Original Article

Beta Thalassaemia

Evaluation of Patterns of Beta Thalassaemia in Children with Microcytic Hypochromic Anemia

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ABSTRACT

Objective: The aim of this study was to evaluate the patterns of Beta Thalassaemia in children suffered with microcytic hypochromic anemia in the different hospitals of Hyderabad region.

Study Design: Cross sectional study.

Place and Duration of Study: This is an experimental based research study that was conducted at Isra University Hospital, Hyderabad and Liaquat University of Medical and Health Sciences Hospital Jamshoro Hyderabad from June 2011 to February 2012.

Materials and Methods: Hundred (100) children from either gender attending outpatient department of Isra University Hospital and Liaquat University Hospital Hyderabad were selected for this study. Whole blood sample (10ml) was collected from each children. Peripheral smears were made and found that all children have suffered with findings of microcytic hypochromic anemia. Children with other than microcytic hypochromic anemia were excluded from this study. All parents of children were interviewed regarding family history and marriages with cousin and other findings or histories were also recorded on a well designed performa.

Results: Whole blood samples were analyzed for the evaluation of the patterns of Beta Thalassaemia in 100 children who were affected with microcytic hypochromic anemia. Out of 100 children, 08 of them were diagnosed as having beta thalassaemia minor, one as Beta Thalassaemia major and the remaining 91 were non thalassaemics. Among 08 Beta Thalassaemic minor children, 05 were females and 03 were males and only one male child was suffered with beta thalassaemia major.

Conclusion: It is concluded that the prevalence level of beta thalassaemia is higher in Hyderabad, which is an alarming. Many factors such as poor facilities for diagnosis, lack of awareness among clinicians, consultants and pathologists and high cost of molecular diagnosis play a contributory role in the propagation of the beta thalassaemia in the Pakistani population. These factors become a serious hindrance for the prevention of thalassaemic program in Pakistan.

Key Words: Beta-thalassaemia, Complete Blood Count (CBC), Peripheral Blood Smear, Cellulose Acetate Haemoglobin Electrophoresis.

INTRODUCTION

Thalassaemia is the most common monogenic disorders across the world[1]. One of the severe form of anemia occurring in childhood associated with organomegaly and characteristic bony deformities has been clearly documented[2,3]. Thalassaemias are a heterogeneous group of genetic disorders of haemoglobin synthesis, that result from a reduced rate of production of one or more of the globin chain(s) of haemoglobin[4,5]. Basic molecular defect in thalassaemia is mutations in the globin gene. These defects are numerous and include deletional or non-deletional mutations. Mutations usually have a geographic and ethnic distribution in each specific population[6]. Clinically thalassaemias are divided into alpha, beta, delta beta, or gamma delta beta thalassaemias according to the type of chains affected. Basically, there are many types of thalassaemias that are based according to the types of globin chains. The most clinically relevant types are alpha and beta-thalassaemia[7].

Pathophysiology of beta thalassaemia that is caused by molecular defect in beta globin gene results in absent or inadequate production of beta globin chains leading to premature haemolysis that cause a different clinical severity of beta thalassaemia[8]. Beta thalassaemia minor (β thalassaemia trait) carriers are generally asymptomatic, but may be suffered from anemia during physiological conditions such as childhood, pregnancy and stress. Beta thalassaemia major which is also known as Cooley's anemia, is a fatal condition that is associated with severe haemolytic anemia, jaundice, organomegaly, bone deformities and requires lifelong regular blood transfusion. While beta thalassaemia intermedia is a clinical condition which is intermediate between beta thalassaemia major and beta thalassaemia trait[9].

Although nutritional anemia are still very common but thalassaemia is the most common inherited disorder in Pakistan, resulting in considerable morbidity and Mortality[10]. Pakistan has a population of 160 million people. The annual rate of population growth is 3% and almost 40% of the population is below 15 years of age. Due to strong cultural preference for consanguineous marriage in Pakistan it is estimated that there is relatively high prevalence of inherited disorders[10,11]. The carrier frequency of beta thalassaemic gene is estimated to be around 6% in Pakistani population[12]. Therefore present study is designed to evaluate the patterns of thalassaemic syndrome such as beta thalassaemia minor, major and intermedia in anemic children at Hyderabad, Sindh. This study will help to create awareness about thalassaemia among pathologists, clinical physicians in the rural and urban areas to facilitate thalassaemia prevention program occurring in this district.

