Original Article

Effect of multiple blood transfusions on hormonal profile in thalassemia children

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Objective: To investigate the possible effect of multiple blood transfusions on different hormones in thalassemia children.

Methodology: This experimental study was conducted over 12 months in Allied Hospital, Ali Zeb Foundation, and Thalassemia center in Hilal-e-Ahmar Hospital, Faisalabad, Pakistan and a total of 80 children were included in the study. They were divided into two groups; 40 β-thalassemia patients and 40 healthy controls. Each group was further subdivided based on gender and age, so that in each subgroup there were ten subjects of same gender with age ranging from 0-5 years and 6-10 years. Blood samples were analyzed for serum levels of triiodothyronine (T_3), thyroxine (T_4), testosterone, estradiol and progesterone. All the data was analyzed by using Statistical analysis of variance (ANOVA).

Results: The values of T_3, T_4, testosterone, and estradiol were significantly lower ($p<0.01$) irrespective of age and gender in thalassemia childrens when compared to normal children. However, the progesterone level was significantly higher ($p<0.01$) in thalassemia children than normal control.

Conclusions: Multiple blood transfusions induced various endocrine abnormalities in thalassemia children. Regular evaluation of oxidants and antioxidants status and hormones should be carried out in thalassemia patients during initial few years of life. (Rawal Med J 2014;39: 265-269).

Key Words: Thalassemia, blood transfusions, hormones, children

INTRODUCTION

Thalassemias are a heterogeneous group of genetic disorders characterized by decreased synthesis of one of the two types of polypeptide chains (either α or β) that constitutes the normal adult human hemoglobin molecule (HbA).[1] The adult hemoglobin tetramer molecule is composed of two alpha and two beta globin chains (α^2 β^2) each linked to a heme molecule. Patients of beta-thalassemia have symptoms like anemia, skeletal and/or endocrine changes and splenomegaly. The alpha thalassemia is very rare and found sporadically in the different areas of the world. On the other hand, the β-thalassemia is very common and endemic worldwide. The disease is most prevalent throughout the Mediterranean region, African continent, Middle East, Iran, Indian subcontinent, Burma and throughout Southeast Asia in a line stretching from southern China to the Malay Peninsula, and Indonesia islands.[2] It can be found with a carrier (heterozygote) in nearly about 4.5% of the world population or 150-200 million people in more than 60 countries. The total annual incidence of symptomatic individuals is estimated at 1 in 100,000 throughout the world and 1 in 10,000 people in the European Union.[3] In addition, each year more than 400,000 infants are born with serious hemoglobinopathies and carrier frequency is about 270 million.[4] There is a growing concern that thalassemia may become a very serious threat of life in future.[5]

In Pakistan, thalassemia is the most prevalent genetic disorder. The number of patients has touched the figure of about 8-10 million carriers.[6]

Large population size, consanguineous marriages and high birth rates make the disease highly rampant. The number of thalassemia homozygotes born is approximately 7000 each year with an overall carrier frequency of 5-5.65 % in Pakistan.[7,8] This familial disease presents with anemia in first year of life and child needs frequent blood transfusions throughout his life. The goal of transfusion therapy is to treat anemia, suppress erythropoiesis and inhibit gastrointestinal iron absorption.[9] Over the past few decades, regular blood transfusions and iron chelation have remarkably improved the quality of life and changed it from a fatal disease of childhood to a chronic disease compatible with extended life.[10] However, these recurrent blood transfusions result in iron overload and numerous complications, which appear in adolescents and young adults.[11] These complications are usually related to a number of factors such as insufficient blood transfusions, infections transmitted by transfusions, allosensitization, iron-overload associated with cardiac, endocrine and liver complications due to chelator's toxicities.[11] Iron intoxication generally damages almost all the endocrine glands such as pituitary, gonads and the pancreas though thyroid, parathyroid and adrenal glands are rarely involved.[12] Since several endocrine glands may be affected in β-thalassemia patients and their functions may be disturbed, that's why the present study was carried out to estimate the possible effects of multiple blood transfusions on serum hormones in various groups of thalassemia childrens.

METHODOLOGY

The study was conducted on 80 children (40 normal healthy control and 40 with thalassemia) of both genders ranging from 1-10 years old, selected from Allied Hospital, Ali Zeb Foundation, and Thalassemia center in Hilal-e-Ahmar Hospital, Faisalabad, Pakistan. The children were divided in to eight groups based on gender and age, in such a way that each group has ten subjects of same gender with age 0-5 years and 6-10 years. The arrangement of subjects into different groups is as follows: Group 1: Thalassemia males (n = 10), age (1-5 years); Group 2: Normal males (n = 10), age (1-5 years); Group 3: Thalassemia females (n = 10), age (1-5 years);
Group 4: Normal females (n = 10), age (1-5 years); Group 5: Thalassemia males (n = 10), age (6-10 years); Group 6: Normal males (n = 10), age (6-10 years); Group 7: Thalassemia females (n = 10), age (6-10 years); and Group 8: Normal Females (n = 10), age (6-10 years).
Blood samples were analyzed in the laboratories of the Department of Physiology & Pharmacology, University of Agriculture, Faisalabad. Hormonal analysis included Testosterone (pg/ml), Triiodothyronine (T_3; ng/ml), Thyroxine (T_4; µg/dl), Estradiol (pg/ml) and Progesterone (ng/dl). Serum assays were determined by using commercially available Kits.
Data obtained were subjected to three ways analysis of variance (ANOVA), and Duncan Multiple Range Test (DMRT) was applied in case of significant difference between groups (Duncan, 1955). The level of significance was kept at p ? 0.01.

