Original Article

Assessment of The Immunization Status of Individuals Vaccinated By Hepatitis B Virus Vaccine In Khartoum State

Kafi SK[1]*, Khalil SSO[2], Musa HA[3]

ABSTRACT

Background: Hepatitis B virus (HBV) is widely distributed all over the world. Quite a large number of the population worldwide is exposed to the virus. It is estimated that 378 million are chronically infected and at risk of developing serious complications like liver cirrhosis and hepatocelluar carcinoma leading to death. Fortunately, an effective vaccine has been introduced to prevent infection with the virus. The rate of infection by HBV has been effectively reduced by universal vaccination with that vaccine. Post-vaccination concentration of vaccine induced neutralizing antibodies against HBsAg above 10 mlU is considered protective against the virus.

Objectives: To assess the state of immunization of Sudanese individuals previously vaccinated by HBV vaccine in Khartoum State.

Methodology: A total of 90 individuals previously vaccinated with HBV vaccine and 70 unvaccinated persons (control) had their blood tested anti-HBsAg. Specimens negative for anti-HBs Ag were further tested for total (IgM & IgG) anti-HBcAg and HBsAg by ELISA. Specimens positive for total anti-HBcAg were further tested for IgM anti-HBcAg.

Results: The majority 76(88.4%) of the vaccinated subjects and 4.28% of the control were found to be positive for anti-HBsAg. Out of the 76 subjects with positive antibody response; 68.4% showed good antibody response (protected against the virus). The remaining (31.6%) showed low antibody response and are therefore at risk of infection. In general 42.2% of the vaccinated subjects and all the control group revealed low or no antibody response and are at risk of infection. The results of this study showed statistically significant difference in the antibody response between those who received three vaccine doses (72.7%) and one dose (18.8%) with P value < 0.05. Two (2.22%) of the vaccinated subjects have developed HBV infection, compared to four (5.71%) of the control group.

Conclusion: Antibody response to HBV vaccine was found 88.4% of the vaccinated subjects, however, considerable number of the vaccinated subjects revealed low or no antibody response. Individuals, who received three doses of the vaccine, had statistically significant antibody response than those who received only one dose.

Keywords: Hepatitis B vaccine, Immunization, IgG, IgM.

Hepatitis B virus is an ubiquitous virus with a global distribution[1]. It is one of the world's most common and serious infectious agents.

1. Shamsoun Khamis Kafi, MD Department of microbiology, Faculty of Medical laboratory Science, The National Ribat University, Khartoum, Sudan.
2. Sana'a Salaheldin Khalil Osman, University of Medical Sciences and Technology, Khartoum, Sudan.
3. Prof. Hasan Abdelaziz Musa, Department of microbiology, Faculty of Medicine, The National Ribat University, Khartoum, Sudan.
* Correspondent: 0912296063

Over 2 billion people worldwide have been exposed to hepatitis B virus (HBV) infection and at least 378 million are chronically infected and at risk of severe disease and death. Nearly 25% of all carriers develop liver cancer and liver cirrhosis. HBV infection causes more than one million deaths every year[1-3]. Studies conducted in Sudan revealed high prevalence of HBV infection[4].

HBV infection rate has been effectively reduced by universal vaccination. Concentrations of vaccine-induced neutralizing

antibodies (anti-HBsAg) above 10 mlU are generally regarded as sufficient for protection against the virus[5]. The duration of protection provided by hepatitis B vaccination is yet unknown, but the presence of immune memory cells can be evaluated indirectly by measuring the immune response to a booster dose of the vaccine. Several reports have unequivocally demonstrated that some individuals with antibodies against hepatitis B core antigen as the only hepatitis B virus serological marker; are chronic carriers for HBV[6]. Detection of hepatitis B surface antigen (HBsAg) by enzyme immunoassay (EIA), utilizes both polyclonal and/or monoclonal anti-HBsAg and target the major "n" determinant of HBsAg, although other targets may also be available for detection. Such assay may be unable to detect HBsAg "a" determinant mutants[7,8]. Previous studies conducted in Nigeria, Taiwan and Italy showed antibody response to HBV vaccine to be 61%, 75.8% and 90.3% respectively[9-11].

The aim of this study was to assess the immunization status of individuals vaccinated with HBV vaccine in Khartoum State in comparison with normal non-vaccinated control.