MATERIALS AND METHODS

This study was carried out to assess the patterns of beta thalassaemia in children with microcytic hypochromic anemia in the population of Hyderabad Sindh, Pakistan. A total number of hundred children were selected in this study and was carried out from the period of June 2011 to February 2011. All children presented in outpatient department of Isra Hospital and Liaquat university hospital Hyderabad, Pakistan were included. The parents of children were interviewed and detailed information was taken regarding the sign and symptoms, family history, marital status, marriage patterns particularly closed family marriages. All the information was kept and recorded in Performa. A prior permission was obtained from the outpatient department Incharge and the ethical committee of the hospital for interview. Whole blood sample was collected by venepuncture technique and following tests were performed such as Complete Blood Count, Morphological examination of blood smear, Cellulose Acetate Haemoglobin Electrophoresis.

Inclusion criteria: Children having microcytic hypochromic anemia and peripheral smear findings suggestive of microcytosis and hypochromia.

Exclusion criteria: Children with anemia other than microcytic hypochromic.

Sample collection: Whole blood sample (10ml) was collected by venepuncture from all subjects recruited in the study. For Complete blood count and Peripheral smear 7ml of blood was transferred to a tube containing EDTA anticoagulant and another 3ml was transferred to another EDTA containing tube for the determination of Cellulose Acetate Haemoglobin Electrophoresis.

RESULTS

Whole blood samples from hundred (100) subjects of either sexes were analyzed for the evaluation of microcytic hypochromic anemia. Out of total eight subjects diagnosed as having beta thalassaemia minor, one was beta thalassaemia major and the remaining 91 were non thalassaemics. Among 08 beta thalassaemia minor, 05 were females and 03 were males and only one male was beta thalassaemia major, Fig 1.

Table No.1: Basic haematological parameters in female beta-thalassaemia minor

Haematological parameters	Mean	Std. Deviation
Hb (g/dl)	9.50	0.87
RBC (millions/µl)	4.84	0.48
MCV (fl)	66.50	5.10
MCH (pg)	19.78	1.38
MCHC (g/dl)	29.74	0.96

Table 2: Basic haematological parameters in male beta-thalassaemia minor

Haematological parameters	Mean	Std. Deviation
Hb (g/dl)	10.73	0.40
RBC (millions/µl)	5.17	0.18
MCV (fl)	67.06	2.28
MCH (pg)	20.53	0.92
MCHC (g/dl)	30.60	0.30

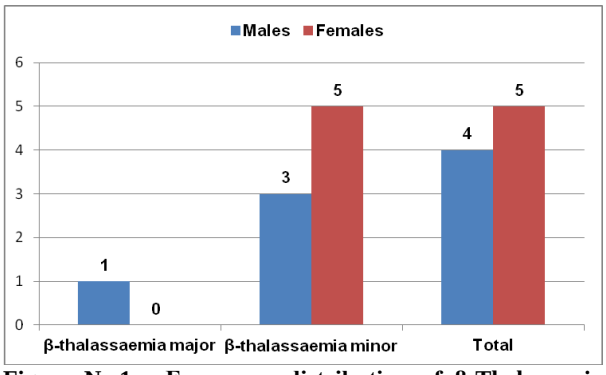


Figure No.1: Frequency distribution of β-Thalassemia Major, Minor subjects in male and female

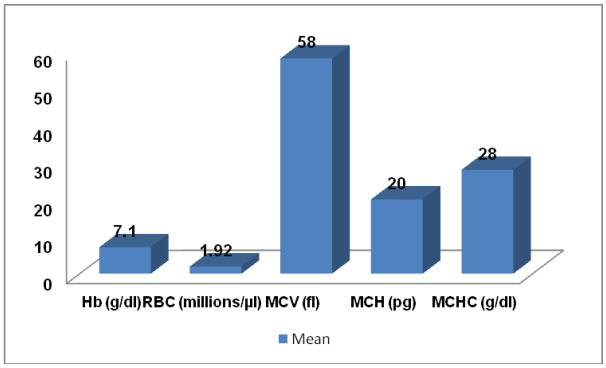


Figure No. 2: Basic haematological parameters in male beta-thalassaemia major

In the present study different heamatological parameters were carried out for the diagnosis of different patterns of beta thalassaemia. In case of beta

