Abstract

Background
Viral hepatitis is a major global health problem affecting millions of people worldwide. The main objective of the present study was to estimate the seroprevalence of hepatitis C and hepatitis B in Saudi adults undertaking premarital screening voluntarily.

Methodology
This observational retrospective study was conducted at King Khaled General Hospital, Al Majmaah, Saudi Arabia, for a period of 3 years (October 1, 2016 to September 30, 2019). The prevalence of hepatitis B and hepatitis C infections was calculated using Microsoft Excel and the hematological data analysis was performed using SPSS software. A total of 3755 premarital Saudi adults who voluntarily participated in the premarital screening were included in this study. Subjects were screened for hepatitis C virus (HCV) and hepatitis B virus (HBV). The demographic and epidemiological parameters included patient age, sex, nationality, and seropositivity for HBV and/or HCV.

Results
Of the 3755 subjects, eight (0.24%) and 12 (0.36%) subjects were positive for HCV and HBV, respectively. Among the 12 HBV-positive subjects, nine (75%) subjects were men, and three (25%) subjects were women. However, among the eight HCV-positive subjects, five (62.5%) subjects were men, and three (37.5%) subjects were women.

Conclusion
This study concluded that the prevalence of HBV infection was slightly higher than that of HCV infection among the Saudi adult population of Al Mjamaah. In addition, both HBV and HCV were less prevalent in women than in men in the study population.

Keywords: Seroprevalence, Hepatitis, Hepatitis C virus, Hepatitis B virus, Infection

Introduction
Viral hepatitis being a major global health problem affects approximately 400 million people globally and is responsible for about 1.4 million deaths each year, most (95%) of which are due to hepatitis B and hepatitis C. Viral hepatitis is caused by five different types of viruses, including hepatitis virus A, B, C, D and E. The hepatitis B virus (HBV) and hepatitis C virus (HCV) can cause chronic hepatitis. HBV and HCV are transmitted through exposure to infected blood and/or semen. More than 95% of the hepatitis B infected adults resolve the infection during the acute phase. However, about 80% of the patients with hepatitis C progress to chronic infection. Viral hepatitis is mostly diagnosed by testing the virus specific serological markers. The diagnosis of HBV infection requires the detection of few serological markers, including anti-HBs/anti-HBe antibodies, and HBsAg. HBsAg is the serological hallmark of infection as it can be detected within 1–10 weeks post-infection and its persistence for >6 months is indicative of chronic infection. HCV screening is generally performed using anti-HCV antibodies and is occasionally confirmed by HCV RNA test.

Hepatitis B is the most widespread viral hepatitis, with approximately 57 million people infected with HBV worldwide. Its prevalence is higher in the Middle East than in Europe and the United States. Its prevalence ranges from 0.6% in Iraq to >8% in Sudan. The number of HBV-infected persons in the Eastern Mediterranean region is >21 million (3.3% of the population). Hepatitis C affects approximately 3% of the world population; however, it has a high prevalence of approximately 10% in developing countries. In 2007, viral hepatitis was ranked as the second most common viral disease in Saudi Arabia. Previously, in 1998, Saudi Arabia witnessed a high prevalence (8.3%) of hepatitis B. In a community based epidemiological study HBsAg seroprevalence of approximately 7% was reported in Saudi children. These reports of high prevalence of HBV led to state and scientific intervention, and as a result, the universal HBV immunization was started in the country in 1989. The immunization program successfully resulted in a decrease in the prevalence of HBV in the country. Data on the prevalence of HCV in Saudi Arabia are insufficient and controversial. The premarital screening program estimates the prevalence of HCV to be 0.33%, while screening centers for blood donors indicate HCV infection rates of 0.4%–1.1%. In addition, HCV prevalence of 1.0%–1.9% has been reported in a systematic review that included most of the published reports on the subject.

The insufficiency of confirmed data leads to diminished planning for the prevention and treatment strategies of the disease. Hence, there is a need for considerable community-based epidemiological studies to obtain more accurate incidence rate of HCV and HBV infection in Saudi Arabia.
Therefore, the present study was performed to estimate the prevalence of HBV and HCV infection among adult population of Al Majmaah, Saudi Arabia. To accomplish this study, adults who attended the health service at King Khaled General Hospital (KKGH) for premarital screening were included and examined. The study population included adult men and women of Saudi nationality from Al Majmaah. In addition, the effects of HBV and HCV seropositivity on hematological parameters were also evaluated.

Materials and Methods

Source/study population

The present study was conducted at the premarital screening center of KKGH and involved 3755 adult Saudi people from the Al Majmaah region (1953 men and 1802 women, aged 20–55 years). The 3-year clinical data (October 1, 2016 to September 30, 2019) were analyzed using a statistical analysis system. The study was approved by Central Institutional Review Board, Ministry of Health, Saudi Arabia (IRB No. 2019-0126E).

Saudi nationals visiting the health service for premarital screening with no recent illness, surgery, or transfusion of blood or blood components were included in the study. Individuals who underwent premarital screening with a recent history of health issues, major surgery, or transfusion of blood and/or blood components in the previous 6 months were excluded from the study.