METHODOLOGY

This prospective cross sectional analytical study was conducted during the period from March to May, 2012. Ninety individuals already vaccinated were randomly recruited to participate in the study irrespective of age, gender and duration of immunization. Immune-compromised, patients with chronic diseases or on immune suppressive therapy were excluded. Seventy non vaccinated subjects were included as control group. After agreeing to be enrolled in the study each subject was asked to fill a questionnaire composing of the basic information such as gender, age, residence, duration of vaccination and number of vaccine doses received. Thereafter, three ml of venous blood were collected from each subject under the study and drawn into plain container, allowed to clot at room temperature. The clotted samples were centrifuged at 3000 rpm for five minutes. The sera were separated into other containers and stored in a deep freezer at -20°C till tested. All the sera were then tested for anti-HBsAg using enzyme immune assay (Statfax). Specimens negative or with low titer of anti-HBs Ag were further tested for total antibody(both IgG and IgM) against hepatitis B core antigen (HBcAg) and hepatitis B surface antigen. Specimens positive for total anti-HBcAg were tested for IgM antibody against the core antigen (IgM anti-HBc Ag). All the ELISA kits used for testing the antibodies and antigens were purchased from KEWEI company (Beijing KEWEI clinical diagnostic reagents INC Add: No 19, Gucheng X Rd, Shijing Shan District Beijing, China).

RESULTS

In this study a total of 90 individuals previously vaccinated with HBV vaccine with mean age 27.58 years, range 12 - 70 years were included. Additional 70 unvaccinated subjects with mean age 32.2 years, range 16 - 70 years were included as control.

Among the vaccinated group, the majority (88.4%) showed antibody response and a remarkable number (14/ 90) were found to be negative for anti-HBsAg (non-responders) (Figure 1). Out of the vaccinated individuals with positive antibodies against HBsAg, 52(68.42%) showed good response to vaccine (protected against HBV infection) and the rest (31.58%) revealed poor antibody response (at risk of infection). On the other hand only 3(4.28%) of the unvaccinated (control) group were found to be positive for antibodies against HBsAg and all revealed poor response to the vaccine (Table 1).

Antibody response to HBV vaccine was found to be statistically better in the vaccinated group compared to the controls (P value< 0.05). Antibody response was good in 72.7% of those who received 3 doses of the vaccine compared to 18.8% of those who received only one dose

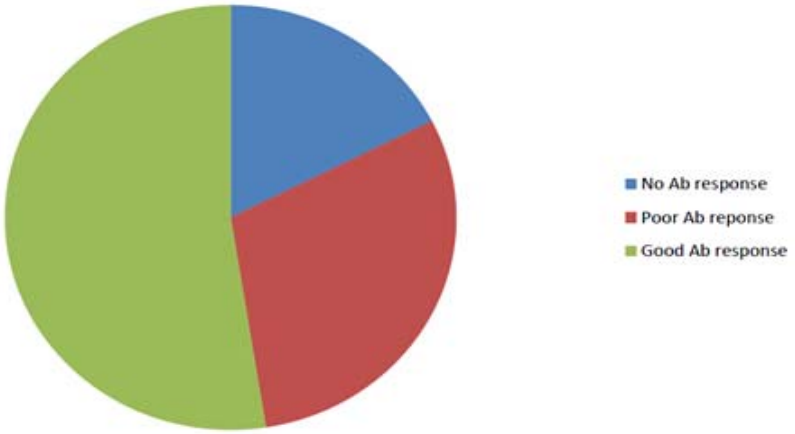

Fig 1: immunization status of vaccinated subjects (n=90)

Table 1: Immunization status of the unvaccinated group

Antibody response status	Frequency	Percentage
No response	67	95.71
Poor response	3	4.28
Good response	0	0
Total	70	100

(Table 2). Childhood immunization with HBV vaccine (3 doses) was found to be better regarding antibody response to the vaccine. These findings revealed statistically significant correlation between antibody response and number of vaccine doses received (P value<0.05).

Two (2.22%) of the vaccinated subjects and four (5.71%) of unvaccinated were found to be positive for HBsAg.

DISCUSSION

The majority (88.4%) of the vaccinated subjects included in this study showed antibody response to the vaccine (≥ 10 mlU/ml). However a considerable percentage (14.6%) is non-responders (without antibody response). Out of those with antibody response to the vaccine, 68.4% showed good response (immune) and the rest (31.6%) poor response (low immunity). In the control group, only 4.28% were found to have antibodies against HBsAg, all of them have low antibody levels (not protected). The obtained results reveal statistically significant variation in the antibody response between the studied population and the control groups (P value < 0.05).