thalassaemia female subject, the mean hemoglobin (Hb) level was 9.50±0.87 (g/dl), red blood cell count (RBC) 4.84±0.48 (millions/µl), Mean Corpuscular Volume (MCV) 66.50±5.10 (fl), Mean Corpuscular Hemoglobin (MCH) 19.78±1.38 (pg) and Mean Corpuscular Hemoglobin Concentration (MCHC) 29.74±0.96 (g/dl) were noted (Table 1).

In beta-thalassaemia minor male subjects the haemoglobin (Hb) 10.73±0.40 (g/dl), red blood cell count (RBC) 5.17±0.18 (millions/µl), Mean Corpuscular Volume (MCV) 67.06±2.28 (fl), Mean Corpuscular Haemoglobin (MCH) 20.53±0.92 (pg) and Mean Corpuscular Haemoglobin Concentration (MCHC) 30.60±0.30 (g/dl) were found respectively (Table 2).

In beta-thalassaemia major male the haemoglobin (Hb) 7.10 (g/dl), red blood cell count (RBC) 1.92 (millions/µl), Mean Corpuscular Volume (MCV) 58.0 (fl), Mean Corpuscular Haemoglobin (MCH) 20.0 (pg) and Mean Corpuscular Haemoglobin Concentration (MCHC) 28.0 (g/dl) were found (Fig 2).

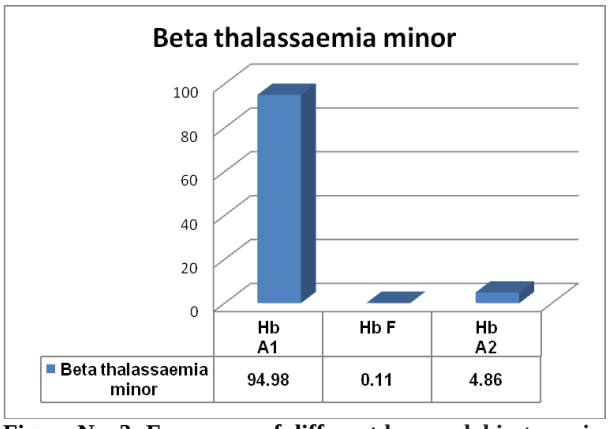
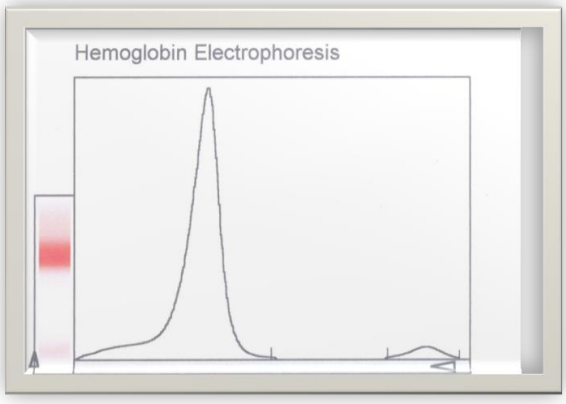


Figure No. 3: Frequency of different haemoglobin types in Beta Thalassemia minor subjects



Hb A1 (95.4) Hb F (0.1) Hb A2 (4.5)

Figure No.4: Hb-electrophoresis of beta-thalassaemia minor

Similarly in case of beta thalassaemia minor subjects haemoglobin-electrophoresis showed haemoglobin A 94.98±0.46 (g/dl), haemoglobin F 0.11±0.35 (g/dl) and haemoglobin A2 4.86±0.46 (g/dl) as shown in Fig 3. In case of beta thalassemia major, the Hb electrophoresis showed 2.3% Hb-A, (Fig 4) 94.6% Hb-F and 3.1% Hb-A2 respectively (Fig 4).