Blood sampling

Fresh whole blood (5 mL) was collected from all the study subjects and divided into two parts; one part was transferred to a tube containing ethylene diamine tetra acetic acid (EDTA), mixed gently and stored at ambient temperature and analyzed for hematological parameters within 48h of collection. The other part was transferred into a plain tube (without any anticoagulant) and allowed to clot by leaving it undisturbed at room temperature for 45–60 min. The clotted blood samples were centrifuged for 10 min at 1000g in a refrigerated centrifuge and the resulting supernatant (serum) from each sample was collected into separately labelled clean and sterile tubes and stored at –20°C until analysis.

Serological tests for HCV and HBV

HCV and HBV serological markers were identified and evaluated using the ARCHITECT Immunoassay System (Abbott, Abbott Park, Illinois, USA) according to the manufacturer's instructions. The detection of HBV surface antigen (HBsAg) and anti-HCV antibodies was performed using HBsAg ULTRA kit (Bio-Rad, France) and anti-HCV PLUS version 3 kit (Bio-Rad, France), respectively. The initial screening results were confirmed by retesting positive samples.18

Hematological analysis

Hematological tests for healthy normal individuals and HBV- and HCV-positive individuals were performed using an automated hematometry analyzer, CELL-Dyn Ruby (Abbott Laboratories, Diagnostic Division, Abbott Park, Illinois, USA)

Statistical Analysis

Statistical analysis for estimating the prevalence of HBV and HCV was performed using Microsoft Excel. The hematological data was analyzed by one-way ANOVA test using SPSS software. The results were considered statistically significant if p values were <0.05.

Results

Seroprevalence of HBV and HCV

In total, 3755 participants (1953 [52.01%] men and 1802 [47.99%] women) were recruited from October 1, 2016 to September 30, 2019. All participants were Saudi nationals aged 20–55 years. All participants underwent premarital screening and were examined for HBsAg and anti-HCV serum markers. As shown in Table 1, 0.32% (n = 12) and 0.21% (n = 8) subjects were positive for HBsAg and anti-HCV, respectively, and 99.47% (n = 3735) subjects were negative for both the serum markers tested. The yearly distribution of HBV- and HCV-positive cases in the study population is shown in Figure 1.

On further analysis of our data and results, we observed that among the 1953 men, 99.28% (n = 1939) were seronegative, 0.26% (n = 5) were positive for HCV, and 0.46% (n = 9) were positive for HBV (Table 2). Among the 1802 women, 99.67% (n = 1796) were seronegative, 0.21% (n = 8) were positive for HBsAg and anti-HCV, respectively, and 99.47% (n = 3735) subjects were negative for both the serum markers tested. The yearly distribution of HBV- and HCV-positive cases in the study population is shown in Figure 1.

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Table 1: Prevalence of viral hepatitis among 3755 participants undergoing premarital screening in Al Majmaah, Saudi Arabia (from October 1, 2016 to September 30, 2019)

| Category                | No. of cases (n) | Percentage (%) |
|-------------------------|------------------|----------------|
| HBV positive            | 12               | 0.32           |
| HCV and HBV Negative    | 3735             | 99.47          |
| Total                   | 3757             | 100            |

Table 2: Combined results of HCV and HBV positive male and female cases/groups.

| Total study population (N=3755) | HBV +ive cases (n=12) | HCV +ive cases (n=8) |
|-------------------------------|-----------------------|----------------------|
| Male, n (%)                  | Female, n (%)         | Male, n (%)         | Female, n (%) |
| 1 9 5                         | 1 8 0                 | 3 0 (0.46)         | 3 0 (0.17)   |
| (52.01)                      | (47.99)               | 5 (0.26)           | 3 (0.17)    |
| >30 yrs = 7                  | >30 yrs = 1           | >30 yrs = 4        | >30 yrs = 1 |
| <30 yrs = 2                  | <30 yrs = 2           | <30 yrs = 1        | <30 yrs = 2 |

Table 3: Comparison of hematological parameters between healthy controls, HBV positive and HCV positive individuals.

| Hematologic parameters | Healthy controls | HBV positive | HCV positive |
|------------------------|------------------|--------------|--------------|
| RBC (10^6/mm^3)        | 5.121 ± 0.24     | 4.95 ± 0.45  | 4.86 ± 0.45  |
| HB (g/dL)              | 14.912 ± 0.52    | 14.69 ± 0.88 | 14.36 ± 1.12 |
| HCT (%)                | 45.463 ± 4.09    | 45.98 ± 3.09 | 42.94 ± 2.8  |
| MCV (fL)               | 86.556 ± 3.66    | 89.55 ± 4.31 | 88.62 ± 4.7  |
| MCH (pg)               | 29.273 ± 0.75    | 30.47 ± 0.93 *** | 29.56 ± 0.41 |
| MCHC (g/dL)            | 34.58 ± 0.86     | 35.26 ± 0.74  | 34.58 ± 0.43 |
| RDW                    | 12.844 ± 0.86    | 12.13 ± 0.51 * | 12.16 ± 0.56 |

Values are shown as Mean ± SD. Hb: Hemoglobin, RBC: Red blood cell, HCT: hematocrit, MCH: mean cell hemoglobin, MCHC: mean cell hemoglobin concentration, MCV: mean cell volume, RBC: red blood cell, RDW: red cell distribution width.

