DISCUSSION

Hepatitis B virus infection occurs worldwide and is an important cause of acute and chronic viral hepatitis. About 2000 million people are infected by HBV and about 350 million people live with a chronic form of the disease.[151] Egypt is considered a country of intermediate endemicity of HBV (2-8% prevalence rate).[152] In areas with high and intermediate endemicity of HBV, widespread infection may occur in infancy and childhood. These infected children rarely develop acute disease, but 25 to 90% become chronic carriers who can transmit the disease for many years.[154,155] Immunization with hepatitis B vaccine is the most effective means of preventing HBV infection and its consequences. Thus, integrating HB vaccine into childhood immunization schedules has been a crucial step to interrupt HBV transmission, especially in endemic areas.[155-157] Hepatitis B vaccines are highly immunogenic, inducing a protective anti-HBs antibody titer (≥10 mIU/mL), in more than 95% of healthy children and young adult. The exact immunological mechanism and duration of long term protection against HBV is not yet fully understood.[154,157]

The persistence of B-cells committed to surface antibody production has been shown to persist beyond disappearance of antibody. There has been considerable work on both B and T-cell assays to assess ongoing immunity. However, this has not led to any clear answers, and the methods are difficult to transfer to highly endemic low-income countries where the question is most relevant. This has led to the use of a "test" dose of vaccine to ascertain whether the subject still has a secondary response to the vaccine, implying memory. Hall[158] stated that there is still a need to gather evidence on two critical endpoints: protection against infection and protection against disease.

All arms of the immune system appear to be affected in children treated for leukemia. During the course of chemotherapy, there may be substantial qualitative and quantitative defects in humoral immunity and antibody response. Partial or complete loss of protective antibody titers against vaccine preventable diseases makes leukemic children (especially younger ones) more susceptible to infections than general population.[43,44]

As regards HBV infection, its prevalence is higher in immunocompromised compared to immunocompetent patients, and the disease course is often aggravated.[157] Most haemato-oncological patients should be considered at risk either for acute acquired HBV infection or HBV reactivation. Increased risk is due to increased exposure to potentially contaminated blood products (despite conventional screening) as well as disease and treatment related factors.[159,160]

Little information is available on the robustness and duration of immunological memory in severely immunocompromised individuals.[142] The present study's aim was to evaluate the immunogenicity of HB revaccination in non immune children (assessed in previous study),[150] treated for ALL after completion of chemotherapy and determine factors affecting anti-HBs titer in those children. It included 25 leukemic children (Group I), and 50 normal children (Group II), as controls. All children have received three doses of recombinant HB vaccine as part of routine infant immunization program. Only the leukemic children were revaccinated with 3 doses of HB vaccine Engerix B (1/2 ml IM at 0, 1, 2 months) and response to re-vaccination was assessed 1-6 months after the third dose.

As regards immunity to Hepatitis B vaccine, the results of the present study revealed that among 25 non immune children who have completed chemotherapy for ALL and re-vaccinated with 3 doses of HB vaccine (Group I), only 4 (16%) were non-immune to HB vaccine (anti-HBs <10mIU/mL), and 21 (84%) children had protective anti-HBs titer (anti-HBs ≥10mIU/mL). Their mean anti-HBs titer was 145.39 mIU/mL at a mean age of 12.99 years. In normal controls of the present study (Group II), mean age was 12.58 years. Mean anti-HBs titer was 85.47mIU/ml. Only 26% of children were non-immune to HB vaccine, while 74% of children were immune.

When the leukemic children were compared to the healthy controls, more leukemic children (84%) were immune against HBV than normal children (74%) but the difference did not reach statistical significance. However, mean anti-HBs titer in leukemic children was significantly higher than in normal children (145.39 vs 85.74 mIU/ml).

So revaccination of non immune children treated for leukemia improve theire immune status to reach that of their normal counterparts even with statistically significant higher mean anti-HBs titer than normal controls who did not receive any additional doses of the vaccine.

