Hepatitis B Virus Immunity Level in Relation to Vaccine Schedule Among Medical Laboratory Health Care Providers in Machakos County, Kenya

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Abstract

Background: Hepatitis B Virus (HBV) is categorized in the genus *orthohepadnae* virus and it encompasses double stranded DNA virus. Viral infection caused by HBV can either be acute or chronic infection. Acute type of HBV infection occurs in the initial six months from birth due to exposure to HBV and in adults there might be no symptoms and no complaint of any clinical signs or symptoms.

Methods: Study design used was a cross-sectional study involving all Medical laboratory health care providers was conducted. Hepatitis B Virus surface antigen rapid test and enzyme linked immunosorbent assay (ELISA) test were used to test the blood samples collected from the subjects and the results were analyzed using chi-square and verified using Pearson Correlation Coefficient.

Results: The study found that the number of participants who were unvaccinated were 16(17.39%) of which 2 had protective immunity and 14(66.66%) had low immunity level. Participants who had received first dose of HBV vaccine were 12 of which 3(14.28%) had low immunity level while 9(12.5%) had protective immunity level. Participants who had received second doses of the vaccine were 11(11.96%) of 2(9.52%) had low HBV immunity level where as 9(12.5%) had protective immunity level. Participants who had complete HBV vaccine doses (3 doses) were 52(56.52%) of which 2(9.52%) had low immunity level while 50(70.42%) had protective immunity level.

Conclusion: The research concluded there is need for completion of HBV vaccine schedule of the three doses so as to increase the immunity level since 70.42% of respondents who had completed the three doses had protective immunity levels above the cutoff point recommended by the WHO (≥10 mIU/ml).It is advisable to get a booster vaccine after completion of the three HBV doses according to the vaccine schedule.

Keywords: Hepatitis B Virus immunity, Vaccination, Healthcare providers.

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1. Introduction

Hepatitis B Virus (HBV) being a double helix DNA virus of genus *orthohepadnavirus* in the *hepadnaviridae* family (Schaefer, 2007). The other viruses in the same category include: Hepatitis A virus (HAV), Hepatitis C virus (HCV), Hepatitis D virus (HDV) and Hepatitis E virus (HEV). Hepatitis B Virus is considered among world's major viral infection causing disease with infected people estimated at 257 million (Wijayadi et al, 2018). Almost 296 million people confirmed with chronic form of HBV infection, with estimated 1.5 million new infections every year (WHO, 2019). According to WHO 2019, an estimated 820000 deaths occur from primary liver cancer and liver cirrhosis and hepatocellular carcinoma. Pacific Region (Western) and African Region people suffering from chronic infection of Hepatitis B virus where 116 million and 81 million respectively (WHO, 2019).

Hepatitis B virus surface antigen proteins normally appear in blood between 1 to 10 weeks after infection. Surface antibodies of Hepatitis B virus appear after Hepatitis B virus surface antigen has disappeared from the immune system and they make one develop immunity against Hepatitis B virus infection. The worldwide consensus of the lowest acceptable protective concentration of Hepatitis B virus antibodies is 10 mIU/ml to assure protection. Hepatitis B virus infection symptoms of include fatigue, malaise, nausea, clay colored stool, vomiting, jaundice, dark urine, liver failure and abdominal pains. Vaccination remains very key in preventing HBV infection among medical laboratory health care providers although there has been no full implementation of the policies on vaccination due to low funding of health care systems in the low-income countries (Ogundele et al, 2018). Humoral antibody response is well known for clearing the circulating HB virus pathogens and inhibiting its spread while the Cellular mediated immune response clears the virally infected cells (Fusichari et al., 2009). The study was carried out in Machakos County Health Facilities. The study will really help in curbing or decreasing the spread of HBV infection among Medical Laboratory Health Care Providers in Machakos County as it will give the policy advice on vaccination after the outcome of the results.

2. Materials and methods

2.1 Study site

The study was carried among laboratory health care providers working in Machakos county health facilities. Machakos county borders Kajiado to the South West, Kirinyanga to the North West,

Makueni to the South, Embu to North, Kitui to East, Nairobi and Kiambu counties to the West. Machakos County stretches from latitudes 0° 45' South to 1° 31' South and longitudes 36° 45' East to 37° 45' East. The county has an altitude of between 1000-1600 meters above sea level.

2.2 Target population

The study was carried out among the Medical laboratory health care providers in Machakos county health facilities.

2.3 Study design

A cross-sectional study involving all Medical laboratory health care providers was conducted. A cross-sectional study was used because it was a study which was conducted once at a particular time where the investigator will measure the outcome and the exposures in the study participants at the same time. The participants were selected based on the inclusion and the exclusion criteria set for the study.

