Efficacy of Primary Hepatitis B Immunization in Children with Acute Lymphoblastic Leukemia

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ABSTRACT

Background: *Children with acute lymphoblastic leukemia (ALL) carry a high risk of hepatitis B virus (HIV) infection. The present study was conducted to see if prior routine hepatitis B vaccine received as a part of national immunization program could prevent HBV infection in these children.* ***Methodology:*** *Ninety-six children with ALL were screened for HBV. Children were divided into three groups according to their initial HBV serology; previously vaccinated children (Group I) (n=34) previously unvaccinated and seronegative children (Group II) (n=56),and unvaccinated but HBsAg negative and anti-HBs positive children (group III) (n=6). Sixty-seven of 96 (69.7%) children received vaccination. The schedule was initiated during the third month of maintenance therapy and each course consisted of three doses given at one month interval.* ***Results:*** *Anti-HBs seroconversion following the first course of three doses of hepatitis B vaccination in group I, II and III was 57%, 33% and 100%, respectively. It increased to 97% in Group I, 62.5% in Group II, 100% in Group III. HBsAg positivity was found in 11 children (11.5%) and all of them developed chronic hepatitis B. Ten of them were in Group II whereas only one child was in Group I (P<0.04).* ***Conclusion:*** *This data reveals that routine HBV vaccination within the national immunization program plays an important role in decreasing subsequent hepatitis B infection in children with ALL.*

Key words: *Acute lymphoblastic leukemia, Children, Hepatitis B vaccine.*

INTRODUCTION

Hepatitis B is one of the most important causes of acute and chronic hepatitis world over. Children with leukemia are especially at high risk for developing hepatitis B infection due to immunosuppression secondary to chemotherapy, radiotherapy, and multiple blood transfusions(1). Majority of them develop chronic hepatitis(2). This may play an adverse prognostic role in terms of their disease-free survival because of delay in chemotherapy(3). The prevalence of hepatitis B virus (HBV) infection in healthy population in Turkey is reported as 5-15%(4,5). Both the high incidence of HBV and the lack of immunization throughout Turkey increased the risk of HBV infection for all children. However, the mandatory vaccination program against HBV has been in place since 1998 in our country. The aim of this study was to see if the routine hepatitis B vaccination in healthy children could decrease the risk of HBV infection in patients with acute lymphoblastic leukemia (ALL).

METHODS

Hepatitis B virus serology data of 96 children with ALL diagnosed between January 1998 and December 2004 were retrospectively analyzed. All children were in continuous first complete remission treated according to BFM-ALL-95 chemotherapy protocol(6). Blood products given during therapy were routinely screened for HBV, HCV and HIV by the Abbott-Axsys-System assay.

Children born in 1998 or thereafter had been immunized according to Turkish compulsory vaccination schedule with recombinant vaccine against hepatitis B surface antigen. The primary vaccination schedule for HBV included three doses of vaccine administered at the first week of life, 2nd and 6th months after birth.

The vaccination status of patients for HBV was recorded from their vaccination cards. All patients were screened for hepatitis B virus at diagnosis and prior to commencing each course of hepatitis B immunization during chemotherapy by testing HBsAg, anti-HBs, anti-HBeAg, HBeAg, HBcIgM, and HBcIgG. Assays were done by the Abbott-Axsys-System assay (a qualitative, third-generation micro particle enzyme immunoassay).

Children were divided into three groups according to their initial HBV serology;

Group I: The ones, who received primary HBV vaccination prior to diagnosis of leukemia and were HBsAg negative and anti-HBs positive (*n*=34).

Group II: *Seronegative children*: The ones who were not previously vaccinated and their HBV serology (HBsAg, anti-HBs, anti-HBeAg, HBeAg, HBcIgM, and HBcIgG) were all found negative at the time of diagnosis (*n*=56).

Group III: The ones who had not received primary vaccination but, were HBsAg negative, anti-HBs positive (*n*=6).

Children with HBsAg positivity at the initial diagnosis were excluded from the study. Sero-negative children during chemotherapy were vaccinated with hepatitis B vaccination when their leukocyte and lymphocyte count were above 3000/ mm^3 and 1000/mm^3, respectively. Vaccination was initiated during the 3rd month of maintenance therapy and consisted of three doses given at 0, 1 and 2 months. The vaccination dose was 40 μg per dose. Antibody titer above 10 IU was accepted as protective against HBV infection. The ones who failed to develop protective antibody titers against hepatitis B vaccination were re-vaccinated for the second and third time with the same schedule. Children who remained seronegative following 3 vaccination courses were accepted as failure.