* Significant difference: * p<0.05 and *** p<0.001

Discussion

Premarital screening is particularly important because it can prevent diseases in the future generations and provide insight into the incidence of several diseases in the adult population\(^1\). Premarital screening is mandatory in Saudi Arabia and provides an excellent platform to estimate the prevalence of several diseases\(^2\). In Saudi Arabia, consanguineous marriages are the main factor for births with a high risk of carrying a disease, especially viral hepatitis and hemoglobinopathies\(^3\). As there is a considerable incidence of hepatitis in Saudi Arabia, it is recommended to acquire data on the prevalence of the disease for planning/improving its prevention and treatment strategies. However, the relevant studies reported in the literature are insufficient to acquire complete data. In this regard, we have previously reported the prevalence of various transfusion-transmitted diseases, including HCV and HBV, in blood donors of Al Majmaah, Saudi Arabia\(^4\). In the present study, we have reported the prevalence of HCV and HBV in adult persons who underwent premarital screening at KKGH, Al Majmaah. The study results showed that seroprevalence of HBV and HCV was higher in men (0.46% and 0.26%, respectively) than in women (0.17% and 0.17%, respectively). The prevalence of HBV and HCV infection in couples tested through the premarital screening program in a previous similar study was 1.51% and 0.33%, respectively, which is slightly higher than that reported in our results\(^5\). Furthermore, a study conducted in the Qassim region showed that the positivity for HBV was 0.7%, 1.5%, and 2.04% in 2008, 2009, and 2010, respectively. The same study also reported the prevalence of HCV to be 0.1%, 3%, and 0.83% in 2008, 2009, and 2010, respectively\(^6\).

The differences in the prevalence rates might be due to regional differences, as the studies were performed in different regions/cities of Saudi Arabia. Furthermore, in these studies, the prevalence of HBV infection was higher than that of HCV infection, which is consistent with the results of our study; however, the prevalence values reported in our study are lower than those reported in previous studies. Several studies have reported a decrease in the incidence and prevalence of viral hepatitis B and C in Saudi Arabia over time\(^7,8,9\). Correspondingly, our results also revealed a lower prevalence of HCV and HBV infection in the adult population. This decline in the prevalence of HBV and HCV may be due to many factors, including timely active vaccination programs, mandatory premarital screening, and complementary immunological and therapeutic approaches that started in Saudi Arabia in 1989.

In agreement with previous studies, our results confirmed that HBV infection was more prevalent than HCV infection in our study population. Further analysis of seropositive results showed that the prevalence of HBV and HCV infection was lower in women than in men, consistent with previous studies.

Among the nine HBV-positive men, two were <30 years old and seven were >30 years old. However, in three HBV-positive women, one was >30 years old, and two were <30 years old (Table 2). Similarly, among the five HCV-positive men, four were >30 years old and one was <30 years old, and among the three HCV-positive women, two were <30 years old and one was >30 years old (Table 2). However, no cases of HBV/HCV co-infection were identified in this study.

**Hematological findings**

As shown in Table 3, the analyzed hematological parameters, including hemoglobin levels, total RBC count, hematocrit, mean cell volume and mean cell hemoglobin concentration, did not show any statistically significant differences between HBV- or HCV-seropositive and seronegative individuals. However, a statistically significant difference was observed in the values of mean cell hemoglobin (MCH) and red cell distribution width (RDW) of the HBV-seropositive subjects compared to those of the seronegative ones (p<0.005).

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with previous studies conducted in the Riyadh region\textsuperscript{24,25}. Hematological analysis revealed that the slight differences in the hematological parameter between the HBV- or HCV-seropositive subjects and the seronegative individuals were statistically insignificant, except for MCH and RDW, that too in case of HBV only (Table 3). Some of our hematological findings are in agreement with those of Ahmad et al (2018) and Sabry et al (2007)\textsuperscript{26,27}, but not with some other findings available in literature\textsuperscript{28,29}. Our main focus was to estimate the prevalence of HBV and HBC, and the small sample size of HBV- and HCV-seropositive groups could be the reason for the some controversial results.

Conclusions
This study reports the prevalence of HCV and HBV infection in the adult population of Al Majmaah, emphasizing a higher prevalence of HBV infection than that of HCV infection and a higher prevalence of both diseases in men than in women.

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Ethical Approval
All procedures in this study were performed in accordance with the ethical standards of the Central Institutional Review Board (IRB) of the Ministry of Health, Saudi Arabia (central IRB log number 2019-0126E).

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Conflicts of interest
The authors have no conflicts of interest.

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