

An evaluation and comparison of HBV among hospital patients in the province of Babylon

Hider M. H. Al-Shirifi ^{1,*}, Hiba Jasim Hamza ², Zahraa Adnan Faraj ³ and Abbas K. Al-Mansoori ⁴

¹ Department of Environmental health. Environmental Sciences College, Al-Qasim Green University, Iraq.

² Department of Medical and Basic Sciences. College of Nursing, University of Babylon, Iraq.

³ Department of Biotechnology, Faculty of Biotechnology, Al-Qasim Green University, Iraq.

⁴ Department of Genetic Engineering, Faculty of Biotechnology, Al-Qasim Green University, Iraq.

International Journal of Science and Technology Research Archive, 2023, 05(01), 001–006

Publication history: Received on 13 May 2023; revised on 27 June 2023; accepted on 30 June 2023

Article DOI: <https://doi.org/10.53771/ijstra.2023.5.1.0065>

Abstract

In Babylon Governorate, the frequency of viral hepatitis B in the general population was discussed. A total of 354 members of the general community with ages ranging from 15 to 45 and a mean of 38.64 years old were randomly selected for the study and tested for anti-HBsAg. Since fourteen samples had positive Anti-HBc IgM results and patients older than 45 had a higher risk of infection (5.6%), the findings indicated that the prevalence of HBV in the general population was (3.95%). Males were more likely to be infected than females (10:4 vs. (71:4:28.6), respectively, according to sex. The rate of infection was 2:1 for the total of (14) positive anti-HBsAg, of which (9) were in rural areas and (5) were in urban areas. Compared to anti-HBsAg, which was obtained by an ELISA test, the diagnostic marker anti-HBc IgM was more consistent with the viral load/Ml of HBV determined by RT-PCR.

Keywords: HBsAg; HBcIgM; ELISA; Minividas; RT-PCR

1 Introduction

The Latin word hepatitis refers to hepatic inflammation (Mehwish et al., 2011). Acute hepatitis is a brief illness that resolves fast, but chronic hepatitis results in long-term illness. The severity of hepatitis relies on a number of variables, including the cause of the liver injury and any underlying disorders in the body (Jou and Muir, 2008). In certain cases, it may result in liver damage, liver failure, or even liver cancer. About 600 000 individuals each year pass away from the effects of hepatitis B, which has infected two billion people globally (WHO, 2012). Anti-HBsAg prevalence differs between the region's nations: 4%–5% in Iraq, 3%–11% in Egypt, 2.6%–10% in Jordan, 2%–6% in the Libyan Arab Jamahiriya, 2.3%–10% in Oman, 5%–6% in Palestine, 7.4%–17% in Saudi Arabia, 16%–20% in Sudan, 6.5% in Tunisia, 2%–5% in United Arab Emirates and 12.7%–18.5% in Yemen (Wasfi and Sadek, 2011).

It is about 3200 nucleotides long and possesses a circular shape of partly double-stranded DNA (Thomas et al., 2005). The entire HBV virion with infectivity can be seen under electron microscopy as a 42–45 nm long spherical form known as a "Dane particle" (Lee and Ahn, 2011). The shells of the virus are composed of two layers. The outer shell is composed of the envelope protein known as HBs protein, which is further subdivided into small, middle, and large HBs proteins (SHBs, MHBs, and LHs proteins, respectively). The inner shell is composed of a core protein known as the HBc protein, which encloses viral polymerase as well as the HBV genome (Ganem and Prince, 2004). This study's objectives are: Assessment of various HBV detection methods, including ELISA, Minivida, and real-time. Examine the prevalence of HBV in the province of Babylon.

* Corresponding author: Dr. Hider, M. H. Al-Shirifi

2 Material and methods

2.1 Distribution of the samples

(354) general population samples were gathered from the central public health laboratory, blood bank, Hilla teaching hospital, and Marjan teaching hospital in Babylon province (age range 6-64 years, mean age 38.64 years).

2.2 Blood collection

Each participant's venous blood was drawn between the hours of 9 and 12 in the public health laboratory. For plasma collection, one ml of anticoagulant (EDTA) was combined. The remaining samples were used to collect serum, which is spun at 1000 rpm for 5–10 minutes to separate it, and then it is chilled or frozen at -20 until it is utilised for the appropriate test. Acon-USA's third-generation enzyme immunoassay kit (EIA-3) was used to determine anti-HBsAg levels. The automated VIDAS system's enzyme-linked fluorescence immunoassay (ELFA) (Minividas Kit/HbsAg, Biomerieux-France) was used to confirm the anti-HBsAg reactivity. The viral load was then determined by RT-PCR utilising the Exiprep™ viral DNA/RNA kit (Bioneer-Koria) to extract the virus DNA and the RT-PCR Amplification kit (Sacace-Italia) to amplify the virus' DNA.