2.4 Sample size determination

According to the research conducted by Kisangau, et al 2018 in Makueni County health facilities the prevalence of HBs Ag was 4%. Hence Fishers et al formula was used to calculate the sample size of the study subjects.

$$n = \frac{Z^2 Pq}{d^2}$$

n=sample size needed.
z=1.96 at 95% confidence interval.
p= the approximate proportion to be found in the target population (0.4)
q=1-P = (0.6).
d=The width of confidence interval chosen (+10%).

$$\frac{1.96^2 \times 0.3 \times 0.7}{0.1^2}$$

=92.196
N =92

2.5 Sampling procedure

Stratified random sampling was used to select respondents from the 9 sub counties of Machakos County. Simple random sampling technique was used to select the participants from the facilities. A third person from the whole population will always be considered for the study till the desired sample size of 92 study subjects is achieved. The participants included in the study were both the vaccinated and unvaccinated medical laboratory health care providers working in Machakos county health facilities. Health care providers who were not medical laboratory officers, those who didn't know their vaccination health status (whether vaccinated or not) and those who were not willing to participate in the study were not included.

2.6 Data collection, assurance and control

Informed consent was sought from all participants. A structured questionnaire in English and translated in Kamba then English was administered to the participants to obtain demographic information. This checked for consistency. Primary HBV vaccination series was confirmed in each laboratory documented personal files to ascertain whether one had completed the three doses of HBV vaccine. Blood samples were collected from the participants and proper labelled. The samples were transported in cold chains to the testing laboratory to maintain samples integrity. Good standard operating procedures were adhered to so as to protect personnel from infection by using proper personal protective equipments (PPEs). Bin liners and sharp boxes for proper waste segregation for disposal so as to protect both the personnel and the environment were used. Quality Controls using both the positive and negative controls were run together with the samples to enhance the integrity and reliability of the results.

2.7 Sample collection

The subjects were prepared before sample collection, advised on how samples were to be collected and the potential risks which may occur after sample collection(there might be a little pain and or hematoma at the site of sample collection).Tourniquet was applied on the arm so as to locate the sub cubital fossa vein, which was then alcohol swabbed. 3-4 ml of blood was drawn from the vein and transferred in to a plain vacutainer bottles using closed system method.

2.8 Hbs Ag Rapid testing

One drop of blood (approximately 25 ul) from the plain vacutainer was put on to the pad of the test strip, then 1 drop of buffer approximately 40 ul was added and read the results after 15minutes of incubation at room temperature (18^0C- 25^0C). Positive Results were if two red lines appear. One line should be in the control region (C) and the other line should be at the test region (T). Negative Results were if one red line appear at the control region (C) and none at the test region (T).

2.9 Hbs Ab quantitative ELISA Test

Subject to preparation of test standards as per the test kit requirements, the test was run. All Standards and samples were added in duplicate to the micro ELISA strip plate. Standard reagent (50µl) was added to the standard well. Test samples (10µl) of each and sample diluent 40µl were added to testing sample wells respectively. Nothing was added on to the blank well. 100µI of horse radish peroxidase enzyme (HRP) conjugate reagent was then added to each of the wells, covered with an adhesive strip and incubated for 60 minutes at 37°C. The wells were then emptied and washed four times using a wash Solution (400µl) using a manifold dispenser. Complete removal of liquid at each step was essential for good performance. After the last wash, the remaining wash solution was removed by decanting. The plate was then Inverted and blotted against clean paper towels. Chromogen solution A (50µl) and chromogen solution B (50µl) was added to each well. Mixing gently was done and incubated for 15 minutes at 37°C, protected from light. A stop solution was added (50µl) to

each well. The color in the wells changed from blue to yellow. The Optical Density (O.D.) at 450 nm was done using a microtiter plate reader within 15 minutes. To determine the results, a standard concentration curve was plotted against the optical density determined for each standard concentration on the y-axis against the corresponding concentration on the x-axis of each test sample.

2.10 Data Management, analysis and presentation

Data collected was entered and cleaned using SPSS version 25. Codes were used instead of names to maintain confidentiality, integrity and retrieval of the data of the study subjects. Descriptive statistics which included measures of central tendency that is mean, median and mode were analyzed. Data was analyzed using chi square and cross tabulation for the association between the two categorical variables and verified using Pearson Coefficient correlation. Descriptive information was presented using graphs and pie charts while data analyzed using inferential statistics was presented using tables.