RESULTS

The mean values of different hormones in thalassemia and normal children are shown in Table 1. The hormonal profile of normal and thalassemia children in relation to age groups is presented in Table 2.

Table 1. Mean ± S.E. values of hormones in normal and thalassemia children with respect to age and gender.

Parameters	Group A (0-5 years)				Group B (6-10 years)			
	Normal male	Thalassemia male	Normal female	Thalassemia female	Normal male	Thalassemia male	Normal female	Thalassemia female
Testosterone (pg/ml)	0.90±0.01^{b}	0.20±0.01^{e}	0.80±0.01^{c}	0.07±0.01^{fg}	1.60±0.02^{a}	0.10±0.01^{e}	0.70±0.01^{d}	0.05±0.004^{g}
T_3 (ng/ml)	92.01±1.1^{c}	72.01±2.46^{c}	98.03±0.93^{bc}	62.10±1.64^{f}	101.05±1.12^{b}	80.04±1.17^{d}	111.07±0.93^{a}	64.00±0.68^{f}
T_4 (µg/dl)	7.20±0.25^{bc}	5.31±0.16^{ef}	6.80±0.16^{c}	5.80±0.20^{de}	8.10±0.09^{a}	6.41±0.21^{cd}	7.90±0.26^{ab}	4.90±0.18^{f}
Estradiol (pg/ml)	5.51±0.20^{b}	2.60±0.06^{de}	8.10±0.09^{a}	5.10±0.17^{b}	4.21±0.05^{c}	3.90±0.15^{c}	2.05±0.08^{e}	3.10±0.17^{d}
Progesterone (ng/dl)	3.90±0.15^{cd}	6.51±0.22^{a}	4.51±0.14^{bc}	5.10±0.17^{b}	2.90±0.11^{e}	3.20±0.21^{de}	4.90±0.17^{b}	4.90±0.17^{b}

$^{a-g}$ Means shearing similar superscripts within a row do not differ ($p < 0.01$).

The data from the table revealed that the level of both hormones T_3 and T_4 are noticeable lower than their respective controls in both groups A and B. Testosterone and progesterone levels are considerably higher to the relevant control groups, but estradiol value is dramatically decreased than normal children in group A.

Table 2. Mean values of different hormones in normal and thalassemia children with respect to age.

	Group A (0-5 years)		Group B (6-10 years)	
Parameters	**Normal Children**	**Thalassemia Children**	**Normal Children**	**Thalassemia Children**
Testosterone (pg/ml)	0.85±0.01[c]	1.35±0.01[a]	1.15±0.02[b]	0.075±0.01[d]
T_3 (ng/ml)	95.02±1.19[b]	77±2.46[c]	106±1.12[a]	72±1.17[d]
T_4 (µg/dl)	7.0±0.25[b]	5.55±0.16[c]	7.6±0.09[a]	5.65±0.21[c]
Estradiol (pg/ml)	6.80±0.20[a]	3.85±0.06[b]	3.13±0.05[c]	3.5±0.15[c]
Progesterone (ng/dl)	4.2±0.15[b]	5.80±0.22[a]	3.1±0.11[d]	4.05±0.21[c]

[a-d] Means shearing similar superscripts within a row do not differ ($p < 0.01$).

Table 3. Mean values of different hormones in normal and thalassemia male and female children.

	Male		Female	
Parameters	**Normal Children**	**Thalassemia Children**	**Normal Children**	**Thalassemia Children**
Testosterone (pg/ml)	1.25±0.08[a]	0.15±0.01[c]	0.75±0.01[b]	0.06±0.004[d]
T_3 (ng/ml)	96.53±1.30[b]	76.02±1.16[c]	104.55±1.63[a]	63.05±0.89[d]
T_4 (µg/dl)	7.65±0.17[a]	5.86±0.18[b]	7.35±0.20[a]	5.35±0.17[b]
Estradiol (pg/ml)	4.86±0.18[a]	3.25±0.17[c]	5.07±0.70[a]	4.10±0.26[b]
Progesterone (ng/dl)	3.40±0.15[b]	4.86±0.41[a]	4.71±0.12[a]	5.00±0.12[a]

[a-d] Means shearing similar superscripts within a row do not differ ($p < 0.01$).

