

Effect of Adding Levamisole on Seroconversion Response to Hepatitis B Virus Vaccination in Hemodialysis Patients: A Single-Center Experience

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Introduction. Hepatitis B virus (HBV) infection is much more common in hemodialysis patients than the general population. Up to half of hemodialysis patients do not have adequate protective HBV antibodies after HBV vaccination. We studied the effects of adding levamisole, as an immunomodulator and adjuvant agent, on seroconversion response to HBV vaccination in hemodialysis patients. **Materials and Methods.** Thirty-six hemodialysis patients were divided into 2 groups. The first group received 40 µg of HBV vaccine intramuscularly at 0, 1, and 6 months plus 100 mg of oral levamisole per day for 12 days. The second group received the same amount and method of vaccine and placebo. Serum antibody levels were measured in each group after 0, 2, and 4 months after the last dose of vaccination.

Results. Anti-HBV antibody level in the patients who received levamisole was lower than that in the control group. Antibody levels in the levamisole group at 0, 2, and 4 months after the last dose of vaccination were 44.4%, 77.8%, and 77.7%, respectively. In the control group, response rates at 0, 2, and 4 months were 55.6%, 72.2%, and 77.8% respectively ($P = .04$, $P = .12$, and $P = .08$, respectively).

Conclusions. Anti-HBV antibody level was significantly lower immediately after HBV vaccination when it was accompanied by levamisole administration. However, no significant differences were observed between the two groups at 2 and 4 months. Further evaluation is recommended to assess the effect of adding levamisole on Hepatitis B surface antibody titer in hemodialysis patients.

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INTRODUCTION

Hepatitis B virus (HBV) infection affects more than 2 billion people worldwide.¹ It is more prevalent in high-risk populations such as dialysis patients than in the general population, because of the greater exposure to HBV during dialysis procedure.² Despite control and preventive measures, viral hepatitis B still seems to be a major concern in medical centers with hemodialysis facilities,³ and all dialysis patients should be vaccinated for HBV.⁴ However, end-stage and pre-end-stage renal disease patients have an impaired immune response to recombinant HBV vaccination.⁵ Therefore, any

effort to increase seroprotection rates in uremic patients after HBV vaccination is important. In this regards co-administration of zinc, γ -interferon, granulocyte-macrophage colony-stimulating factor, and levamisole are used for enhancing of seroconversion rate after HBV vaccination.⁶

Levamisole, a phenylimidothiazole, is an antihelminthic drug that stimulates T cells and macrophages and improves cellular immunity by increasing the secretion of chemotaxis and proliferation of these cells.⁷ We studied the effect of adding levamisole on seroconversion rate of HBV vaccination in hemodialysis patients.

MATERIALS AND METHODS

Forty patients on maintenance hemodialysis (3 times per week) from 2 dialysis centers were enrolled in this study from 2006 to 2007. They were patients without any history of HBV vaccination. The exclusion criteria were a hepatitis B surface antibody (HBsAb) titer higher than 10 mIU/mL, receiving immunosuppressive drugs, a history of malignancy, a positive human immunodeficiency virus antibody test, and a positive hepatitis C virus antibody test. The study protocol was approved by the local ethics committee and the patients provided informed written consent. Using permutation blocking method, the patients were randomized into 2 groups. The first group (levamisole) received 40 µg of recombinant HBV vaccine intramuscularly at baseline and 1 and 6 months, plus 100 mg of levamisole (Porsina Pharmacy Co, Tehran, Iran), 6 days before and 6 days after vaccination. The second group (control) received the same vaccination protocol, except for placebo instead of levamisole. Vaccination and drug administration were done after hemodialysis sessions. The patients, hemodialysis technicians, and laboratory staff were all blinded to randomization. The patients were followed up by measurement of serum HBsAb level using an immunoenzymometric assay (Radim, Rome, Italy) after the 3rd dose of vaccination and at 2 and 4 months interval thereafter. Patients with a serum antibody level more than 10 MIU/mL were considered as responders. Vaccines used for this study were made by LG Life Sciences LTD (New Delhi, India).

Data Collection and Analysis

Other than HBsAb levels, demographic data including age, gender, race, and weight, along with duration of dialysis and serum level of albumin were collected. The collected data were analyzed using the SPSS software (Statistical Package for the Social Sciences, version 15.0, SPSS Inc, Chicago, Ill, USA). The repeated measures analysis of variance and the Pearson chi-square test were utilized in the analyses. *P* values less than .05 were considered significant.

RESULTS

Of the 40 patients enrolled in the study, 4 were excluded. In the levamisole group, 1 patient developed severe gastrointestinal complications

Baseline Data of Hemodialysis Patients

Parameter	Levamisole Group	Control Group
Mean age, y	50.33	43.22
Mean weight, kg	55.38	57.80
Mean serum albumin, g/dL	3.68	3.99
Mean hemoglobin, g/dL	9.05	9.38
Mean dialysis duration, mo	54.61	46.38

due to levamisole and thus stopped receiving the drug. Another patient in this group received kidney transplantation. In the control group, 2 patients died due to tuberculosis and myocardial infarction. Of the remaining 36 patients, 21 were men (10 in the levamisole group and 11 in the control group). As shown in the Table, age, weight, serum albumin level, hemoglobin concentration, and duration of dialysis were comparable between the two groups.

