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# Influence of Smoking on Immunological Response to Hepatitis B Vaccine among Medical Students and Health Care Workers in Khartoum State, Sudan

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**Abstract:**

***Background and Aims**: An important way to prevent hepatitis B infection is vaccination especially among high-risk populations including healthcare workers (HCWs). Unfortunately, immunologic response to the vaccine is not uniform. Multiple different factors such as gender, age, body mass index (BMI), smoking and underlying diseases can influence the immunologic response. So, this study was aimed at evaluation the impact of smoking in the post-vaccination immunologic response of medical students and HCWs in Khartoum state, Sudan.*

***Methods:** In this retrospective cohort study, smokers and non smokers medical students and HCWs completely vaccinated against HBV were enrolled. Data of the factors which could probably alternate the immunologic response were collected by direct interviewing questionnaire. Serum specimens were collected and evaluated by Electrochemiluminescence immunoassay (ECLIA) for Anti-HBs Ab.*

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*Titer ≥10 IU/ml was supposed to be protective. Collected data were analyzed using SPSS 16.00, P value <0.05 was considered significant.*

***Results:*** *Among total of 40 smokers (mean age =28.75 range from 21-45 years) and 40 non smokers (mean age of 26.62 range from 17-40 years), All of them were males only. 28(70%) of smokers and 34(85.0%) had a protective level of anti-HBs (≥10 IU/ml). Responsiveness was low but not statistically different in smokers than non-smokers (RR=2.0 95% CI=.832 - 4.805, P=0.108). But amount and duration of smoking is significantly associated with non protective level of anti-HBs (P=.001 and P=.05) respectively.*

***Conclusions:*** *Our study data emphasize a relation between amount and duration of smoking and their significant association with HBV vaccine non responsiveness. Study recommends periodic monitoring of anti-HBs assay for all health care workers, especially for smokers.*

**Key words:** Hepatitis B, HBV Vaccine, Smoking, Anti-HBs, Vaccination, Sudan.

## INTRODUCTION:

From the immemorial time man has been interested in trying to control disease and for well being.[1] Hepatitis-B virus infection is a global public health problem with approximately 400 million people chronically infected.[2]

Infection due to Hepatitis B virus results wide spectrum of liver diseases ranging from fulfillment hepatitis to cirrhosis and hepatocellular carcinoma.[3]

Despite advances in antiviral therapy, only minorities of chronic hepatitis B patients have a sustained response. Thus primary prevention by vaccination remains the main thrust in the control of hepatitis B infection.[4]

Recombinant DNA Vaccine is available since 1987.[5] Intramuscular vaccine administration at 0,1,6 month produces 85 -90% seroprotection rate in adolescents.[6,7]

When primary vaccination produces hepatitis B surface antibody (HBs Ab) level >100 IU/ml, it is considered to be adequate response or the vaccine is called responders. If between 10-100 IU/ml, then hypo responders and if it is <10 IU/ml, then there is no response or non responder.

Hepatitis B surface antibody titer >10 IU/ml is considered to be a marker of sustained immunity.[8] Factors associated with decreased immune response increasing age, smoking, obesity, gender, and genetic factors.[9]

**Smoking:** Tobacco smoke consists of several thousand toxic and carcinogenic substances.[10]

Ingredients of tobacco smoke have numerous extremely harmful effects on human health and are related to the occurrence of various diseases such as respiratory infections, chronic obstructive pulmonary disease (COPD) and lung cancer.[11] Tobacco smoke might affect the function of the immune system.[12]

Numerous studies in vitro and in vivo have demonstrated so far that there is an immunosuppressive effect of tobacco smoke on T and B-lymphocytes.[13, 14]

Many previous studies evaluated the immunological response following hepatitis B vaccination in different countries of the world reported a responsiveness level range almost from 85%-90%, Paltkov *et al.,* reported 86.5% responsiveness to vaccine in Israel, 2003.[15] Also Yen *et al.,* reported 86.4% responsiveness to vaccine in Taiwan, 2005. [16] And Saberifiroozi *et al*., who reported 87.3%, total response in Iran, 2006. [17]

Other few studies assessed the decreased immunogenicity and it is association with smoking status and it concluded that there is a statistical difference in the response to vaccine according to smoking status. Like finding of Mansour *et al*., who reported 3(60%) out of 5 smokers in his study population did respond to vaccine properly.[18]

## MATERIALS AND METHODS:

This is retrospective cohort study comprised of 80 participants (40 smokers and 40 non smokers' controls) of medical students and health care workers (HCWs) in Khartoum state, Sudan. They were vaccinated against hepatitis B. Ethical clearance was obtained from research ethical committee of Faculty of Graduate Studies and Ministry of Health Khartoum State, verbal consent was obtained from Hepatitis B vaccinated students and HCWs who participated in this study. Data was obtained from participants regarding age, weight, height, smoking history and amount of cigarette smoked per day and the time from last vaccination dose using direct interviewing questionnaire; Body mass index (BMI) was obtained by dividing the weight in kilograms by the square of height in meters.