The results of the present study were consistent with Viana et al[161] who studied thirty three children, over fifteen months old at time of diagnosis of ALL, treated for ALL after cessation of chemotherapy. These patients did not receive any additional hepatitis B vaccine doses after diagnoses of the disease. Base line anti-HBs titers were collected after a minimum period of four weeks from the end of chemotherapy then booster dose of hepatitis B vaccine was applied and, after another period of four weeks the serologic test was repeated. After booster dose of hepatitis B vaccine 26 (81.2%) of the patients became immune to HBV (anti-HBs≥10mIU/mL). When the individuals who had finished treatment but had not received the booster dose of HB vaccine were compared with the Control Group, there was a smaller number of potentially protected individuals against HBV among the leukemic children, the difference was statistically significant (p-value = 0.035). This difference virtually disappeared after the patients had received booster dose of HB vaccine.

Zignol et al[56] conducted a similar study but on children who were successfully treated for paediatric malignancies including ALL. Median age at diagnosis of malignancy was 6 years (range: 1–18 years). They have all completed the compulsory vaccination schedule in Italy (three doses of HB vaccine were given at 4 months, 6 months, and 11 months of age). Booster dose of hepatitis B vaccine was administered to 32 children who had lost protective HB vaccination serum titers Recovery of protective anti-HBs titer occur in 29 patients(22/23 patients with hematological malignancy and 7/9 patients with solid tumors) for over all efficacy of 90%.

On the other hand lower response to hepatitis B re-vaccination indicated by Yu et al.[57] They tested paediatric sarcoma patients at the end of chemotherapy for immunity to several vaccines in the United States. Their median age was 14.8 years. Patients had pre-treatment detectable antibody titer within the range of healthy children's antibody titers of comparable age. However, after chemotherapy, 71% of patients had negative titers for at least one infectious disease. Patients most commonly had negative titers for hepatitis B (64%). Forty one patients received a hepatitis B revaccination .Of the seven whose post re-vaccination anti-HBs titers were tested, 5 (71.4%) had positive titers (anti-HBs ≥10mIU/mL), and 2 patients had equivocal titers. This difference in the immune response to HB revaccination can be explained by two factors. First, they studied paediatric sarcoma patients who receive

different chemotherapy protocols than ALL patients of the present study. Second, the patients were revaccinated approximately 9.2 months after completing chemotherapy which is a relatively short interval. This may explain an incomplete reconstitution of patients' immune system.

Similarly lower response to hepatitis B revaccination reported by Baytan et al[162]They included 19 ALL patients with lost immunity to hepatitis B virus who have been previously vaccinated with Recombinant HB vaccine according to Turkish compulsory infant immunization program (at birth, 2 and 6 months). At a median time of 15.9 months after completion of chemotherapy, Revaccination of these patients was initiated during 3rd month of maintenance therapy and consisted of three doses of HB vaccine at 0,1and 2 month. The vaccination dose was 40µg/dose. Seven (36.8%) remained seronegative and revaccinated for the 2nd time, 12 patients (63.2%) developed protective anti-HBs titers (anti-HBs ≥10mIU/mL). This difference in the response to hepatitis B revaccination may be explained by that in Bayatan et al revaccination was initiated during the 3 rd month of maintenance therapy, While in the present study hepatitis B vaccine doses were administered at least 1year after cessation of chemotherapy.

On the contrary, Brodtman et al[55] reported much lower response to hepatitis B revaccination. They studied eighty children with ALL, at least one year after completion of chemotherapy. Almost 50% of children were persistently non immune to HBV despite re-immunization. They concluded that children in remission from ALL have high prevalence of humoral immune defects that are not related to any specific chemotherapy regimen. This antibody deficiency may place children with ALL at risk for development of these bacterial and viral diseases even after completion of chemotherapy. Pediatricians, oncologists or both should periodically monitor humoral immunity after chemotherapy and re-vaccinate these children as needed, to ensure prolonged immunoprotection

In the present study, there was no significant difference in sex between non-immune and immune patients to HB vaccine. This is in agreement with Zignol et al[56]

Mean age at diagnosis of leukemia in the present study was found to be higher in immune patients compared to non immune ones, (6.33 vs 4.81 years), but the difference did not reach statistical significance *(p*=.299). Moreover, negative statistically non significant correlation between the age at diagnosis and the anti-HBs titer was found (*p*=.904). That means that the older the age at which leukemia is diagnosed, the more the chance for the child of being immune and the lower the anti-HBs titer.. In agreement with Viana et al[161] and Volc et al[164] who found that age at diagnosis was non-significant predictor of immunity.

