DETECTION OF HETEROZYGOUS BETA THALASSAEMIA IN THE SIBLINGS OF KNOWN BETA THALASSAEMIA MAJOR CHILDREN

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ABSTRACT

Objective: *To detect heterozygous beta(β) thalassaemia trait in the siblings (brothers & sisters) of known β-thalassaemia major children by haemoglobin (Hb) electrophoresis.*

Material and Methods: *This prospective analytical study was conducted at Postgraduate Medical Institute Lady Reading hospital Peshawar. Subjects were divided into two groups. Group:1 included 60 families consisting of 120 subjects (siblings) of known B-thalassaemia major children as patients and Group:2 included 15 families consisting of 30 children as control group. HBA2 was determined by electrophoresis elution method. The criteria used to fit the diagnosis of beta thalassaemia trait were HbA_2 level of $\geq 3.6\%$.*

Results: *Out of 120 siblings of known beta thalassaemia major children, 73(60.83%) were detected as β-thalassaemia trait. Three (10%) subjects were identified as beta thalassaemia trait in control group.*

Conclusion: *This study shows that beta thalassaemia trait is more common in the siblings of beta thalassaemia major children as well as it is not uncommon in normal families.*

Key Words: *Beta Thalassaemia Major, Siblings, Beta Thalassaemia Carrier (Trait).*

INTRODUCTION

Thalassaemia is the commonest inherited disorder present world wide.[1] Beta thalassaemia is the most common single gene disorder in Pakistan.[2] There would be approximately 8-10 millions trait (carrier) in Pakistan on average of 5–8 % as the national carrier rate.[2,3] It has become a world wide problem due to increasing immigration of ethnic groups with the high prevalence of thalassaemia.[4] The management of homozygous state by blood transfusion and iron chelation therapy has prolonged the survival of these children.[5, 6] However, the cost of this kind of management for all thalassaemic children is higher than the current health related expenditure of government.[7,8] Under these circumstances the best approach will be to prevent the birth of children with thalassaemia major. The single most effective method of prevention is pre-marital screening and genetic counseling. This requires population screening for beta thalassaemia trait.[8,9] It is heterozygous state which may present with chronic anaemia along with splenomegaly and skeletal changes to an almost symptomless state. Clinically these subjects may present with feature of iron deficiency anaemia and if iron is given they develop iron over load.[10-12] It is important to exclude iron deficiency anaemia in which HbA_2 is reduced or normal.[13] In the developed countries much attention has been directed to the prevention of disease by detection of thalassaemia trait and marriage counseling.[14] By using this prevention program in Sardinia the incidence of homozygous patients decreased from 1:250 live births to 1 :1000 live births.[14] The present study was mainly designed to detect the incidence of beta thalassaemia trait in the siblings (brothers and sisters) of the Beta thalassaemia major children.

AGE AND SEX DISTRIBUTION IN B–THALASSAEMIA TRAIT SUBJECTS (n=73)

Age (years)	Sex		Total	Percentage
	Male	Female		
1 - 9	12	13	25	34.25
10 - 19	16	14	30	41.10
20 - 29	8	10	18	24.65
Total	36	37	73	100%

Table 1

MATERIAL AND METHODS

This was a prospective analytical study and carried out in the Pathology department of Postgraduate Medical Institute Lady Reading Hospital Peshawar. Those families which had beta thalassaemia major child were included in this study. A total of 60 families comprising of 120 siblings of the affected families were selected as group 1. Fifteen normal families having 30 normal children who had no beta thalassaemia major child in the family were taken as control subject Group:2. Both groups belong to district Peshawar. All were thoroughly examined for jaundice, hepatosplenomegaly and lymphadenopathy. The relevant information regarding age, sex, ethnic origin (caste/tribe) and places of birth / inhabitance, family history and consanguinity were recorded in proper proforma and the data was analyzed on SPSS version 10.0. Seven ml of venous blood was collected from each of the subject through a clean venipuncture and distributed in the following manner. Two 2.0 ml blood was mixed with potassium EDTA used for blood counts, and red cell indices and retic count. Two 2.0 ml blood was mixed with potassium EDTA for estimation of HbA_2 and HbF. Three 3.0 ml blood allowed to clot. Clear serum preserved at -20C for estimation of serum ferritin.

All complete blood count and red cell indices were measured on Micros cobas 16 haematology auto analyzer with in two hour of sample collection. Estimation of HbA_2 was done spectrophotometrically after elution of the haemoglobin bands.[15] HbF was measured by modified Betke method.[16] Serum ferritin was estimated by ELISA method. Reticulocyte count was performed by supra vital staining.

The criteria used for the diagnosis of β-thalassaemia trait was haemoglobinA_2 level of $\geq$ 3.6.

RESULTS

A total of 73 cases (60.83%) of heterozygous beta thalassaemia were detected in survey of 60 families of the known β thalassaemia major children. Maximum cases 41.10% were detected in the 10 - 19 years of age group (table.1), followed by 1 - 9 years of age (34.25%).

In most of the children (78.25%) HbA_2 was in the range of 3.6 - 6 %, while 15.25% subjects of beta thalassaemia trait had HbA2 level 6.1 – 7% and only 6.50% of these subjects had HbA2 level in the range of 7.1 – 9.0%.

In the control subjects three persons out of thirty had HbA_2 level of more than 3.6% and they were the beta thalassaemia trait cases.

