

Hematological Parameters and Elements Level in three Different Groups of Thalassemia Major

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Objective: To follow up the changes in red blood indices and elements level of children with thalassemia in major in the Northwest region of Saudi Arabia.

Design: Prospective case control study.

Methods: A total of 112 children (both genders) aged between newborn to 14 years including controls were studied. Blood samples were collected and the following indices were analyzed: RBC, Hb, HCT, MCV, MCH, MCHC, PLT, and RDW. The study also included some elements analysis from serum by Spectrophotometer.

Results: Sixty-eight children's with thalassemia (43 males, 63% and 25 females, 37%) were divided into three groups. Group A of 13 children from newborn to 2 years, Group B totals 19 from 3 to 8 years and group C of 36 children aged from 9 to 14 years. The result showed that there is no significant difference in blood count parameters between the three patients group except Hb concentration in the third group, where it was lower

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than the other groups. The platelet concentration in the third group was also found to be higher than the other two groups. Analysis of metal ions results showed no significant differences for serum Ca^{2+} , Pi and Mg. Interestingly our data showed a lowered concentration of Zn^{2+} with increasing age.

Conclusion: This study demonstrated a lower Hb for older group "C" due to hypersplenism and lower platelets in the splenectomized patients. Lower concentration of Zn^{2+} with age is perhaps due to the chelation therapy, a finding that suggests the use of zinc supplement to prevent the zinc insufficiency long term effects.

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α -Thalassemia is the most prevalent genetic disorder affecting human population¹. Its geographic distribution is highly variable in a number of defined areas, but the highest levels are found in population with a high incidence of malaria, where it can reach an alarming level of 80%². These disorders are caused by genetic mutation leading to absence of one or more α globin chains in the haemoglobin tetramer³.

Developed DNA methods make it possible to detect not only the most prevalent deletions that give rise to α thalassemia but also to determine the number and types of deletion present⁴⁻⁶. Normal subjects have four α genes ($\alpha\alpha/\alpha\alpha$), two on each chromosome. There are four kinds of α -thalassemia due to deletions of the structural α globin genes. In one of these cases only one gene is deleted (α^+ -thalassemia), another type of α -thalassemia trait is caused by deletion of two genes ($-\alpha/-\alpha$) or ($- -/\alpha\alpha$), the loss of three genes corresponds to haemolytic anemia known as Hb H disease ($- -/-\alpha$), and the loss of four α genes produce Hb Bart also known as hydrops fetalis syndrome^{7,8}.

Numerous studies have been conducted in different regions of Saudi Arabia to identify different types of α -thalassemia and to determine the α -gene mutations frequency⁹⁻¹³. The result has shown the presence of both deletion and non-deletion type of α -thalassemia, but at variable frequencies in different regions of the country. The heterozygous ($-\alpha/\alpha\alpha$) and homozygous ($-\alpha/-\alpha$) forms of α -thalassemia have been identified at a high prevalence in some regions of Saudi Arabia¹⁴. In the eastern region of Saudi Arabia, more than 50% of the population appears to have a clinically silent form of α -thalassemia and Hb H disease is recognized with increasing frequency¹⁵. The heterozygous α -thalassemia ($- -/\alpha\alpha$) has been show to occur at a very low prevalence, confirming that hydrops fetalis ($- -/- -$) and Hb H disease ($- -/-\alpha$) are rare in Saudi Arabia despite the high frequency of α -thalassmias¹⁶. In this study, we present the haematological and elemental differences in three different age groups with α -thalassemia patients in Al-Madinah Al-Munawarah city (Northwest of Kingdom) to follow up the progression of this disease in different age groups and in this part of Saudi Arabia. Beta thalassemia trait is reported in Jeddah area with prevalence rate of (0.96%)¹⁷.