3 Results and discussion

HBV infections start when the immune response that usually clears the virus does not occur or is insufficiently strong to be effective; as a result, infections are more prevalent among people with low immunity due to poverty. (1997, Hoofnagle). According to table 1, the current study found that the prevalence of anti-HBsAg in the general population was (3.95%), with the age group (45) having a greater prevalence of infection (5.6%) than the age group (25–44), which had a prevalence (5.5%). This may support the idea that these age groups are more exposed to the risk factor of infection. This exposure may occur during sexual activity, work, or travel, but statistical analysis showed a relationship between age and the age of infected individuals in these age groups compared to the ages included in this study (p 0.05).

Table 1 Tests result of anti-HBsAg in general population by ELISA test

Age groups	No. test	Anti-HBs Ag+	Anti-HBc IgM+		ELISA		
			NO	Index	Mean of the O.D(650nm) of negative control	Mean of the O.D(650nm) of positive control	Mean of the O.D(650nm) of patient samples
>15	95	2	1	0.67	0.011	0.496	1.64
15-24	98	4	3	0.89	0.034	0.360	1.42
25-44	72	3	2	0.83	0.03	0.322	1.86
45<	89	5	1	0.49	0.06	0.400	2.00

The statistical analysis relieved significant difference between the mean of optical density of patients samples and negative control in all age groups (LSD(0.05)1.783)

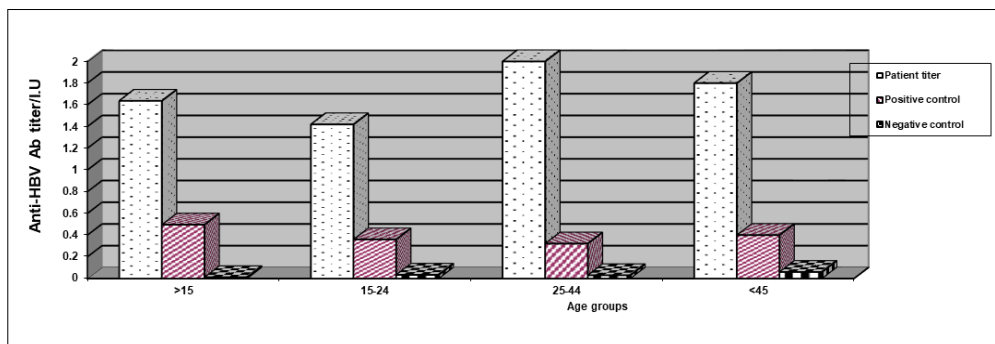


Figure 1 The status of anti-HBs Ag in different age groups of general population by ELISA test

Table 1 showed that the prevalence of anti-HBsAg in the general population was (3.95%), and that out of a total of 14 positive samples, 7 also tested positive for anti-HBcIgM, a marker for recent or acute HBV infection (Yin and Tong, 2006). This finding was also supported by (Al-Awady et al., 2008), who explain that although the age of infection is primarily at birth due to vertical transmission (from mother to child), the age of presentation is between 25 and 35 years old because the patients are asymptomatic and were unintentionally identified by routine testing while giving blood, getting married, or working, all of which are common activities for people in this age range. According to several risk exposures of ageing groups, such as injections with syringes, blood transfusions, and invasive operations, the incidence of infection in the older age groups can be explained. Numerous findings published by Allwright et al. in 2000, Sandesh et al. in 2006, and Memon et al. in 2010 support the findings of the current investigation. According to the sex; male was more infected with disease than female (10:4) with rate (71.4: 28.6) as relive in the figure (2):