DISCUSSION

Thalassaemia has become a worldwide clinical problem due to increased immigration of ethnic groups with high prevalence of thalassaemia[13]. Pakistan is one of the country included in the "thalassaemic belt with highest prevalence of thalassaemia[14]. The carrier frequency of beta thalassaemia is estimated around 6% in Pakistani population. Other provinces of Pakistan including Sindh, Baluchistan and Khyber Pakhtunkhwa the prevalence of beta-thalassaemia is most common[15].

Married couples consisting of two carriers have 25 percent chances that any child they have will be affected. The estimated rate of birth of affected infants is 1.3 per 1000 live births and 5000-9000 children with beta thalassaemia major born per year, although no scientific literature is documented till now in Pakistan[16,17].

Recent studies have suggested that low-income status and lack of awareness, preference to marry within the ethnic groups and consanguineous marriages are contributed to the increased incidence of this disease in the Pakistani population[17,18].

In the present study prevalence of beta thalassaemia minor is 8 % in hundred of microcytic hypochromic children which is higher, because of selected microcytic hypochromic population. Beta thalassaemia and iron deficiency anemia are the most common microcytic hypochromic anemia in Pakistan. Resources of screening beta thalassaemia minor before marriages are minimal in Hyderabad so higher frequency of beta thalassaemia trait has been noticed in this city. This result is consistent with a study reported in India 2003[19]. They also reported that the prevalence of the beta thalassaemia trait in the population of Punjab is 5.5%. Another study was reported that also showed highest frequency of β-thalassaemia trait in Gujarat, followed by Sindh, Punjab, Tamil Nadu, South India and Maharashtra[20].

The red cell indices is an important diagnostic tool for the assessment of beat thalassaemic trait. In such cases MCV and MCH are low while MCHC is normal. Red cell count is often more than 5-0×10^7/µl. In the present study initially red blood cells indices and peripheral blood smear were performed which also showed that all beta thalassaemia minor subjects have MCV less than 67.06 fl, MCH less than 20.53 pg and MCHC was found almost normal. Additionally the peripheral smear of all subjects showed altered red cell morphology such as hypochromia, microcytosis and anisopoikilocytosis. This present study is almost in line with another study

conducted in Lahore to know the significance of red cell indices in the diagnosis of beta thalassaemia trait[21]. Similarly a study conducted that also showed the basic haematological parameters where MCV was less than 77fl and MCH less than 26.4 pg in all the cases of thalassaemic minor subjects the finding was almost similar with the present study[22].

Hb-electrophoresis is another tool for the final diagnosis of beta thalassaemia In this study Hb-electrophoresis were also performed only in those subjects whose red cell indices and peripheral blood smear were suggestive of beta thalassaemia. The results showed that haemoglobin A2 level of all beta thalassaemia minor subjects have showed 4% or more which appears to be highly significant for the diagnosis of heterozygous beta thalassaemia. This finding is also in concurrence with previous study where MCV value was 77 fl or less and haemoglobin A2 4% or more was taken as the primary screening tool for the diagnosis of beta thalassaemia trait[23]. Similarly another study also conducted that showed the characterization of beta thalassaemia minor, when Hb A2 is 4.0– 6.0% [24] while in the present study the mean level of Hb A2 in beta thalassaemia minor was 4.86±0.46.

In rural areas of Hyderabad, where there is deficiency or lack of latest equipment for the diagnosis of beta thalassaemia. So provisional diagnosis can be done on simple morphological criteria, which is based on microcytic red cells, target cells and basophilic stippling on peripheral blood smear. Facility for Hb electrophoresis is not available at many places in Hyderabad and also high cost makes a refusal for poor patients to reach at final diagnosis. Red cell indices given by automated cell counters can be reliably used in these areas to differentiate beta thalassaemia trait from other microcytic hypochromic anemia.

CONCLUSION

It is concluded from this study that prevalence of beta thalassaemia is higher in Hyderabad Sindh, which is an alarming situation. This fact is not neglectable, as many factors such as poor facilities for diagnosis, lack of awareness among clinicians and pathologists and expensive cost of molecular diagnosis, are contributing in the propagation of the beta thalassaemia gene in the Pakistani population. These factors become a serious hindrance for the thalassaemia prevention program in Pakistan. Furthermore there is an urgent need to bridge this wide gap of diagnostic facilities for every patient at low cost value so that they easily access medical care.

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