Moreover, the values of estradiol and progesterone are significantly ($p < 0.01$) elevated whereas the level of testosterone is drastically lower than the normal children in group B. Furthermore, the values of T_3, testosterone, progesterone and estradiol are remarkably decreased in thalassemia children of group B when compared to thalassemia children of group A, suggesting that the age factor have a depressing effect on these hormones. However, the age factor did not have any effect on T_4 level in thalassemia children, as the values of T_4 are not differing statistically ($p< 0.01$) in group A and B.

Table 4. Mean values of different hormones in normal and thalassemia children.

Parameters	**Normal Children**	**Thalassemia Children**
Testosterone (pg/ml)	1.00±0.06[a]	0.11±0.01[b]
T_3 (ng/ml)	100.54±1.21[a]	69.54±1.38[b]
T_4 (µg/dl)	7.50±0.13[a]	5.60±0.13[b]
Estradiol (pg/ml)	4.97±0.36[a]	3.68±0.17[b]
Progesterone (ng/dl)	4.05±0.14[b]	4.93±0.21[a]

[a-b]Means shearing similar superscripts within a row do not differ ($p < 0.01$).

All the hormone levels were significantly ($p<0.01$) lower as compared to normal children except progesterone in both male and female groups (Table 3). The overall values of testosterone and T_3 are noticeably lower in thalassemia females than males indicating a suppressing effect on these hormones in females. Moreover, the level of estradiol was considerably higher in females than males. However, T_4 and progesterone did not differ significantly ($p<0.01$) in both genders.

Most of the hormones levels such as testosterone, T_3, T_4 and estradiol are remarkably decreased in thalassemia children as compared to normal children indicating that multiple blood transfusions have some deleterious effects on these hormones in thalassemia children (Table 4). However, progesterone level significantly ($p<0.01$) increased in thalassemia children when compared to normal children.

DISCUSSION

Blood transfusion and chelation therapy are two vital measures for the treatment of thalassemia patients. Since few years, these two remedies have greatly improved the quality of life as well as life span of age up to 30 years; however frequent blood transfusions and poor compliance to therapy causes progressive iron overload, which is a major clinical complication in thalassemia.[13] Iron depositions in various endocrinal glands is responsible for the hormonal imbalance. Factors like hypoxia due to persistent anemia and perfusion defect, also contribute to the derangement.[14]

The anterior pituitary gland is particularly sensitive to free radicals oxidative stress, resulting in hormone secretion disorders mainly gonadotrophins. The accumulation of toxic

quantities of iron in the body leads to the formation of reactive oxygen species (ROS) that ultimately causes multiple endocrine damages.[15] The oxidative damage to thalassemic RBCs can cause their accelerated apoptosis and ineffective erythropoiesis,[16] leading to the rapid release of free iron which further exacerbate the condition. Various changes in RBCs of thalassemia patients are associated with constant oxidative stress within the cells caused by precipitation of excess α-globin chains and release of free iron.[17]

In this study, both T_3 and T_4 were significantly ($p<0.01$) lower in thalassemia children when compared to control group. This is in line with the previous studies.[18] The low levels of T_3 and T_4 in the present study suggest that the damage to the thyroid tissue may start much earlier in adolescence in both males and females. High level of free iron causes the iron deposition in thyroid gland, with consequent fibrosis of the glandular parenchyma, and progressive thyroid dysfunction. Although the long-term natural history is poorly understood, it is thought that high cost, non-availability of parenteral chelating agents, and poor compliance are prone to iron toxicity.

In our study, serum testosterone level was significantly ($p<0.01$) lower than the normal children in both genders. These results are in line with the previously reported studies.[19] These reduced levels are attributed to iron deposition in gonadotrophic cells, which eventually lead to delayed puberty and hypogonadotrophic hypogonadism.[20] Primary and secondary characteristics of sexual development are usually delayed in these patients.[20]

Serum estradiol level was also found significantly ($p<0.01$) lower in thalassemia children, which is consistent with previously documented studies.[21] The lower level of estradiol may be possibly due to the deposition of iron in the granulosa cells of ovaries. Moreover, delay in the age of menarche and poor breast development is also observed in thalassemia patients.[22]

The serum progesterone level was significantly ($p<0.01$) higher in thalassemia children when compared with the normal children. These results are contrary to some reports.[22,23] One of the likely reasons seems to be that other progesterone producing organs like adrenal cortex may show compensatory hypertrophy to produce more progesterone. This aspect of high progesterone in thalassemia children needs to be further explored in subsequent studies.

CONCLUSIONS

Multiple blood transfusions induced various endocrine abnormalities in thalassemia children. There were significantly low levels of T_3 and T_4, testosterone and estradiol, irrespective of age and gender. However, high level of progesterone was found in thalassemia children. These findings warrant regular evaluation of oxidants and antioxidants status and hormones in thalassemia patients during initial few years of life, and proper treatment and replacement therapy should be employed when necessary.

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Conflict of Interest: None declared
Rec. Date: Mar 1, 2014 Accept Date: May 25, 2014

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