Individuals who were vaccinated with vaccination schedule of 0, 1, 6 months were included in the study while individuals with a history of jaundice and individuals with a history of incomplete vaccination were excluded.

All study participants were males because smoking is a social stigma for females in Sudan.

## EXPERIMENTAL WORK:

**Specimens collection:** blood samples were collected under aseptic technique by medial vein puncture using 5 ml syringe into plain tube to obtain serum by centrifugation at 5000 rpm for 10 min. serums was kept in -20°C till serological study was performed.

**Specimens processing:** specimens were processed by **E**lectro**c**hemi**l**uminescence **i**mmuno**a**ssay (ECLIA) using cobas e411 full automation immunoassay analyzers.

**Test principle:**

- First incubation Anti-HBs in the sample (40 µL), biotinylated HBsAg (ad/ay), and HBsAg (ad/ay) labeled with a ruthenium complex react to form a sandwich complex.
- 2nd incubation: After addition of streptavidin-coated microparticles, the complex becomes bound to the solid phase via interaction of biotin and streptavidin.
- The reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then removed with ProCell. Application of a voltage to the electrode then induces chemiluminescent emission which is measured by a photomultiplier.
- Results are determined via a calibration curve which is instrument-specifically generated by 2-point calibration and a master curve provided via the reagent barcode.

**Data analysis:** The data retrieved from the questionnaires and anti-HBs antibody result were analyzed using the Statistical Package for Social Sciences (SPSS) version 16.0, Relative Risk (RR) with 95% Confidence Interval (CI) & 2 sided P-value generated from Pearson's chi square tests used to test the statistical significance.

**RESULT:**

A total of 80 participants (40 smokers as test group and 40 non smokers as control group) with history of complete vaccination schedule to hepatitis B were enrolled.

As shown in Table 1, participants of the smokers and non smokers groups did not differ at inclusion in age (range from 21-45 mean=28.75 years) *vs* (range from 19-40 mean=26.62 years). There was no difference between the two

groups concerning distribution of BMI (mean =24.1 *vs* 22.8) for smokers and non smokers respectively. Also there was no deference regarding duration from vaccination time (mean= 3.07 *vs* 3.18 years).

Out of 40 participants 28(70%) of smokers had a protective antibody titer (more than 10 IU/ml) while 12(30%) had antibody titer less than 10 IU/ml which considered not protective and participants labeled non responders.

Among non smokers control group, 34(85.0%) participants had a protective level of antibody titer ≥10 IU/ml. However 6(15%) participants did not had a protective antibody titer as shown in table 2.

The frequencies of non responders was higher in smokers (30%) compared to (15%) in non smokers, RR= 2.0, 95% CI= .832 - 4.805 (P= .108); not statistically different considering P value ≤.05. (figure1)

In smokers group the percentage of non responders were higher (100%) in participants smoked 11-20 cigarettes/day compared to (20%) in participants smoked 1-10 cigarettes/day which was statistically different (P= .001) considering P value ≤.05. (Table3)

Also as shown in Table3 the percentage of non responder were lower in participants with smoke history duration of 1-5 years (12.5%) compared to participants of 6-10 years (42.9%), 11-15 years (40%) which was statistically different (P= .05) considering P value ≤.05.

**Table 1: Age, BMI, and Duration from vaccination descriptive characteristic of study populations:**

| groups | | Minimum | Maximum | mean | Std. deviation |
|---|---|---|---|---|---|
| **Smokers (N=40)** | Age(years) | 21 | 45 | 28.75 | 6.456 |
| | BMI(%) | 19.40 | 29.80 | 24.077 | 3.2999 |
| | Duration from vaccine(years) | 1 | 5 | 3.07 | 1.071 |
| **Non smokers (N=40)** | Age(years) | 19 | 40 | 26.62 | 5.830 |
| | BMI(%) | 17.2 | 32.7 | 22.768 | 3.0315 |
| | Duration from vaccine(years) | 1 | 8 | 3.18 | 1.583 |

**Table 2: Smokers versus non smokers characteristics of vaccine responsiveness:**

| Study groups | Non Responders, N,(%) | Responders, N,(%) | Total | Relative risk | 95% Confidence Interval | P-value |
|---|---|---|---|---|---|---|
| Smokers, | 12(30%) | 28(70%) | 40(100%) | - | - | - |
| Non smokers, | 6(15%) | 34(85%) | 40(100%) | - | - | - |
| | | | | **2.000** | **.832 - 4.805** | **.108**** |

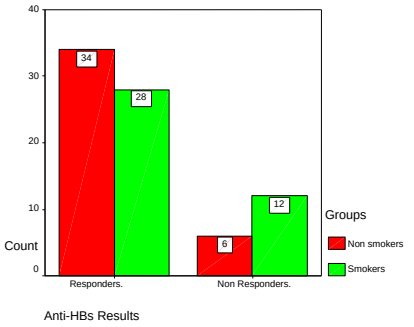
**Not significant different at the 0.05 level.