Chronic hepatitis was defined as having an ALT level of more than three times the upper limit of normal, persistence of HBsAg and HBV-DNA for longer than 6 months. Percutaneous liver biopsy at least 6 months after the cessation of chemotherapy was performed on all children with chronic hepatitis B and/or hepatitis C after consent. A histological activity index (HAI) score for chronic hepatitis was determined on liver biopsy samples according to Knodell classification(7).

RESULTS

All children were in continuous first remission for a median time of 27±13.2 (range 12-84) months. Forty patients (41.7%) were found to be anti-HBs positive owing either to previous primary vaccination or HBV infection before the onset of ALL. Fifty-six out of 96 children (58.3%) were found to be seronegative for HBV at diagnosis (***Table* I**).

The leukemia risk groups, anti-HBs titers and lymphocyte counts in each group before starting vaccination during ALL treatment are displayed in ***Table* II**. Nineteen (56%) children in group I lost their immunity at median 15.9±10 (range 7-24) months of chemotherapy. The rest (44%) remained anti-HBs positive during their follow-up period (mean 27±13.2 months). However, in Group III, all children became seronegative during the same period. The levels of anti-HBs titers, mean age and lymphocyte counts prior to vaccination in each group were not significantly different from each other (***Table* II**).

The HBV status and outcome of HBV vaccination during chemotherapy are given in ***Table* III**. Sixty-seven out of 96 (69.7%) children were eligible for vaccination. Children with lost immunity in Group I (*n*=19) received 1st course of HBV vaccine. Seven children remained seronegative and were re-vaccinated. Of them, two were non-responders. Anti-HBs positivity was observed in

TABLE I HBV STATUS OF CHILDREN AT THE TIME OF DIAGNOSIS OF ALL

	Previously vaccinated (Group I)	No vaccination (Group II)	Previous HBV infection (Group III)	Total
Number of patients (*n*)	34	56	6	96
Number Anti-HBs (+) patients	34	none	6	40 (41.7%)
Number Anti-HBs (–) patients	none	56	none	56
Male/Female	13/21	22/34	3/3	38/58
Mean age (year)	6.5±3.3*	7.7±4.3*	6.4±3.45*	5.37±3.99

**P*>0.05

TABLE II HBV STATUS OF CHILDREN BEFORE VACCINATION

Patients		Standard risk group	Medium risk group	High risk group	Anti-HBs titers (IU/dL)	Lymphocyte counts (mm^3)
Group I (*n*=34) previously vaccinated	Loss of immunity	4	11	4	354±125*	989±198*
	remained anti-HBs (+)	7	7	1	575±224*	1013±275*
Group II (*n*=56) not vaccinated	No immunity	16	34	6	none	1009±246*
Group III (*n*=6) previous HBV infection	Loss of immunity	1	2	3	389±129*	1121±367*

**P>0.05*

these 2 non-responders following the 3rd course of vaccination. Ten children out of 56 (19.6%) in Group II were lost due to either relapse or toxicity during chemotherapy and 4 became HBsAg positive before. Therefore, only 42 patients (75%) in this group were eligible for the vaccination. Twenty-four seronegative children following the first course were re-vaccinated for the second time. Seven of them remained seronegative and received the 3rd course. All 6 patients with anti-HBs positivity at initial diagnosis in Group III became seronegative and developed protective anti-HBs titers after the 1st course of vaccination. The positive titers in this group continued on follow-up after completion of therapy. Anti-HBs positivity in Group I increased to 97% whereas it was 62.5% in Group II after completing the extended vaccination courses. The difference between the groups was not significant. Eleven children out of 96 (11.5%) developed chronic hepatitis B infection. Majority of them (*n*=10) were in Group II. The mean time in developing HBV infection was 14.2±7.4 months.

There were no significant difference between each group's results following the 1st, 2nd and 3rd courses of vaccination with *P* values 0.07, 0.1, and 0.9, respectively. However, the frequency of breakthrough HBV infection was significantly higher in children who received no primary vaccination prior to the diagnosis of ALL as compared to children who received the routine hepatitis B vaccination prior to diagnosis of ALL (*P*=0.04).

DISCUSSION

In the present study, 25 out of 40 children (62.5%) with positive Anti-HBs titers lost their immunity at mean 15.9±10 months (range 6-21 month) during chemotherapy. In various studies, the loss of humoral immunity to viral vaccination antigens has been described, suggesting the need for revaccination in children with leukemia, although successful response is not achieved in all such individuals(8-14). However, Fioredda, *et al.* from Italy(15) reported that protective antibody titers