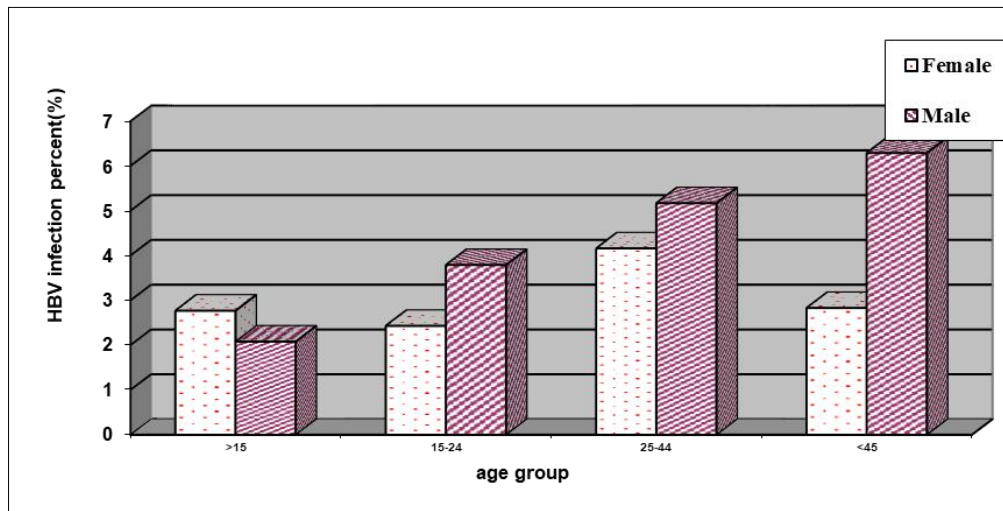


Figure 2 The relation between age groups and HBV infection in general population according to age and sex

This outcome was consistent with other studies, such as Khan's (2011). Additionally, (Dennis et al., 2005) and (Alexander et al., 2007) found that men predominate over women in all populations of anti-HBsAg carriers, supporting the well-documented fact that higher anti-HBsAg seroprevalence has been reported in male than in female for populations in some Asian countries. Women are more likely than men to clear anti-HBsAg, despite the fact that this is a well-established but poorly understood factor in chronicity (David and Daniel, 2003). These findings support earlier research in this area conducted in Iraq (Husain, 1997) and 2012 (Heim). There were a total of 14 positive anti-HBsAg tests; 8 of them were in urban areas, and the remaining 6 were in rural areas, with a rate of infection of 2:1. In this study, the hepatitis B virus distributions by place of residence showed that there was a significantly higher prevalence of the virus in urban than rural areas ($p > 0.05$). This conclusion could be the result of improved health education in urban than rural regions, which promotes early disease diagnosis. The increased frequency in urban regions may be attributed to the crowded nature of cities, which may promote HBV and HCV transmission. These findings are consistent with those of earlier research conducted in Iraq by Hussin (1997) and Al-Awady (2008). The distributions of the hepatitis B virus in this study showed a significant difference ($p > 0.05$) between the high prevalence of the virus in low economic status, medium economic status, and low economic status. With HBV, the proportion of those with good, medium, and low economic standing was (1:1:2), respectively. The current study supports earlier research by Mistik and Balik (2001), which revealed that those with lower socioeconomic status and less hygienic living conditions are more likely to contract HBV than other people.

It is consistent with the findings of other studies conducted in Japan (Dennis et al., 2005), which demonstrated that lower socioeconomic states have higher rates of HBV prevalence. These results were confirmed by additional findings published by Alter (1993) and Murphy et al. (1994). The distributions of the hepatitis B virus in this study, according to educational level, showed that patients with low educational level (primary and secondary school education) compared to those with high educational level (graduate and post graduate education), showed a high prevalence of them with a significant difference ($p > 0.05$). According to this study's findings, persons with lower educational levels are more adversely affected by HBV at a ratio of (6:1) than those with higher educational levels.

Table 2 Distribution of patients by (residency, economical status and educational level)

The parameter	Hepatitis B	
	No (%)	
Residency	Urban	125 (8\125)= 6.4
	Rural	229 (6\299=2.6)
Economic Status	Good	59 (2\59=3)
	Medium	118 6\118=5)
	Low	177 (6\177=3)
Educational level	High	118 (2\118=1.69)
	Low	236 (12\236= 5.1)

Table 3 Tests results of anti-HBsAg in general population by ELISA, Minividas and RT-PCR

Age groups	Anti-HBs Ag+(ELISA)		Minividas value IU/MI		RT-PCR	
	NO.	Index	No.	Value	No.	Viral load IU/MI
>15	2	20.5	2	15.14	2	13.07*107
15-24	4	14.2	3	13.90	3	85.4*105
25-44	3	20.0	8	17.80	8	11.06*106
<45	5	15.72	13	17.52	13	1.668*107