Table 2: Immunization status of the vaccinated individuals according to the number of received doses of HBV vaccine

Number of vaccine doses	Total tested	No Ab. response		Poor Ab. response		Good Ab.response	
		Freq	Percent	Freq	Percent	Freq	Percent
1	16	3	18.75	10	62.50	3	18.75
2	27	7	25.92	5	18.51	15	55.55
3	44	4	9.00	8	18.18	32	72.72
3(childhood)	3	0	0.00	1	33.33	2	66.66
Total	90	14	55.55	24	26.66	52	57.77

The results of the current study are in agreement with a study conducted by Odusanya and his coworkers in Nigeria. They reported that 61% of vaccinated subjects developed protective antibodies compared to 18% for the control group[9]. In Taiwan, Yen-Hsuan and his colleagues reported HBsAg seropositivity of 75.8% and 70.7% among persons younger than 15years and between 15 -20 years respectively[10]. Higher antibody response (90.32%) was reported in Italy by Campagna and his companion[11].The variation in the antibody response in these studies can be attributed to the study populations with regards to age, residence, economical and atmospheric conditions and possibly vaccines qualities. Remarkable number of vaccinated persons did not show antibody response to the vaccine. This may be due to problems in the vaccination process as it is observed that some persons received only one vaccine dose more over some of them might had immunity defects. Two (2.22%) of vaccinated group and four (5.71%) of the control group were found positive for HBsAg. This finding is comparable with the figure reported by Voranush Chongsrisawat *et al* in Thailand, who reported HBsAg rate of 4% among vaccinated children[12]. Figures from Nigeria were 2% and 11.8% for vaccinated and unvaccinated groups respectively[9].

CONCLUSION

Remarkable percentage of HBV vaccinated persons showed no or poor antibody response to the vaccine and therefore at risk of infection by HBV. In fact some of them were found to be infected evidenced by detection of HBsAg in their blood. A considerable number of individuals received incomplete dose of vaccine.

REFERENCES

1. Hollinger FB and Liang TJ. Hepatitis B Virus. In: Knipe DM et al., eds Fields Virology, 4th ed. Philadelphia, Lippincott Williams & Wilkins, 2001; 2971 - 3036.
2. Robinson WS. Hepatitis B viruses. General features (human). In: Webster RG and Granoff A, eds. Encyclopedia of virology, London, Academic press Ltd, 1994; 554 - 569.
3. Mahoney FJ and Kane M (). Hepatitis B vaccine. In: Plotkin SA and Orenstein WA, eds. Vaccines, 3rd ed. Philadelphia, W.B. Saunders Company, 1999; 158 -182.
4. Gasim I. Gasim, Hamdan Z. Hamdan, Sumaia Z. Hamdan and Ishag Adam. Epidemiology of Hepatitis B and Hepatitis C virus infection among hemodialysis patients in Khartoum, Sudan. Journal of Medical Virology 2012; 84:52 - 55.
5. Howard CR and Allison LMC. Hepatitis B surface antigen variation and protective immunity. Intervirology 1995; 38:35 - 40.
6. Wolfagang Jilg, Emilia Sieger, Reinhart Zachoval, Hermann Schätz. Individuals with antibodies against hepatitis B core antigen as the only serological marker for hepatitis B virus infection: high percentage of carriers of hepatitis B and C virus. J Hepatol. 1995; 23 (1)14 -20.
7. WHO. Hepatitis B vaccines. Releve epidemiologique hebdomad aire/ Section d'hygiene du Secretariat de la Societe des Nations, Weekly Epidemiol Rec/ Health section of the Secretariat of the league of Nations 2009; 84: 405 - 419.
8. Jongerius JM, Wester M, Cuypers HT, van Oostendorp WR, Lelie PN, van der Poel CL, van Leeuwen EF. New hepatitis B virus mutant form in a blood donor that is undetectable in several hepatitis B surface antigen screening assays. Transfusion 1998; 38: 56 – 59.
9. Odusanya OO, Alufohai E, Meurice FP, Ahonkhai VI. Five- years post vaccination efficacy of hepatitis B vaccine in rural Nigeria. Human Vaccin 2011; 7(6): 625 - 9.
10. Yen-Hsuan NI, Mei-Hwei Chang, Li-Min Huang, Huey-ling Chen, Hong-Yuan Hsu, Tai-Yuan Chiu, Keh- Sung Tsai and Ding-Shinn Chen. Hepatitis B virus infection in children and adolescents in Hyperendemic Area: 15 years after Mass Hepatitis B vaccination. Annals of internal medicine 2001;135 (9):796-800.
11. 11. Capagna M, Siddu A, Meloni A, Murru C, Masia G, Coppola RC. Epidemiological impact of Mandatory vaccination against hepatitis B in italian young Adults. Hepat Mon. 2011; 11(9): 750 - 2.
12. 12. Voranush Chongsrisawat, Pornsak Yoocharoen, Apiradee Theamboonlers, Piyanit Tharmaphornpilas, Porpit Warinsathien, Supakarn Sinlaparatsamee, Siriraj Paupunwatana, Kasemporn Chaiear, Sawan Khwanjaipanich and Yong Poovorawan. Hepatitis seroprevalence in Thailand: 12 years after hepatitis B vaccine integration into the national expanded programme on immunization. Trop Med Int Health 2006; 11(10):1496-502