Original Article

Prevalence of Haemoglobinopathies in Children Presenting with Anemia at DHQ Hospital Zhob, Balochistan

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ABSTRACT

OBJECTIVE: To find the prevalence of haemoglobinopathies in children presenting with anemia at District Headquarter Hospital Zhob.

METHODOLOGY: The cross-sectional study was carried out at District Headquarter Hospital (DHQ) Zhob from March to August 2021. Patients aged 6 months to 15 years presented with signs and symptoms of anemia were included in the study and investigated for haemoglobinopathies throughcomplete blood count, peripheral smear, iron studies (serum ferritin), haemoglobinanalysis by haemoglobinelectrophoresis, high-performance liquid chromatography and sickling test. All the data, including age, gender, and diseases was recorded and analyzed with the help of a statistical package for social sciences (SPSS 21).

RESULTS: A total of 2258 children were clinically suspected of anemia and were advised the sequential blood tests for diagnosing Haemoglobinopathies. Out of these, 239 cases lost to follow up. The remaining 2019 caseswere included, comprising1026 males (50.81%) and 993 females (49.18%) with a mean age 8.09±3.78 years. Haemoglobinopathies constituted 8.9%ofpediatric diseases and 56.7% of anemia cases. The most common haemoglobinopathy was beta-thalassemia major (29.4%), followed by sickle / beta-thalassemia compound heterozygous (28.3%), sickle cell disease (28%), HbH"alpha thalassemia" (2.8%), HbD trait (2.3%), HbE/Beta-thalassemia (2%), HbD Homozygous (1.8%), HbD/beta-thalassemia (1.5%), HbE trait (1.1%)and Beta-thalassemia minor (0.9%). There were 11 cases (1%) reports labeled as borderline.

CONCLUSION: Haemoglobinopathies constitute a major portion of diseases and the most common cause of anemia in the pediatric population of Zhob.

KEYWORDS: Haemoglobinopathy, anemia, Thalassemia, Vitamin B12, Folate, Iron deficiency anemia

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INTRODUCTION

Haemoglobinopathies denote genetic defects collectively affecting either Haem structure or synthesis[1]. They are divided into two categories. First are those that affect the structure of hemoglobin (Hb) and produce abnormal Hb structural variants usually due to substitution of one amino acid by another for example hemoglobin C, S, D and E Second types, called thalassemia's, primarily affects the rate of synthesis of one of the globin chains causing an imbalance in the production of Hb subunits[2]. Haemoglobinopathies are the most common genetic diseases of the world, with an estimated 7% prevalence[3]. Thalassemia and sickle cell disease are the commonest hemoglobinopathies[4]. Approximately 5% of the world population carries a gene for thalassemia or sickle cell disease, and in few regions, it could be as high as 25%[5].

Haemoglobin is made up of four sub-units; each of the sub-units consists of one heme group and one polypeptideglobin chain. The polypeptideglobinchain of adult hemoglobin (Hb A) contains two alpha and two beta chains. These chains are similar in length but differ in the sequence of amino-acids. Fetal Haemoglobin (Hb F)contains two alpha and two gammaglobin chains. Thalassemia occurs due to quantitative aberrations in globin chain synthesis. Alpha thalassemia eventuates from deficient alpha-globin chain synthesis, and beta-thalassemia occurs due to deficient beta globin chain synthesis. In a newborn, levels of Hb F which is the predominant type of Hb, gradually decreases and reach adult levels (<1% of total Hb) by the end of the first year of life as Hb A begin to be produced. In beta thalassemia's which affects the making of Hb A, this transformation of HbF to Hb A is disturbed and cause Hb F to remain significantly higher than normal. Thalassemia's are characterized by ineffective erythropoiesis, hemolytic anemia, iron overload and associated complications. On the other hand, sickle hemoglobin (Hb S), a structural variant, occurs due to mutation in the gene for the beta-globin chain resulting in the replacement of glutamic acid with valine at the sixth amino acid

position of the beta-globin chain. This alters the red cell membrane and normally a flexible round red blood cell changes into a fragile elongated crescent shaped cell there by reducing the red cell life[6]. Sickled cells often form aggregates, causing a microvascular obstruction which leads to painful crisis, emboli, osteonecrosis, and multi-organ damage[6].