TABLE III HBV STATUS AND OUTCOME OF HBV VACCINATION DURING VACCINATION

	Previously vaccinated (Group I) (*n*=34)	Titer (IU/dL)	No vaccination (Group II) (*n*=56)	Titer (IU/dL)	Previous HBV infection (Group III) (n=6)	Titer (IU/dL)
Ist course	19		42		6	
Anti HBs (+)	11 (57%)*	527.3±410	14 (33%)*	350±221	6 (100%)	734±423
Anti HBs (–)/HBs Ag (–)	7 (36%)	none	24 (57%)	none	none	_
HBs Ag(+)	1 (5%)	none	4 (10%)	none	none	_
IInd course	7		24			
Anti HBs (+)	5 (72%)*	720±433	15 (62%)*	275±140		
Anti HBs (–)/HBs Ag (–)	2 (28%)	none	7 (30%)	none	_	
HBs Ag(+)	none	none	2 (8%)	none		
IIIrd course	2		7			
Anti HBs (+)	2 (100%)*	790±127	6 (85%)*	634±355		
Anti HBs (–)/HBs Ag (–)	none	none	1 (15%)	none	_	
HBs Ag(+)	none	none	none	none		
Following the 3 courses of vaccination						
Anti HBs (+)	33 (97%)*	787±137	35 (62.5%)*	884±385	6 (100%)	667.3±456
Anti HBs (–)/HBs Ag (–)	none	_	1	_	none	_
HBs Ag(+)	1**	_	10**	_	none	_

**P=0.06 **P=0.04; when Group I compared to Group II*

were present in 81% of children with ALL after a median time of 12 months. The authors did not recommend further antibody determination and re-vaccination in these children. Their result is in contrast to our finding. Although, there is no clear explanation for the high rate of protective titers in the Italian study, the difference can be explained by the low number of high risk patients in that study. Persistent abnormalities of T, B and NK-cell subsets have been previously described even after 6 months of treatment in heavily treated patients(16). Majority of the children in the current study also received either intermediate or high risk chemotherapy protocols and all were lymphopenic.

The present study showed that 44% (*n*=5/34) previously vaccinated children remained anti-HBs positive throughout the leukemia treatment. On the other hand, all of the 6 anti-HBs positive children with no prior vaccination became seronegative**.** Their anti-HBs positivity was related to the previous natural HBV infection. Although, there are not enough patients in this group to make an optimum comparison, it might be speculated that vaccination could have resulted in more augmented and persistent immunity than the infection itself. Good evidence supporting this idea actually comes from various studies about HBs specific B- and T-cell responses detected after HBV vaccination in healthy individuals(17,18). Böcher, *et al.*(19) have shown that a significantly high level of circulating anti-HBs-secreting B cells enriched in the bone marrow years after vaccination. In addition to that, long term response of T cell memory to HBsAg after HBV vaccination play a crucial role in preventing chronic HBV infection(20).

In the current study, the timing for vaccination was during maintenance therapy when their leukocyte count was above 3000/mm^3 . The response rate ranged between 62.5% and 97%. Our previous study in children with cancer, including 42 patients

WHAT IS ALREADY KNOWN?

- Children with acute lymphoblastic leukemia carry a high risk of developing hepatitis B infection.

WHAT THIS STUDY ADDS?

- Primary hepatitis B vaccination during childhood decreases the prevalence of chronic hepatitis B in children with leukemia.

with ALL, have also proven that a good response rate was achieved (88%) after giving three doses of HBV vaccine during maintenance therapy(21). However, three different studies from India had much poorer rate of seroconversion varying from 10.5% to 28.6% when they vaccinated these children during induction therapy, with 3 to 5 repeated double doses (40 μg)(22-25). We used the same dose and similar vaccination schedule as in Indian studies. The main difference from Indian experience was the vaccination timing in the current study *i.e.*, during maintenance therapy instead of induction period. Although there is no consensus regarding the optimal timing of re-vaccination in children with cancer, we strongly recommend vaccinating these children when they are in complete remission with high leukocyte count after completing the intensive chemotherapy period.

A higher response rate following immunization was achieved in Group I compared to Group II. In various studies, it has been also demonstrated that memory B cells that met the antigen before, show a bigger antibody response when they meet the same antigen again(26). Our result supports this finding showing that the previous vaccination enhances immunity.

In this study, 11 children developed HBV infection during the first year of treatment. All of them went on to develop chronic hepatitis B infection. Passive prophylaxis with hepatitis B immunoglobulin (HBIg) may have protected them from infection. There are studies showing the efficacy of using HBIg for preventing infection from HBV(22). However, we were not able to use it due to its high cost. The risk of developing chronic hepatitis B infection in an earlier study from our center was reported as 48% in children with cancer(21). This high figure has proved the necessity of HBV vaccination in these children. In addition, our data clearly shows that children with previous vaccination had significantly lower rate of HBV infection than the children receiving no primary vaccination. This finding strongly supports that HBV vaccine as a part of routine vaccination before they develop ALL is a key factor in decreasing the incidence of viral hepatitis in children with cancer.

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