2.11 Ethical Considerations

The research sought the Ethical approval from Mount Kenya University Ethical Research Committee (MKUERC) and also permit from National Council for Science, Technology and innovation (NACOSTI). The research also sought approval from Machakos County Director of medical services.

3. Results and Discussion

3.1 Hepatitis B Virus Point Prevalence

The study managed to obtain 92 samples from medical laboratory health providers. After carrying out the Hepatitis B surface antigen test, two samples turned positive and 90 samples were found to be negative. The calculation of the point prevalence of Hepatitis B virus among medical laboratory health care providers in Machakos County was done as indicated below;

$$P = \frac{number\ of\ infected}{total\ number\ of\ study\ subject}\ x\ 100$$

$$= \frac{2}{92}\ x\ 100 = 2.17\%$$

P= 2.17%

The study found out that the point prevalence of Hepatitis B virus among medical laboratory health care providers in Machakos County was found to be 2.17% (Figure 1).

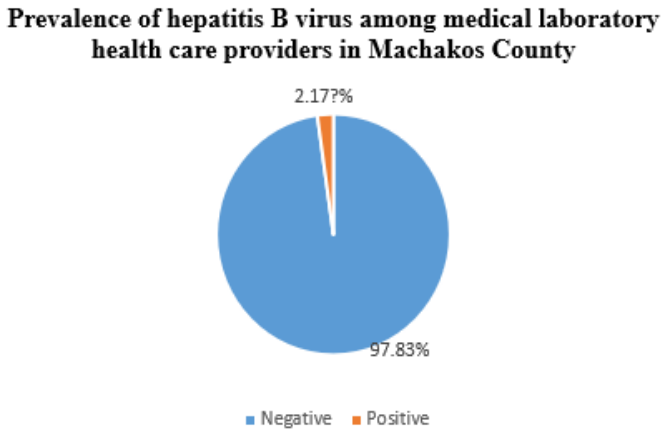

Figure 1: prevalence of Hepatitis B virus among medical laboratory health care providers in Machakos County. Source field data (2022)

3.2 Immunity level to Hepatitis B vaccine

The study found out that the total number of subjects for study were 92 out of which 21 (22.82%) had low immunity level (<10 mIU/ml) and 71 (77.17%) had protective immunity level (≥10 mIU/ml). The total number who had no (0) vaccine were 16 (17.3%) out of which 14 (66.66%) had low immunity level and 2 (2.81%) had protective immunity level. We had a total of 12 (13.04%) who had single dose out of which 3 (14.28%) had low immunity level while 9 (12.5%) had protective immunity level. For the participants who had received 2 doses were total of 11 (11.96%) out of which 2 (9.52%) had low immunity and 9 (12.5%) had protective immunity. 52 (56.52%) had received 3 doses out of which 2 (9.52 %) had low immunity levels and 50 (70.42%) had protective immunity level. There was 1 (1.40%) participant who had a booster vaccine and had a protective immunity (table 1).

For those who had not received the Hepatitis B vaccines were 16 of which 14 had low immunity and 2 had protective immunity. However, there was no significance difference since the P value was greater than 0.05. For those who had received Hepatitis B vaccine with 1 dose was 12 of which 3 had low immunity and 9 had protective immunity. The number of participants with protective immunity was higher as compared to those with low immunity. However, there was no significance difference between the 2 immunity levels since the p value was greater than 0.05. Those who had received 2 doses of Hepatitis B virus vaccine were 11 of which 2 had low immunity level while 9 had protective immunity level.

The number of participants with Hepatitis B virus protective immunity level was higher than the number with low immunity level. There was no significance difference between the 2 levels of immunity since the p value was greater than 0.05. Those who had received 3 doses of Hepatitis B vaccine were 52 participants of which 2 had low immunity level while 50 had protective immunity level. The number of participants who had protective immunity and 3 doses vaccination was higher than the ones with low immunity level. There was significance difference between

the 2 levels of immunity since the p value was less than 0.05. Those who had low immunity across the schedule of receiving HBV vaccine, there was a decline in number of participants from those who had not received the vaccine and those who had received 1 dose of HBV vaccine and a decrease showed a significance difference singe p value was less 0.05.

The number of participants further decreased on the 2nd dose of which there was no significance difference (p>0.05) from those who had received the 1st dose. The number of participants did not change at the 3rd dose of HBV vaccine schedule compared to those who had received the 2nd dose. There was no significance difference between the participants who had received 3 doses and 2 doses. Those with protective immunity and had received the 1st dose, the number of participants was higher than those who had received despite having a protective immunity of which there was no significance difference since p value greater (p>0.05). Those who had protective immunity and received HBV vaccine, the number of participants was equal to those who had received the 1st dose and there was no significance difference as shown in table 1. Those who had protective immunity level and had received 3 doses of HBV vaccine were many as compared to those who had received the 2 doses of Hepatitis B vaccine.