The present study revealed that there was no significant difference in mean post chemotherapy interval between non-immune and immune patients (*p*=.0.402). No significant correlation was found between post chemotherapy interval and anti-HBs titer (*p*=.100). When comparing mean anti-HBs titer of patients who have ended chemotherapy recently (≤2 year) to those who finished since a longer interval (>2 year), anti-HBs was higher in the former group, but the difference did not reach statistical significance, (*p*=.180). These findings are consistent with Viana et al[161] and Volc et al[163]

In Group Ia 100% children's diagnosis was B-ALL, While in Group Ib 95.2% was B-ALL and only 4.8% was T-ALL., showing no relation between the type of leukemia and immunity status after HB revaccination, (p=1.000). Brodtman et al[55], reported similar results as they found no significant difference in the expression of protective anti-HBs titer according to the ALL phenotype.

In the present study, the type of protocol, whether old protocol or standard/high risk modified CCG (attached as appendix) used for treatment of leukemia had no significant impact on the immune status of patients to HB revaccination, (p=.663). This was previously observed by Baytan et al[162]where patients have been treated using standard or high risk arms of BFM-ALL-95 protocol.

In the present study, although it was noticed that more immune patients to HB vaccine have received ≥10 units of blood products, the number of blood units transfused to the patients throughout their treatment did not affect significantly their immunity status, (p=.642). This is in agreement with Mokhles et al[164]who studied immunity to HB vaccine in multitransfused thalassemic patients from Mansoura.

Comparing anti-HBs titers before and after revaccination, anti-HBs titers were significantly increased after application of 3 doses of hepatitis B vaccine (145.39mIU/mL after revaccination vs 2.38 before revaccination.

Zignol et al[56] reported figures approaching to the present study. Quantitative determination of the increase in serum titers was performed for 13 out of 29 patients who received a booster dose of hepatitis B vaccine, the median serum titer 4 weeks after booster administration was 123 IU/L (range, 24-1000 IU/L).

Lower figures reported by Celkan et al [165] who studied a total of 43 patients. All of the cases were ALL. Four of the cases were high risk and received more intensive chemotherapy according to BFM-95 protocol. In 14 patients who were seronegative for HBV initially an antibody titer of 91 ± (0.8mIu/ml) was measured after the third dose of hepatitis B vaccine. This difference in quantitative increase of antibody titers may be explained by two main factors. First, vaccine doses were administered to patients on maintenance therapy for more than 6 months while in the present study vaccine schedule was applied at least 1 year of completion of chemotherapy. Second, different vaccine type and vaccination schedule applied in the previous study. Gen Hevac B® (Pasteur) was used for vaccination and patients were vaccinated on a 0, 1, 6 months schedule and re-evaluated 2 months after the last dose.

Considering the response to hepatitis B revaccination, the present study showed that 4 (16%) of leukemic children showed anti- HBs titer <10 mIU/ml(non responders), 6 (24%) showed anti- HBs titer 10-100 mIU/ml(hypo responders), and 15 (60%) showed anti-HBs titer >100 mIU/ml (good responders).

In normal controls of the present study (Group II), mean age was 12.58 years. Mean anti-HBs titer was 85.47mIU/ml. Only 26% of children were non-immune to HB vaccine, while 74% of children were immune.

Several earlier studies have been conducted on children immunized with HB vaccine during infancy in Egypt.[164,166-170] all have been vaccinated with three doses of a recombinant HB vaccine (Engerix B) at 2, 4 and 6 months of age as part of routine compulsory vaccination. These studies reported different rates of seroprotection against HBV. These differences may be attributed to several factors: First, these studies have been

conducted in different governorates and included children were gathered by different methods. Second factor is that they were conducted on children of different age groups. Third, is the time factor, as these studies were not conducted simultaneously so immunity to HB vaccine might have been affected by other factors related to vaccine quality, technique of vaccination or exposure to HBV in the community. Lastly, differences in laboratory methods used for assessment of the immunity to HB vaccine in these studies might make a comparison between their results not practical.