Table 3 shows that although the higher mean titers for both ELISA and RT-PCR are the same (2.00 and 1.668*10⁷, respectively), the index mean of anti-HBsAg produced by ELISA technique does not entirely correlate with viral load of the virus. As shown in table (3), this was also attained at a lower mean titer. This goes back to the fact that all results were obtained automatically by the apparatus which measured the results value depending on Calibrator 1 (represent positive control) and Calibrator 2 (represent negative control). The same anti-HBsAg prevalence was obtained by Minividas technique but it was more specific than ELISA technique. By using the cutting-edge molecular technology RT-PCR, the viral load of the positive specimens was assessed. The results of this study's human and automated procedures were consistent across all examined blood samples. Since the employed kit of (IgM-HBc) was based on the competitive combination principle, the results in Fig. 3 in the general population group refer to positive correlations with (IgM immunoglobulin) as a component of adaptive immune response and viral load of RT-PCR test. The findings are consistent with a 2008 study by Han et al. who discovered that the IgM anti-HBc and HBV DNA viral load combination has a positive colouring and enhances diagnostic capability. The high viral load titer in children under 10 who were infected. This can be explained by the fact that their mother was either unvaccinated or diseased when they were born, or it could be due to a contaminated device or a hospital-related infection (nasocomial infection). Since the majority of those in this age group had positive anti-HBc IgM test results, the higher virima observed in the age group (31-40) indicates that there are newly infected individuals in this age group. Higher age virus loads exhibit the gradual reduction. This may be due to the fact that these groups were exposed to the infectious agent before a diagnosis was made and that because of their advanced age, they were more likely to undergo medical procedures and receive blood transfusions.

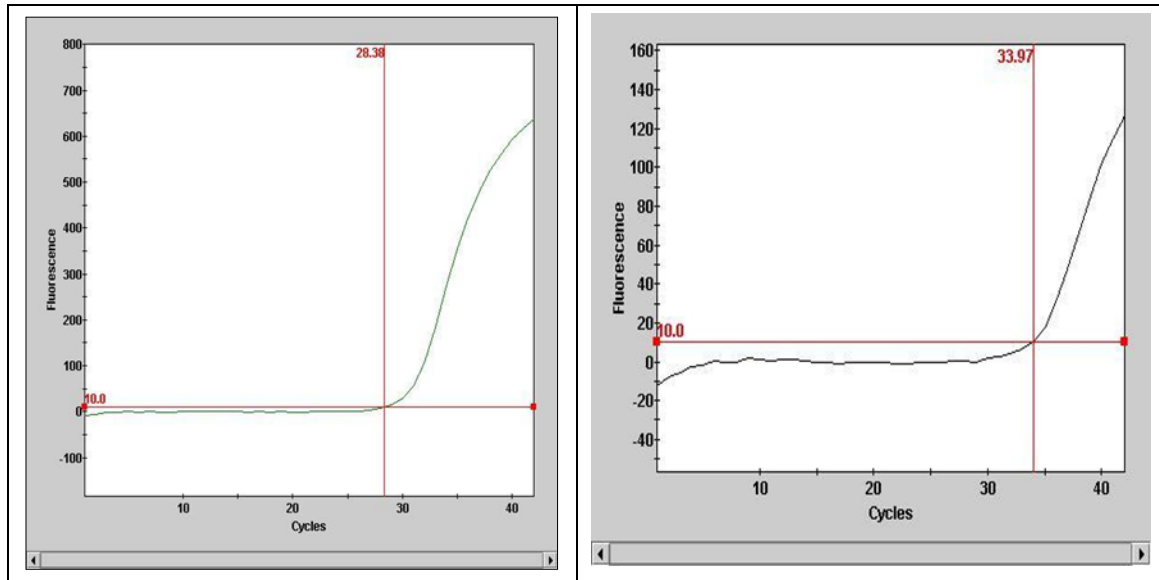


Figure 3 Relative fluorescence vs – cycle number – amplification plot showing two positive cases with two different (Ct)s (28.3 and 33.9 which indicates different levels of viral genome

4 Conclusion

In Babylon province, the prevalence of HBsAg among the general population is 3.96%, which is significantly greater than the prevalence of HBsAg among healthy blood donors (0.76%). Significant correlations were found between the HBsAg infection and male sex, urban areas, low socioeconomic status, and low educational attainment. Hepatitis B disease prevalence can be evaluated by determining the viral load of the infection using RT-PCR.

Compliance with ethical standards

Acknowledgments

I am grateful to all of those with whom I have had the pleasure to work during this and other related projects.

Disclosure of conflict of interest

The authors have no conflicts of interest to declare.