The prevalence of beta-thalassemia is reported to be 5-8% world-wide and in Pakistan[7]. While limited data is available about the prevalence of sickle cell disease in Pakistan[8]. The majority of data available which has been reported by laboratories from main cities, are far from its prevalent regions. In such regions compound heterozygosity of beta thalassemia with a sickle or other thalassemia is not unusual. This results in a complex spectrum of genetic and clinical disorders[2]. Several gene mutations and polymorphisms confer protection against malaria, e.g. sickle cell trait, alpha thalassemia, G6PD (glucose-6-phosphate dehydrogenase) deficiency. As a result, the prevalence of these haemoglobinopathies is very high in malaria-endemic areas[9].

Haemoglobinopathies are a great challenge for the health care system and affected families. Early detection and characterization of haemoglobinopath-ies areof utmost importance so that families can be counseled and warned of serious consequences/ complications associated with haemoglobinopathies which include iron overload, recurrent infections, cirrhosis, endocrine failure, pulmonary hypertension, hepatobilliary cancers, arthropathies, transfusion related complications and early death[10].These cases require lifelong care, and the health care system should provide adequate diagnostic, pre-natal/pre-marital screening and treatment facilities in the prevalent areas.

The present study aims to find out the prevalence of haemoglobinopathies in the Northern region of Balochistan. Zhob is a big district with a population of 310544, located in Northern Balochistan abutting Khyber Pakhtunkhwa (neighboring province) and Afghanistan. The district headquarters hospital Zhob is the only major health facility in the whole area and provides health care to the population of Zhob and surrounding areas. This wouldbe of beneficial in formulating policies for management and prevention of thalassemia's and associated Hb (haemoglobin) variants in farthest parts of the country.

METHODOLOGY

This cross sectional study was carried out in the pediatric department of District Headquarter Hospital (DHQ) Zhob from March to August 2021. Total 12739 patients were seen/checked in the pediatric department of hospital during study period. Permission was obtained from the ethical review board of DHQ hospital for subject research. Consent of parents or guardians was acquired for including the cases in the subject study. A convenient sampling technique was used for this study.

Patients aged between 6 months to 15 years presented with signs and symptoms ofanemiai.e pallor, irritability, failure to thrive etc, Patients who had a family history of haemoglobinopathies, Patients in the specified age bracket, who were already diagnosed as cases of haemoglobinopathies were included. The cases who were lost on follow-up or whose parents did not consent for inclusion in the study were excluded.

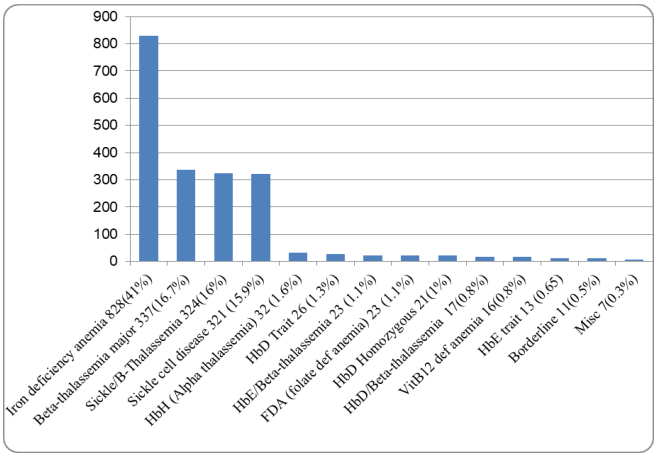
Patients aged between 6 months to 15 years were assessed for signs and symptoms of anemia and haemoglobinopathies like family history, conjunctival pallor, irritability, hepatosplenomegaly and failure to thrive. This was followed by initial laboratory investigations like complete blood picture with red cell indices; TRBC (total red blood cell count),Hb (haemoglobin), MCV (mean corpuscular volume),MCH (mean corpuscular haemoglobin), & RDW(red cell distribution width) and peripheral smear examination. If Hb (haemoglobin) was less than 10g/dl they were further investigated for anemia or haemoglobinopathies accordingly. Children were tested further with iron studies (serum ferritin) and haemoglobin analysis by haemoglobin electrophoresis, high-performance liquid chromatography and sickling test. Complete blood picture including red cell indices like TRBC, Hb, MCV, MCH, RDW and peripheral smear were done in DHQ Hospital Zhob laboratory. For the rest of the tests and haemoglobin studies, patients were directed to various reference pathology laboratories for investigations. On examination of blood peripheral films, the cases with a high MCV (mean corpuscular volume) levels were tested further, and serum vitamin B12 and folate levels were carried out to diagnose these two varieties of anemia. Patients and their attendants were interviewed once they returned back; all the figures were collected and analyzed accordingly. All of the data including age, gender, disease (type of haemoglobinopathy), was recorded and analyzed with the help of a statistical package for social sciences (SPSS 21). Percentages were used to express the frequencies.