El-Sawy et al[167]in1999, studied children from Alexandria city and nearby districts. They reported a seroprotection rate of 53.3% in children 4.5 to 5.5 years old, with anti-HBs geometric mean titer (GMT) of 28.1 mIU/mL. These rates were lower than the rates reported in the present study, although his patients were of younger age. This study was one of the first studies conducted in Egypt looking for long-term immunity to HB vaccine. They explained this low percent of immune children by the fact that the Egyptian schedule for HB immunization (at 2, 4 and 6 months of age) yields to lower anti-HBs peaks. They recommended a fourth inoculation of HB vaccine to all previously vaccinated Egyptian children one or more years after completion of their basic immunization series to boost their present immune protection and suggested the adoption of a new HB vaccine schedule at 1, 2 and 9 months of age.

Later, other studies reported varying percent of seroprotected children against HBV. Shaaban et al[168] studied 242 children aged 6-12 years in Cairo. They reported that overall studied children, only 39.3% were seroprotected (anti-HBs ≥10 mIU/mL). Similar figures were also reported by Shatat et al.[166]

In 2009 El-Desouky et al[170]tested anti-HBs in children from Mansoura (mean 7.8 years). The overall percentage of seroprotected children (anti-HBs titer ≥10mIU/mL) was 43.2%. On the other hand, El-Sayed et al[169] published in 2009 approaching figures to these of the present study. Among healthy children from Menoufeya governorate around 6 years of age, up to 81% had anti-HBs titer ≥10mIU/mL and the median level of anti-HBs titer was 36.22mIU/mL.

In all previously mentioned studies any HBsAg and/or anti-HBc positive child was excluded. A point of weakness in the present study is that such markers were not tested in the control group which makes comparison with other studies less accurate. Moreover, this is an additional factor that might explain the lower percentage of immune patients to HB vaccine in previously mentioned studies compared to the present study. Exposure to HBV infection (evidenced by HBsAg or anti-HBc seropositivity) is known to provoke an anamnestic response with increase of anti-HBs titer.[133,165,170]

More recently in 2012 in Alexandria University Children's Hospital (AUCH), similar study.[150] revealed that among 100 healthy children with mean age of 7 years Mean anti-HBs titer was 61.57mIU/ml. Only 15% of children were non immune to HB vaccine, while 85% of children were immune.

Many international studies studied long-term immunogenicity of HBV vaccine. Every one of these studies enrolled children and/or adolescents at different ages. Moreover, they were conducted in different communities entailing different epidemiology of HBV infection as well as different immunization schedules and different brands of HB vaccines used. These differences may account for the contradicting results they showed.[171-196]

But et al[171]studied Chinese subjects who received 3-dose recombinant HB vaccine in infancy (at 0, 1 and 6 months of age). Included subjects represented a high-risk population because they were family members of HBV carriers. At 10 years follow up there were up to 81.8% of them having protective anti-HBs titer (the geometric mean titer was 80.5mIU/mL). On follow up of the same cohort of subjects at different point of time, 73% were immune at 15 years post vaccination and an even higher percentage of immune patients (76.5%) was found at 22 years. They explained this increase in percent of immune children might be due to boosting effect because of the highly effective anamnestic responses on exposure to HBV.[192,194-196] same conclusion was reached by Ni et al[181]in Taiwan where prevalence of protective anti-HBs was 75.8% in apparently healthy persons younger than 15 years. These figures approaches those reported in the present study.

Moreover, some investigators reported higher percentage of immune children than that reported in the present study. Da villa et al[173] found that 97% of children vaccinated with recombinant HBV vaccine (Engerix B given at 3, 5, and 11 months of age) 5 years earlier, had anti-HBs protective levels. They performed their study in southern Italy, a region with high prevalence of HBV. They suggested that anamnestic response occurring following natural exposure to the virus might explain the very low rate of prevalence of positive anti-HBc among previously vaccinated children.

All previous studies have been conducted in high endemicity areas where natural boosters might influence the persistence of protection. In contrast Duval et al[174] conducted their study in Canada, a low endemicity area and reported figures approaching those of the present study. They found that most of children immunised with recombinant HB vaccines were immune after 5 years (87.4% in those who received Engerix B and 81.6% in those who received Recombivax). The anti-HBs GMT of children vaccinated with Engerix B was higher than in those vaccinated with Recombivax ($p<.0001$).