METHODS

In this prospective controlled trial Sixty-eight children's with thalassemia major disease aged between newborn and 14 years old (both gender) were studied while visiting thalassaemic section in Maternity and children hospital in Al-Madina Al-Munawarah (Northwestern) of KSA. In April – September 2000 the patients were divided into three groups. Group A (13 patients) from newborn to 2 years old, Group B (19 patients) from 3 to 8 years old, Group C (36 patients) from 9 to 14 years old. Forty four normal individuals matched of age and sex were used as normal control. The blood samples were collected before 24 hours of blood transfusion or given medication such as Desferal, 10 ml of blood was withdrawn using disposable syring with needle gauge 22. The blood was divided into two parts; 7 ml in plain tubes were kept at 0°C after separated by centrifugation at 3,000 rpm for 10 minutes (Labofuge, Model 2000 Heraeus) and used to determine the biochemical parameters and elements levels, 3 ml in potassium EDTA tubes were used directly for haematology analysis (RBC, Hb, HTC, MCV, MHC, MCHC, PLT and RDW) by using an automated system (Boehringer Mannheim sysmex, model NT 800). Elemental analysis of the blood serum were carried out using Perckin Elmer, model 5000 automatic absorption spectrophotometer at the micro analysis laboratory, Faculty of Earth science, KAAU, Jeddah.

Statistical Analysis

Data was analyzed using statistical package. For each parameter studied the mean (x), standard deviation (SD), maximum value, minimum value, range and mean standard deviation (MSD) were calculated.

P Values < 0.005 were considered significant.

RESULTS

The data are summarized in table 1, demonstrating that there is no significant differences in blood count parameters between the three patients group except Hb concentration in the third group "C" that was found to be lower than the other groups (A & B) in view of the higher transfusion demand due to hypersplenism.

The PLT concentration in the third group was also found higher than the other two groups, which could be explained on the basis of the fact that many of those patients are splenectomized by this age, while splenectomy is an established cause for transbocytosis¹⁷.

Table 1. Complete Blood Count Parameters in Three Age Groups

Groups	RBC ($10^{12}/l$)	Hb (g/dl)	HCT (%)	MCV (fl)	MCH (Pg)	MCHC (g/dl)	Plt ($10^9/dl$)	RDW (%)
Group A (2 yrs) Control Patients	4.58 ± 0.07 3.24 ± 0.1	2.19 ± 0.18 8.42 ± 0.26	7.63 ± 0.46 3.24 ± 0.67	2.26 ± 0.57 3.57 ± 0.43	6.64 ± 0.25 4.65 ± 0.43	32.11 ± 0.3 4.61 ± 0.71	275.22 ± 3.4 61.46 ± 8.95	35 ± 0.16 12 ± 0.39
Group B (8 yrs) Control Patients	4.55 ± 0.04 3.16 ± 0.1	2.51 ± 0.14 8.5 ± 0.23	7.49 ± 0.22 3.31 ± 0.67	2.50 ± 0.51 3.33 ± 0.43	7.63 ± 0.25 6.09 ± 0.47	2.80 ± 0.48 4.18 ± 0.37	82.42 ± 3.16 97.0 ± 14.58	81 ± 0.45 42 ± 0.54
Group C (14 yrs) Control Patients	4.8 ± 0.07 3.02 ± 0.7	3.39 ± 0.25 7.96 ± 0.14	10.05 ± 0.45 3.37 ± 0.49	3.39 ± 0.62 3.32 ± 0.32	8.68 ± 0.44 5.45 ± 0.32	3.68 ± 0.53 3.63 ± 0.50	37.56 ± 9.75 64.42 ± 21.23	59 ± 0.33 3.51 ± 0.6

± MSD = Mean Standard Deviation P – Values < 0.005 considered significant

A comparison of these parameters in the three groups with healthy matching controls revealed that the concentration of RBC, Hb, HCT, MCH are significantly lower in-patient than in the healthy controls, while no significant difference in MCHC value. The concentration of PLT and RDW are higher in patients than the healthy controls.