**Table 3: distribution of non responder's status according to amount and duration of smoking:**

| | | Amount of smoking | | |
|---|---|---|---|---|
| **Cigarettes/day** | **N** | Responders, N,(%) | Non responders, N,(%) | P-value |
| **1-10** | 35 | 28(70%) | 7(20%) | |
| **11-20** | 5 | 0(0%) | 5(100%) | 0.001* |
| **Total** | 40 | 28(70%) | 12(30%) | |
| | | **Duration of smoking** | | |
| **Smoke duration(years)** | **N** | Responders, N,(%) | Non responders, N,(%) | P-value |
| **1-5** | 16 | 14(87.5%) | 2(12.5%) | |
| **6-10** | 14 | 8(57.1%) | 6(42.9%), | .05* |
| **11-15** | 10 | 6(60%) | 4(40.0%), | |
| **Total** | 40 | 28(70%) | 12(30%) | |

* Significant different at the 0.05 level.

**Figure1: frequencies of Immunologic responses (Anti-HBs ≥10 IU/ml) in smokers and non smokers:**




## DISCUSSION:

Hepatitis B is the most important infectious occupational disease for the health care workers.[19] The Advisory Committee on Immunization Practices defines health care workers as members of the professions of medicine, nursing, dentistry, biomedical laboratory technicians, and emergency medical personnel.[20] The high risk of being infected is the consequence of prevalence of virus carriers in the assisted population, the high frequency of exposure to blood, body fluids and the high contagiousness of hepatitis B virus.[19]

It is well accepted that Hepatitis B vaccination induces protective level of antibody after complete course of vaccination. There are few genuine non responders. [20, 21, 22, 23] So post vaccination screening should be done in health professionals within 1-6 months of complete vaccination.

In UK evidence of HBsAb titer of >100 IU/ml is required before medical students are allowed access to patient. Students with serum level of HBsAb < 10 IU/ml have to repeat the full vaccination and those with 10 -99 IU/ml are required to receive a single booster vaccination.[24,25,26,27]

This study is from few studies which conducted in Sudan, and provide epidemiological data assessing the immune response to hepatitis B vaccine and to our knowledge the first

one to study the influence of smoking as a factor associated with lacking protective antibody titer following vaccination. Although HCWs and medical students are from high risk group, a titer of ≥10 IU/ml considered protective because the sample collected even 5 years post vaccination.

The present study revealed a good protection level (85%) of anti-HBs non smoker males control that consist with finding of Abdul Ahad *et al.,* in Bangladesh that males had (85.88%) protective level of anti-HBs.[28]

Also its similar to 88.5% reported in study conducted in Sudanese medical students in Khartoum state, Sudan by Ibrahim et al. 2014[29], 88.4% by Khalil *et al.,* 2013.[30] and 86%(82% in males) reported in Pakistan by Mohammed Zeeshan *et al.,* 2007.[31]

Considering smoker group, our study result revealed more frequencies of non responder cases 30% compared to 15% of non smoker control (P-value . 108) which is not statistically different.

Regarding smoking status number of studies noted significant association between non responder cases and smoking. For instance (P<.01) reported by Rachel *et al.,* [32] and (P=.0003) reported by Ann *et al.,* [33] and (P<0.05) reported by Mansour *et al.,* [18]

The difference between this study and other results is may be due to amount of cigarettes smoked or how long duration they smoked because this factors dose not controlled.

However this can be interpreted by our study result concerning amount of cigarette smoked per day and duration from participant being smoker which showed significant association (P= .001) and (P= .05) respectively.

## CONCLUSION:

Hepatitis B vaccines are highly immunogenic, but have decreased immunogenicity associated with increasing age, smoking, and male gender. In this study 30% of smokers and 15% of non smokers of participants had no protective level of anti-HBsAb (<10 IU/ml). Since health-care staffs including medical students are a high risk group to be contaminated with HBV, it is preferable to be evaluated for anti-HBs titer 1-6 months after full three-dose vaccination especially when these factors are present.

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**Competing interests:**
Authors declare that they have no any interests for competition.

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