There was a significance difference between the number of participants who had received the 3 doses and those who had received 2 doses since the p value was less than 0.05. The number of participants who had received HBV booster vaccine were less than the number who had completed the HBV vaccine schedule and had a high protective immunity. Comparing the 2, there was a significance difference between the two levels of immunity since the p value was less than 0.05.

Table 1: Level of Hepatitis B virus immunity in relation to vaccine schedule among medical laboratory health care providers

Vaccination schedule	No. of participants per dose	Immunity	No. of cases per immunity category	P value
0	16 (17.39%)	Low immunity (<10 mIU/ml)	14 (66.66%)	0.07
		Protective immunity (≥ 10 mIU/ml)	2 (2.81%)	
1	12 (13.04%)	Low immunity (<10 mIU/ml)	3 (14.28%)	0.062
		Protective immunity (≥ 10 mIU/ml)	9 (12.5%)	
2	11 (11.96%)	Low immunity (<10 mIU/ml)	2 (9.52%)	0.052
		Protective immunity (≥ 10 mIU/ml)	9 (12.5%)	
3	52 (56.52%)	Low immunity (<10 mIU/ml)	2 (9.52%)	0.03
		Protective immunity (≥ 10 mIU/ml)	50 (70.42%)	
Booster	1 (1.09%)	Low immunity (<10 mIU/ml)	0 (0%)	0.010
		Protective immunity (≥ 10 mIU/ml)	1 (1.40%)	

4. Discussion

According to Ocan et al, (2022) a study done in Uganda found that 72% of participants had protective immunity level due to HBV vaccination which is in line with this study. According to Bruce et al 2016, found out that among the people who received HB full vaccination and never received a booster HBV vaccine had protective immune response (≥10 mIU/ml) which is in line with our study because 70.42% of fully vaccinated medical laboratory health care providers had protective immunity (≥10 mIU/ml).

According to Joukar et al (2017), a study carried among North Iranian health care workers found out that 88.9% had developed protective immunity levels of anti-body ≥10 mIU/ml while our study found out that those who had protective immunity were 77.17%(71/92) of the total participants which concurs with this study. According to Ssekamatte et al 2020 reported 57.8 % completion of HBV vaccine dosage among medical health care providers in central Uganda which is line with this study since the completion rate of Hepatitis B vaccination was 56.52% in our study. According to Ocan M et al 2020 a study carried out in Uganda found that 72% of participants had protective immunity level due to Hepatitis B vaccination which is in line with this study.

A research done in Kenya Makueni County by Kisangau et al (2017), found that 80% of the health care workers were vaccinated against HBV while this study found out that the people who were fully vaccinated were 56.52% which is not in line with this study.

A study done among medical students in Southeast Brazil by Batista, (2019) shown that 31.5% had low immunity level after Hepatitis B vaccination which doesn't concur with this study.

Among the study subjects who had received the three doses of HBV vaccine, there was only 1(1.09%) participant with a booster dose and had protective immunity against Hepatitis B virus. This shows the booster dose increases the antibody levels among the people who have already completed the 3 doses of HBV vaccination. A study done by Sajid M et al, 2021 found out that after Hepatitis B booster vaccination, there was an increase in antibody titer levels from 23.4% to 68.0% which concurs with this study.

Conclusion

The research concluded there is need for completion of HB vaccine schedule of the three doses so as to increase the immunity level since 70.42% of respondents who had completed the three doses had protective immunity levels above the cutoff point recommended by the WHO(≥10 mIU/ml).Since Hepatitis B immunity levels wanes with age as evidenced from this study and earlier studies conducted before, it is advisable to get a booster vaccine after completion of the three Hepatitis B doses according to the vaccine schedule.

Recommendations

The participants who turned Hepatitis B surface antigen positive were advised to go for the treatment. All medical laboratory health care providers working in Machakos County health facilities are strongly advised to go for the full Hepatitis B vaccination dosage schedule to improve their immunity levels against HB virus. Upon HBV vaccination completion, there is need for a booster dose

since the immunity level declines with age as evidenced from the study.

Acknowledgements
Much appreciation goes to Medical Laboratory Health Care Providers working in Machakos County health facilities for much co-operation in carrying out this research.

Conflicts of interest
The author declared no conflicts of interest.

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