Several follow up studies were conducted on newborns born to HBsAg-positive mothers.[197-199]They reported comparable rates of seropositivity to anti-HBs as those including children born to known HBsAg negative mothers or to mothers of unknown HBV status. Seventy six percent of children were seroprotected after a median follow up of 5 years in one study.[197]

On the other hand, some studies reported lower percentage of immune children than those in the present study. Zanetti et al[193] studied Italian children vaccinated as infants three pediatric doses of recombinant hepatitis B vaccine (Engerix B) given at 3, 5, and 11 months of age. Protective anti-HBs concentrations were present in 64% of children after 10 years post vaccination. Approaching figures were reported from different areas of the world.[178,189,191]

Others have shown much lower percentages of protected children. Samandari et al[183]enrolled children and adolescents from Alaska who have received recombinant vaccine series (at birth, at 1–3 and 6–9 months). Their results showed that among children of mean age 5.9 years, the percent of seroprotected subjects was only 29%. Earlier, Petersen et al[182] have reported 12.5% seroprotected children at a mean of 5.1 years and no children with retained anti-HBs ≥10 mIU/mL at a mean of 7.4 years. Approaching figures were also reported from Indonesia.[136]

In the present study, among control group there was no significant difference between non-immune and immune children regarding the sex. This is in agreement with several studies that found no relation between sex and immunity to HB vaccine.[167,170,178,202] In contrast, in a study conducted on adolescent Alaskan, multivariate logistic regression model showed that sex was a significant factor affecting immunity to HB vaccine as males were more protected (p=.04). This may be explained by the older age of their patient and the possibility that males had anamnestic response due to higher risk of exposure to the HB virus.[180]

In the present study, mean age difference was detected among normal controls, where non-immune children were significantly older than immune ones, (p=.002). Moreover, a significant negative correlation was found between the age and the anti-HBs titer of control group, (p=0.021). Which means that the older the age of the child the lower the anti-HBs titer.

The findings reported in the present study are in agreement with many studies conducted to determine effect of age on children vaccinated against HBV in infancy.[182-183,187,189,199]

In Egypt, El-Sawy et al[167]reported significantly higher seroprotection rate in younger children compared to older ones (93.3% at 7 months vs 53.3% at 5.5 years, p=.0097). Moreover, there was a significant association between the post vaccination interval and the seroprotection rate, (p=.00028). The anti-HBs GMT was highest (196.2 mIU/mL) in 7 months old children and lowest (28.1 mIU/mL) in 5.5 years old children, (p<.0001). They concluded that although a high initial seroprotection rate was elicited by the current HB vaccination schedule adopted in Egypt, there was a rapid drop in the seroprotection rates over time.

Similarly, Shaaban et al[168]reported different rates of seroprotection according to the age category, (p=.009) and a significant negative correlation between age and anti-HBs levels (r= -0.31, p=.041). Furthermore, by logistic analysis, age was found to be a significant variable for prediction of non-immune state in previously vaccinated children. For every 1 year increase in age there was a 23% increased risk of becoming non-immune after HBV vaccination, (p=.019). Same conclusions were confirmed later by El-Desouky et al[168] where the longer the time lapse since the last dose of HB vaccine, the lower was the seroprotection rate in tested children (p=.004). El-Sayed et al[169] also confirmed there was a highly significant difference in immune status to HB vaccine between children of different age groups as well as a decline in anti-HBs titer with time, (p<.0001).

Results from other countries confirmed the significant effect of age on immunity to HB vaccine. In a long-term Alaskan study, children of older age had significantly lower proportion of seroprotected individuals and lower anti-HBs GMT, (p<.05).[184] But et al[171] reported similar results in China. In a Canadian study,[173] there was a 29-fold decrease from the GMT baseline 5 years after the primary vaccination using Engerix B vaccine and up to 56-fold decrease when using Recombivax Hepatitis B vaccine. Williams et al[191]showed a 56-fold decrease in anti-HBs concentration from one to five years of age.