References

- [1] AL-Awady, H., AL-Shook,M., and AL-Jubory,S.S.,(2008). The Epidemiology of Chronic Hepatitis B in Babylon Province.M.SC thesis. College of Veterinary Medicine/ Babylon University.
- [2] Alexander, I. K., and Kourtis, A. P.,(2007).Hepatitis B Updated. Am J Gastroenterol. 201(3): 297-298.
- [3] Allwright S, Bradley F, Long J, Barry J, Thornton L. and Parry, J.V.(2000). Prevalence of antibodies to hepatitis B, hepatitis C, and HIV and risk factors in Irish prisoners: results of a national cross sectional survey. British Medical Journal; 321:27-83.
- [4] Alter, J.(1993).Viral hepatitis in North America. International Symposium On Viral Hepatitis and Liver Disease company, Tokyo;48-52.
- [5] Blumberg, B.S.,(2006). The curiosities of hepatitis B virus: prevention, sex ratio and demography. Proceedings of the American Thoracic Society.
- [6] David, C. D. and Daniel, D. F., (2003). Scientific American medicine. Web MD inc. USA. 824.
- [7] Dennis, L. K., Eugene, B., Anthony, S. F., Stephen, L. H., Dnal L., and Jameson, J. L. , (2005). Harrison 'sprinciples of internal medicine. 16th edition. McGraw-Hill. USA. 1830

- [8] Ganem, D. and Prince, A.M., (2004).Hepatitis B infection natural history and clinical consequences. England Journal of Medicine; 350:1118-1129.
- [9] Han,Y., Tang,Q., Zhu, W., Zhang,X. and You,L.,(2008). Clinical, Biochemical, Immunological and Virological Profiles of, and Differential Diagnosis Between, Patients With Acute Hepatitis B and Chronic Hepatitis B With Acute Flare .Journal of Gastroenterology and Hepatology ;23 (11) : 1728-1733.
- [10] Heim,S.S., (2012). Survey of viral Hepatitis type A, B, and C in Thikar city –Iraq in the years 2006-2010 .Collage of Education Journal ;2 (1):262-271.
- [11] Hoofnagle, J.H., Doo, E., Liang, T.J, Fleischer, R., Lok, A.S. Hu, W.P., Lu, Y., Precioso, N.A., Chen, H.Y., Howard, T., Anderson, D., and Guan, M.,(2008). Double-antigen enzyme-linked immunosorbent assay for detection of hepatitis E virus-specific antibodies in human or swine sera Clin. Vaccine Immunol. 15 (8): 1151–1157.
- [12] Hussin, A.G.,(1997),Seroepidemiological survey of Hepatitis B surface antigen and antibodies of Hepatitis C in Babylon province. MSc.thesis. Babylon university. collag of medicine.
- [13] Jou JH, Muir AJ, 2008 In the clinic. Hepatitis C. Ann Intern Med.;148: (6-1):6-16
- [14] Khan, S. and Attaullah, S., (2011).Share of afganistan populace in Hepatitis B and C infections .Virology Journal. 8:216.
- [15] Lee, J.M., and Ahn, S.H., (2011). Quantification of HBsAg: Basic virology for clinical practice. World J Gastroenterol.17(3):283-289
- [16] Mehwish, R., Muhamad, I., Hifza, K. and Firoz, K., (2011).An overview of Triple infection with Hepatitis B, C and D viruses.Virology Journal . 8:368
- [17] Memon, M.R., Shaikh ,A.A., Soomro, A.A., Arshad, S., and Abbas shah. Q,(2010). Frequency of Hepatitis Band in patients undergoing elective surgery, J Ayub Med Coll Abbottabad. 22: 2
- [18] Mistik, R. and Balik ,I., (2001).Epidemiological analysis of viral hepatitis in Turkey. Viral Hepatitis Journal; 9:29
- [19] Murphy, E., Bryzman,S., Matijas,L., Williams,A. and Nema,(1994).Demographic determinants of HCV seroprevalence in U.S.blood donors .Americain Journal of Epidemiology, 139: 31
- [20] Sandesh, K., Varghese, T., Harikumar, R., Beena, P., Sasidharan,V.P. and Bindu, C.S.,(2006) .Prevalence of Hepatitis B and C in the normal population and high risk groups in north Kerala. Tropical Gastroenterology; 27(80); 83
- [21] Thomas, H.C., Lemon, S., and Zuckerman, A.J, Viral Hepatitis. In: Kann M, Gerlich WH., Editors. Structure and molecular virology. 3rd ed. Oxford: Blackwell Publishing; (2005).149-18.
- [22] Wasfi, O.A.S and Sadek, N.A. ,(2011). Prevalence of hepatitis B surface antigen and hepatitis C virus antibodies among blood donors in Alexandria-Egypt. Eastern Mediterranean Health Journal.,17; (3).239-241.
- [23] WHO,(2012).Prevention and Control of Viral Hepatitis Infection. Framework For Global Action. www.who.int/csr/disease/hepatitis/GHP_framework.
- [24] Yin, L.K .and Tong, K.S., (2006).Hepatitis B infection: what the primary care doctors should know. Malaysian Family Physician.1 (1):8-10.