RESULTS

A total of 12739 children were seen/checked in the pediatric department of DHQ hospital during the study period. A total of 2258 children were clinically suspected of anemia and were advised the sequential blood tests for diagnosing haemoglobinopathies. Out of these, 239 cases were lost to follow up. The remaining 2019 cases (15.8%) were included. There were 1026 males (50.81%) and 993 females (49.18%).The age range was from 6 month to 15 years with a mean age of 8.09±3.78 years. The prevalence of various types of anemia in pediatric cases is shown in **Figure I**. The most common cause

of anemia was iron deficiency anemia (41%), followed by beta-thalassemia major (16.7%), sickle/beta-thalassemia (16%), sickle cell disease (15.9%).

FIGURE I:
PREVALENCE OF VARIOUS TYPES OF ANEMIA



Total 1145 (9.2%) children were diagnosed as cases of various haemoglobinopathies. Frequencies of haemoglobinopathies are shown in **Table I**. The most common cause was beta-thalassemia major (29.4%), followedbysickle/ beta-thalassemia compound heterozygous (28.3%), sickle cell disease (28%), HbH"alpha thalassemia" (2.8%), HbD trait (2.3%), HbE/ Beta-thalassemia (2%), HbD Homozygous (1.8%), HbD/ beta-thalassemia (1.5%), HbE trait (1.1%) and Beta-thalassemia minor (0.9%). There were11 cases (1%) which were border-line but clinically diagnosed cases of haemoglobinopathies. These were labeled "undetermined".

TABLE I: FREQUENCY OF HAEMOGLOBINOPATHIES (n=1145)

Haemoglobinopathy (Disease)	n=1145	Percentage
Beta thalassemia major	337	29.4%
Sickle/ beta thalassemia compound heterozygous	324	28.3%
Sickle cell disease	321	28%
HbH (Alpha thalassemia)	32	2.8%
HbD trait	26	2.3%
HbE/Beta-thalassemia	23	2%
HbD Homozygous	21	1.8%
HbD/beta-thalassemia	17	1.5%
HbE Trait	13	1.1%
Beta-thalassemia minor	10	0.9%
Sickle cell trait	10	0.9%
Borderline	11	1%
Total	**1145**	**100%**

Total 874 children (6.9%) were suffering from other causes of anemia as mentioned in **Table II.** The most common cause of anemia (other than haemoglobinopathy) was iron deficiency anemia (94.7%), followed by folate deficiency (2.8%) and vitamin B12 (cyanocobalamin) deficiency (1.9%). There were seven cases (0.8%) which had miscellaneous causes, including celiac disease, liver and renal disorders etc.

TABLE II: OTHER CAUSES OF ANEMIAIN THE PEDIATRIC PATIENTS PRESENTING AT THE DHQ HOSPITAL ZHOB (n=381)

Disease	n=874	Percentage
Iron deficiency anemia	828	94.7%
Folate deficiency anemia	23	2.8%
Vitamin B 12 deficiency	16	1.9%
Misc	7	0.8%
Total	**874**	**100%**

The population of Zhob is 310544 according tothe 2017 census of Pakistan. Under 15 years aged population has been reported 46.67% according to Pakistan Bureau of Statistics[11]. The prevalence of haemoglobinopathies was under 15 years population of Zhob is calculated to be 0.79% as;

Prevalence in haemoglobinopathies (<15 years population) = 1145 / 310544 X 46.67%
= 0.79%
= 79/10000 of <15 years children

DISCUSSION

Our study demonstrated frequencies of haemoglobinopathies in DHQ hospital Zhob, identifying iron deficiency anemia as the most common cause of anemia (41%) followed by beta-thalassemia major (16.7%), sickle/beta-thalassemia (16%), sickle cell disease (15.9%).