McMahon et al[180] followed Alaskan children for longer intervals up to 22 years of age. They reported that there was a decrease in rates of seroprotection with time. In a multivariate logistic regression model, they found that initial anti-HBs level (p<.001) and age class (p<.001) were both highly significant predictors of immune status to HB vaccine. The same findings were confirmed in Gambia, by Whittle et al.[189]

In an attempt to summarize the cumulated data on long-term HB vaccine immunogenicity, several review papers were published.[184,203-205] A review published by Leuridan and Van Damme[184]examined literature regarding the need for booster doses against hepatitis B published since 2002. They stated that nearly all recipients of hepatitis B vaccine respond with HBsAg-specific humoral and cell mediated immunity. However, antibody titers decline with time elapsed since vaccination.

In populations studied at 13, 15, and 22 years of age, the proportion of individuals who had lost antibody was limited to 10% or less. Even if these studies showed a loss of humoral immunity, this does not necessarily mean that booster doses will be required.[204] Data for >20 years, demonstrated that, in healthy individuals, vaccination conferred effective protection against acute disease and the development of HBsAg carrier state. This was true even in those with waning or disappearance of anti-HBs as they could mount a rapid and powerful anamnestic response to a vaccine challenge, together with the production of anti-HBs in circulating B-cells. [171,188,190,196]

In contrast, only Chinchai et al[187]recommended considering booster dose of HB vaccine 18–20 years after primary immunization especially in high-risk areas as "Cellular immunity" determined by detection of HBV-specific IFN-γ–producing cells was detected in only 50.6% of studied individuals.

Based on current scientific evidence, booster vaccination against hepatitis B for immunocompetent children and adults is not recommended for long-term protection. Immunocompromised patients, however, should be monitored and receive a booster vaccination if their anti-HBs levels decrease below 10 mIU/mL.[161, 171,174,203-205]

The publication of the CDC (2012) for immunization of children and adolescents "zero to 18 years old" recommends only a 3 or 4 doses HB vaccine schedules for infants with no further boosters.[206]

SUMMARY

Leukemias accounts for about one third of all cancers in children. Acute leukemias constitute 97% of all childhood leukemias, of which 75% are acute lymphoblastic leukemia (ALL). The different subtypes of ALL (B- or T-cell lineages) depend on the stage of lymphoid differentiation at which a leukemogenic event occur.

The improved survival of childhood ALL has focused attention on the late complications of antileukemic therapy. Both short and long-term immunologic effects of chemotherapy have been documented in children treated for leukemia. All arms of the immune system appear to beaffected in those children leaving them at risk for serious bacterial and viral infections.

Partial or complete loss of protective antibody titers against vaccine preventable diseases makes leukemic children more susceptible to infections than general population.

HBV infection occurs worldwide and is an important cause of acute and chronic viral hepatitis. About 2 billion people have been infected by hepatitis B virus and about 350 million people live with a chronic form of the disease. Egypt is considered a country of intermediate endemicity of HBV (2-8% prevalence rate).

Main routes of HBV infections are vertical transmission (perinatal), contact with an infected person (horizontal transmission), transmission through sexual contact and through parenteral, percutaneous and permucosal exposure to blood or other infected body fluids.

Clinically, hepatitis can present in an acute or a chronic form. Infants and children are more susceptible than adults for development of the chronic form of the disease. Chronic hepatitis is defined by at least 6 months of persistent HBV disease.

Preventing HBV transmission during infancy and childhood is a high priority because increased risk of chronicity. Immunization with hepatitis B vaccine is the most effective way of preventing HBV infection and its consequences. Integrating HB vaccine into childhood vaccination schedules is hence a crucial step to interrupt HBV transmission.

Recombinant Hepatitis B vaccines are highly immunogenic, inducing a protective anti-HBs antibody titer ($\geq$10 mIU per mL), in more than 95% of healthy children. There is excellent durability of response after a successful primary series. Even if titers of anti-HBs fall to less than the protective level (10 mIU/mL), most of these infants and young children continue to be protected against HBV infection. Long-term immunity has been found to derive from immunological memory. Cellular memory lasts longer than the humoral immune response, and its function remains efficient even after disappearance of anti-HBs from the circulation.

In cancer patients, HBV infection may be community acquired especially in endemic areas, but are also commonly iatrogenic. The course of disease is often severely aggravated. Moreover these patients are at risk of viral reactivation especially after end of treatment.

Immunity against HBV is significantly affected by chemotherapy. However, to date only a little information is available on the robustness and duration of immunological memory in severely immunocompromised individuals.