The seroconversion rates after intramuscular injection of HBV vaccine were measured. Antibody level in the levamisole group at 0, 2, and 4 months after the last dose of vaccination were 44.4%, 77.8%, and 77.7%, respectively. In the control group, response rates at 0, 2, and 4 months were 55.6%, 72.2% and 77.8% respectively. Anti-HBV antibody level was significantly lower in the patients who received levamisole than those in the control group ($P = .04$) at the time of the last dose of vaccination. However, there was no significant differences between the two groups at the 2nd and 4th months ($P = .12$ and $P = .08$, respectively).

DISCUSSION

Seroconversion rate after hepatitis B vaccination is 90% in general population⁸; however, this rate is lower (50% to 83%) in hemodialysis patients.^{9,10} The frequency of nonresponders to HBV vaccination in hemodialysis patients was found to be almost 50%.¹¹ Therefore, there have been various studies conducted on the use of immune-modulator and adjuvant agents for improving both humeral and cellular immune responses to increase the HBV vaccination response rate in patients on hemodialysis; for example, adjuvants such as γ -interferon, interleukin-2, zinc, thymopentin, granulocyte macrophage colony-stimulating factor, erythropoietin, additional immune modulators (AM3, HB-AS04), and levamisole have been recently used in combination with hepatitis B vaccine in order to increase the rate of responders in dialysis

population¹²⁻²⁴; however, the efficacy of these various agents is still unclear and requires further investigations.

Patients on maintenance hemodialysis had lymphopenia with diminished T4 and T8 lymphocytes with T-cell CD4 dysfunction and abnormal antigen-presenting cell. Peripheral blood lymphocytes from uremics produced less immunoglobulin G and interleukin-2 than that from healthy individuals.^{2,5} These are the main causes of immune defect in hemodialysis patients. We studied the effect of adding levamisole on seroconversion response to HBV vaccination, because levamisole increases the absolute lymphocyte count, especially of T lymphocytes, activates lymphocytes and lymphokine release and improves chemotaxis of granulocytes.²⁵⁻²⁷

Our results showed that antibody level in the levamisole group at 0, 2, and 4 months after the last dose of vaccination were 44.4%, 77.8%, and 77.7%, respectively, compared to 55.6%, 72.2%, and 77.8% in the control group. Anti-HBV antibody level in patients who received levamisole was significantly lower than that in the control group ($P = .04$) at the time of the last dose of vaccination; however, there is no significant difference between the two groups at the 2nd and 4th months after vaccination. Although there are several reports that have shown levamisole plus hepatitis B vaccination enhances protective antibody in hemodialysis patients, in our study, anti-HBV antibody level in patients who received levamisole was lower than that in the control group. In another research in Iran on hemodialysis patients, it was found that levamisole might have no significant effect on enhancing the response.²⁸ In line with previous reports, it was also found that HBV vaccine induced cellular immunostimulation with chronic hepatitis B; however, levamisole treatment did not. The authors concluded levamisole plus vaccine therapy was not superior to vaccine treatment alone; however, this study was performed on children.²⁹

Reasons for these different results can be the dosage of levamisole administration, timing of administration, dosage of vaccine, type of vaccine and levamisole, and host genetic background.³⁰⁻³² Searching for available databases about levamisole effect on HBV vaccine, a 90% seroconversion rate (one of the highest seroconversion rates) was reported by a research group that included 20 µg of

HBV vaccine with more than 95% purity plus 100 mg of levamisole for 12 days, and they suggested intradermal vaccination could have higher antibody response.²² We also use 100 mg of levamisole for 12 days with 40 µg of HBV vaccine, but we have only 77.8% antibody level. It is noteworthy that both studies took place in Iran with almost the same number of patients, but with different types of HBV vaccines.

Although we found lower anti-HBV antibody level in our patients who received levamisole, in fact, levamisole is a safe drug, well tolerated, and available and not expensive. Thus, we suggest its use only in nonresponders to HBV vaccination (patients who do not respond to the vaccination program), as there are reports that show higher responder rate in co-administration of levamisole to routine HBV vaccination program.²¹ It would be better if we performed this research in a larger number of patients and explained the level of antibody by titer. Therefore, we suggest further studies to describe the level of antibody by titer and percentage together, evaluate combination of intramuscular and intradermal hepatitis B vaccination, implement second-generation recombinant HBV vaccine with more purity, and apply other types of levamisole. Other limitations of our study were noncompliance of the patients and gastrointestinal complications that caused drug withdrawal.

CONCLUSIONS

Seroconversion response rate to HBV vaccination in hemodialysis patients who received levamisole was significantly lower than that with placebo at the time of the last dose of vaccination. However, this effect did not last up to the 2nd and 4th months after the last dose of vaccination. Further evaluation to assess the effect of adding levamisole on HBsAb titer in hemodialysis patients is recommended.

CONFLICT OF INTEREST

None declared.

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