In most of the cases HbF was below 1% while only 7.58% subjects had HbF of more than 1%. In the control group all subject had HbF level of less then 1%.

Haemoglobin A_2 level ranged from 3.6 – 9.0%, MCV of less than 75 fl was found in 100% and MCH of 24.4 pg or less in all subjects of beta thalassaemia trait (Table 2).

Altered red cell morphology such as hypochromasia, microcytosis and anisopoikilocytosis were seen in all the cases. The frequency of consanguineous marriage in parents were first cousin 40%, second cousin marriage 16.67%, distant cousin 15.0% and non relative in 28.33% (Table 3).

DISCUSSION

Beta thalassaemsia trait is probably the commonest inherited haemtological disorder in Pakistan.[2,3] It is the second most common cause of hypochromic microcytic anaemia.[17] Hereditary disorders in Pakistan have been marked by infections and nutritional deficiencies. If this country overcomes these acquired diseases successfully, hereditary disorders including Haemoglobinopathies would become important national problem.[18] In the present study, out of 120 siblings of known beta thalassaemia major children 73 (60.83%) were detected to have -thalassaemia trait. The prevalence of beta thalassaemia carrier

HAEMATOLOGICAL DATA IN SUBJECTS HAVING HETEROZYGOUS BETA THALASSAEMIA AND CONTROL SUBJECTS

Figures in parentheses shows the range values

Haematological Parameters	Beta Thalassaemia minor (n =73)	Healthy Population (n = 27)	P values
Hb (g/dl)	10.20 ± 1.55	12.10 ± 1.70	HS
TRBC (x 1012/L)	(6.1 - 12.5)	(11.5 - 14.5)	
PCV (L/L)	4.94 ± 0.84	5.01 ± 0.50	HS
MCV (fl)	(3.95 - 6.80)	(4.05 - 6.05)	
MCH (pg)	35.8 ± 7.21	40.0 ± 6.3	HS
MCHC (g/dl)	(21.10 - 45.60)	(37.00 - 46.00)	
HbF (%)	61.73 ± 4.55	80.2 ± 8.50	HS
HbA2(%)	(48.3 - 74.6)	(76 - 91)	
Retic (%)	20.33 ± 3.76	26.2 ± 2.70	HS
Serum ferritin	(15.4 - 24.4)	(26.8 - 29.9)	
(ng/ml)	23.12 ± 4.45	30.5 ± 3.50	S
	(22.7 - 38.0)	(30.2 - 36.2)	
	0.89 ± 0.25	0.9 ± 0.22	NS
	(0.6 - 1.4)	(0.6 - 1.3)	
	5.00 ± 0.57	3.0 ± 0.27	HS
	(3.6 - 9.0)	(2.5 - 3.4)	
	1.8 ± 0.66	1.60 ± 0.66	S
	(0.8 - 4.0)	(0.8 - 2.5)	
	43.0 ± 18.5	74.0 ± 30.5	HS
	(10 - 104)	(16 - 136)	

Table 2

Key

HS=Highly significant (P< 0.001)

S=Significant (P< 0.05)

NS =Non- Significant (P > 0.05)

N=Number of subjects

state found in the relatives of beta thalassaemia major children is more than 50% reported in a study from India.[19] A study from Pakistan revealed the incidence of beta thalassaemia trait among the siblings of beta thalassaemia major children were 58%.[20] Incidence of beta thalassaemia trait in parents and grand parents were69.17% documented in other study from Pakistan.[21] The incidence has closely followed the Mandelian pattern of inheritance.[22]

In the Fars province of Iran screening programme for beta thalassaemia trait before marriage reduces the prevalence of this genetic disorder[23]. Similarly is Cyprus and Sardinia the incidence of thalassaemia major cases dropped by 96 %.[14,24] The consanguineous marriages are quite common in Pakistan and especially in Pathans. In the present study 71.67% families showed history of consanguinity. This social practice may have compounded the problems, the carrier rate of beta thalassaemia is high in the northern area 8.0% in Pathan.[25]

Haemoglobin A_2 level of 3.6% or more appears to be highly significant level for the diagnosis of heterozygous B-thalassaemia in the

FREQUENCY OF CONSANGUINEOUS MARRIAGES IN 60 FAMILIES

Relationship	No. of Families	Percentage
First cousin marriage	24	40.0
Second cousin marriage	10	16.67
Distant cousin marriage	9	15.00
Non relative marriage	17	28.33
Total	60	100.0

Table 3

present study. This was supported by low MCV, low MCH, raised and abnormal red cell morphology.[20] Similar results were obtained when same red cell indices with red cell count were used in 463 beta thalassaemia trait 195 iron deficiency anaemia patients in the study carried out in new Dehli India.[26]

The parent at risk can be informed about the management and treatment of thalassaemia major. A population base screening programme involving a large sample size can be used to find the exact incidence and to asses the magnitude of this problem on a regional or national scale.[27] Marriage counseling in these carriers is important to reduce the incidence of beta thalassaemia major cases as has been carried out in the developed countries.[28,29]

CONCLUSION

This study shows that beta thalassaemia is very common in the siblings of beta thalassaemia major childrens, more over it is not un common in normal population. Therefore it is recommended to have a mass screening programme for detection of beta thalassaemia trait in general population and in pre marital couples.

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