The present study's aim was to evaluate the immune response to HB vaccine in non immune children treated for ALL after completion of chemotherapy and after revaccination with three dose of hepatitis B vaccine and determine factors affecting anti-HBs titer in those children.

This study was carried out on twenty five non immune children with acute lymphoblastic leukemia (ALL) attending the hematology oncology clinic at Alexandria University Children's Hospital (AUCH), after completion of chemotherapy 1-6 months after re-vaccination with 3 doses of hepatitis B vaccine. There were 14 males and 11 females, their age ranged from 6.17 to 17.08 years with a mean of 12.99 years. Fifty healthy children were also included as controls. There were 22 males and 28 females, their age ranged from 7 to 12 years with a mean of 12 years.

Laboratory investigation included anti-HBs by ELISA have been assessed in leukemic cases and normal children.

All children included in the study have been previously vaccinated with 3 doses of hepatitis B vaccine according to the schedule adopted by the Egyptian Ministry of Health (at 2, 4 and 6 months of age) only leukemic cases known to be non immune to hepatitis B virus received 3 doses of Engerix B1/2 ml IM (at 0,1and 2 months). They were subjected to full history taking including personal data, vaccination history, history of risk factors to HBV infection, data related to leukemia diagnosis, and conditions that can affect individual's immune response. Clinical examination was done stressing on signs of relapse and signs of liver disease.

Overall the 75 subjects included in the study, 58 (77.3%) were immune and 17 (22.7%) were immune. There was no statistical significant difference between leukemic children and normal children as regards age and sex.

More leukemic children (84%) were immune against HBV than normal children (74%) but the difference but did not reach statistical significance. However, mean anti-HBs titer in leukemic children was significantly higher than in normal children (145.39 vs 85.74 mIU/ml).

So revaccination of non immune children treated for leukemia improve their immune status to reach that of their normal counterparts even with statistically significant higher mean anti-HBs titers than normal controls who did not receive any additional doses of the vaccine.

As regards the response to hepatitis B re-vaccination in leukemic children 4 (16%) showed anti-HBs titer<10mIU/ml (non responders), 6 (24%) showed titer 10-100 mIU/ml (hypo responders), and 15 (60%) showed titer >100 mIU/ml(good responders).

The results of the present study revealed that among 25 non immune children who have completed chemotherapy for ALL, only 4 (16%) were persistently non-immune to HB vaccine (anti-HBs <10mIU/mL), and 21 (84%) children had protective anti-HBs titer (anti-HBs ≥10mIU/mL). Their mean anti-HBs titer was 145.39 mIU/mL. There was no significant statistical difference between immune and non immune subjects as regards age. Negative correlation was found between the post chemotherapy interval and the anti-HBs titer leukemic subjects, although it did not reach statistical significance

Mean age at diagnosis of leukemia was found to be slightly higher in immune patients compared to non immune ones and a negative non significant correlation between the age at diagnosis and the anti-HBs titer was found. On the other hand, sex, mean post chemotherapy interval, ALL phenotype, type of protocol used for treatment of leukemia had no significant impact on immune response of patients to HB vaccine. Patients who have completed chemotherapy for ≤2 year had higher anti-HBs titers compared to patients to who have completed chemotherapy for >2 year.

The number of blood units transfused to the patients throughout their treatment did not affect significantly their immunity status to HB vaccine although it was noticed that more immune patients have received ≥10 units of blood products.

In normal controls, mean anti-HBs titer was 85.74mIU/ml. Only 13 (26%) children were non-immune to HB vaccine, while 37 (74%) children were immune. Non-immune children were significantly older than immune ones. Moreover, a significant negative correlation was found between the age and the anti-HBs titer of control group. There was no significant difference between non-immune and immune children regarding the sex.

CONCLUSIONS

From the current study, it can be concluded that:

1. Extra doses of hepatitis B vaccine are recommended in ALL patients submitted to treatment after hematologic recovery.
2. After this, anti-HBs levels should be verified to define the individual's protective status.
3. Revaccination in non immune children treated for leukemia improve their immune status to reach that of their normal counter parts even with higher mean anti-HBs titers
4. There is waning of anti-HBs titer over time in healthy children.