Table 2, showed the results of metal ions analysis in three patient groups. The level of Ca, Pi and Mg has no significant differences in the serum of three patients groups, whereas a considerable increase of Fe and Cu ions concentration. The interest point is the lowered concentration of Zn ion.

Table 2. Comparison of Elemental Parameters between Three Age Groups

Groups	Mg ⁺⁺ (mmol/l)	Pi (mmol/l)	Ca ⁺⁺ (mmol/l)	Fe ⁺⁺ (umol/l)	Zn ⁺⁺ (umol/l)	Cu ⁺⁺ (umol/l)
Group A (2 yrs) Control Subject Value	0.95 ± 0.07 0.79 ± 0.03 P < 0.013	1.81 ± 0.05 1.33 ± 0.04 P < 0.015	2.38 ± 0.05 2.02 ± 0.03 P > 0.005	17.1 ± 0.05 21 ± 0.56 P < 0.0001	85 ± 0.55 8.49 ± 0.33 P < 0.0001	55 ± 0.34 23.68
Group B (8 yrs) Control Subject Value	0.98 ± 0.04 0.68 ± 0.02 P < 0.011	1.66 ± 0.05 1.37 ± 0.04 P > 0.013	2.49 ± 0.03 2.6 ± 0.02 P > 0.005	78 ± 0.48 81 ± 0.74 P < 0.0001	74 ± 0.53 7.88 ± 0.37 P < 0.0001	56 ± 0.37 48 ± 0.19 P < 0.0001
Group C (14 yrs) Control Subject Value	0.97 ± 0.03 0.72 ± 0.02 P < 0.015	1.81 ± 0.06 1.34 ± 0.03 P < 0.015	2.47 ± 0.03 1.9 ± 0.02 P > 0.005	65 ± 0.57 35.94 ± 0.6 P < 0.0001	65 ± 0.57 7.13 ± 0.22 P < 0.0001	44 ± 0.67 81 ± 0.81 P < 0.0001

± MSD = Mean Standard Deviation P – Values < 0.005 considered significant

DISCUSSION

A statistical database of the hematological parameters and trace elements level in the patients of the prevalent type of the disease and follow-up the effectiveness of the treatment program. The erythrocytic indices showed no significant changes between the three different age groups, which indicate that, the haematological parameters of patients during infancy, childhood and adolescents are quite similar with minor variation in some parameters, except of course in severe conditions or those associated with other clinical problems. Our results are quite similar with other studies carried out in other regions of Saudi Arabia¹⁰⁻¹⁴. Analysis of metal ions results showed no significant differences for serum Ca^{+2} , Pi and Mg^{+2} , whereas a considerable increase of Fe^{+2} ion concentration which is normal due to the treatment by red cell transfusion therapy and the lack of adequate utilization of the chelator deferoxamine. The interesting point we observed in our data is the lowered concentration of Zn^{+2} with increasing of age. It has been proven by Hatori, et al in 1995 that administering desferioxamine to young children in metaphysical dysphasia and abnormalities in linear growth. To explore the notion that deferoximine interferes with endochondral growth by chelating zinc, they examined the effect of the drug on chondrocytes maintained in long-term culture. It was found that deferoximine caused a dose-dependent inhibition of a wide range of functions including cell proliferation, protein synthesis, and mineral deposition. Directly relevant to the mineralization process, and hence the observation that the drug dramatically lowers the activity of alkaline phosphatase, a zinc requiring enzyme, it can be thus postulated that perhaps zinc supplement should be tried in preventions the long term consequences of the deferoxamine therapy¹⁶.

CONCLUSION

In this study, which is the first study as our best knowledge. There is no significant difference in the hematologist parameters between the three different age groups except for a lower Hb due to hypersplenism and higher platelets splenectomy in-group C. The elemental analysis showed interesting decline of Zn^{+2} concentration with increasing age, this could well be due to the effect of desferioxamine chelation therapy and thus necessitates that zinc supplement should be clinically evaluated to prevent the complications related to long term zinc insufficiency.

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