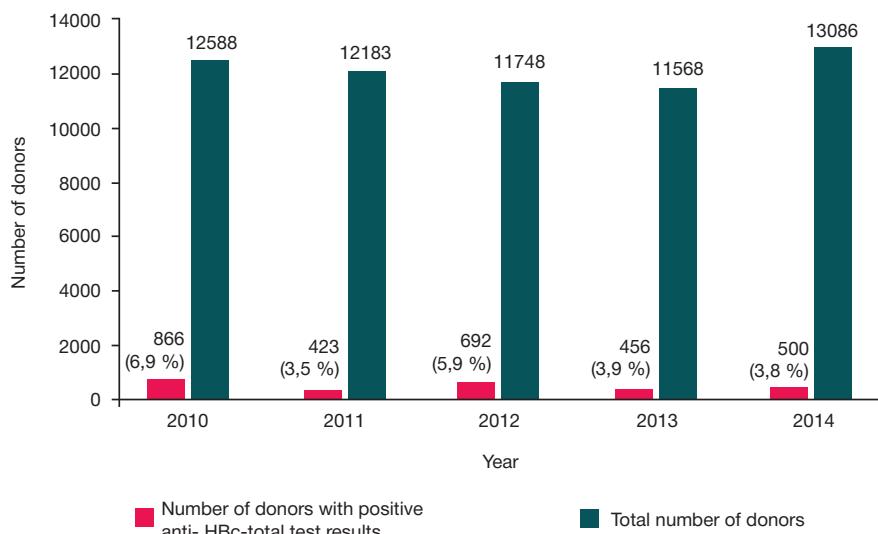
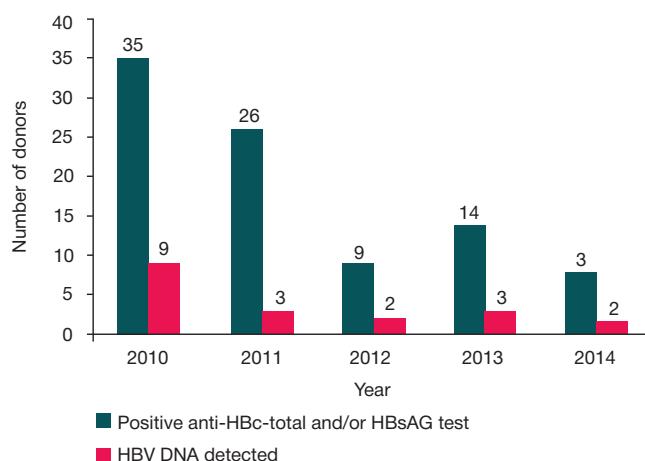


Fig. 3. Share of donors with M-class immunoglobulin in blood, donors with positive anti-HBc-total and/or HBsAg tests, 2010–2014



**Fig. 4.** Number of HBV DNA detection occurrences, donors with positive HBsAg and/or anti-HBc-total tests, 2010–2014

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and further examination of the donor. Disease control center only receives information about donors with positive HBsAg tests. Timely implementation of preventive and anti-epidemic measures requires further medical examination and outpatient observation of individuals with anti-HBc-total and anti-HBc IgM in blood. Otherwise, use of blood from donors with occult hepatitis B increases the risk of transfusion transmission of HBV and its spread in the population [12, 13].

## CONCLUSIONS

Donors with positive tests for HBV markers (HBsAg, anti-HBc-total, anti-HBc IgM, HBV DNA) are found every year. Introduction of anti-HBc-total test to the standard set of HBV diagnostic tests will increase the rate of detection of latent HBV [14, 15]. Setting up seamless workflow and cooperation between blood donation centers and other medical institutions will ensure early detection of active forms of infection and prevention of chronic process, and also help prevent infection of others.

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