Various researchers have reported frequencies of haemoglobinopathies in Pakistan. Waheed U 2012[12] reported the frequency of various haemoglobinopathies to be 28.4% in patients who presented at Islamabad. Among them, 25.6% had thalassemia trait, 1.4% had thalassemia major,1.4% had sickle cell disease. Shabbir S. et al. [13] studied the pattern of haemoglobinopathies on laboratory samples at Karachi and reported 34.2% cases to be affected with haemoglobinopathies. Out of these, 51.8% had beta-thalassemia minor, 24.1% had beta-thalassemia major, 6.7% had HbD trait, 4.5% had sickle/beta-thalassemia, and 3.9% had sickle cell anemia. Hussain J 2015[4]studied frequencies of haemoglobinopathies at the Department of Pathology Gomal Medical College Dera Ismail Khan (Khyber Pakhtunkhwa). They found beta-thalassemia major to be the most common haemoglobinopathy (25.8%), followed by beta-thalassemia trait (11.3%), sickle cell disease (9.7%), sickle cell trait (3.2%), and sickle/beta -thalassemia "double heterozygous" (3.2%) (4).

Ghani R 2002[14] reported 20.6% patients of beta-thalassemia major, 13% beta thalassemia trait, 5.1% of sickle cell disease, 0.76% haemoglobin D (HbD Punjab).

Arab countries also have a high prevalence of these haemoglobinopathies; the reported carrier rate of beta -thalassemia is 1-11%, 1-58% for alpha thalassemia and 0.3-30 % for sickle cell trait[13]. This high rate has been attributed to consanguineous marriages[15]. Similarly, Southern parts of Iran also had a high prevalence of haemoglobinopathies (thalassemia's); 35% for alpha thalassemia and 10% for beta-thalassemia[16].

The majority of these above-mentioned frequencies of haemoglobinopathies in Pakistan are from laboratories of Karachi, Islamabad and Dera Ismail Khan because majority of reference laboratories are located in main cities. However, our study stands out as we have reported the frequencies and prevalence of these haemoglobinopathies from one of the areaswhere they are extensively present. In our research, we found 8.9% of children (checked during the whole year in the pediatric department of the hospital) to be affected with haemoglobinopathies. The prevalence of haemoglobinopathiesin under 15 years old population of Zhob was 0.79%. This prevalence reflects the affected children with haemoglobinopathies. The carrier rate of haemoglobinopathies would be much higher. The undetected carriers are a constant source of homozygous haemoglobinopathies in future generations after inter-marriages. This will help the policy makers of thalassemia control program to redirect their efforts towards these neglected areas to address haemoglobinopathies. The management of haemoglobinopathies includes not only treatment of anemia and iron chelation but also some definitive therapeutic modalities like bone marrow transplants. Special emphasis should be on the prevention of disease which must include pre-marital and extended family screening with facilities in place regionally for prenatal diagnosis.

We recommend that pre-marriage and pre-natal counseling/screening should be made a strict law in such areas. Cyprus was the first country to follow these programs, and they have successfully curtailed the birth of children with these disorders to zero[17]. Education of health professionals and the population and enhancing awareness through print and electronic media and influential people isalsoessential to prevent these genetic disorders.

CONCLUSION

Haemoglobinopathies constitute a major portion of diseases and the most common cause of anemia in the pediatric population of Zhob. Early detection and management of such cases should be done to slow down the progression of haemoglobinopathies to avoid complications. Affected families should be counseled for eventual consequences/outcomes. Pre-marital and pre-natal screening should be made a law.

Ethical permission: District Head Quarters Hospital, District Zhob, Balochistan, Medical Superintendent, letter No. 1137, dated 18-03-2021.

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AUTHOR CONTRIBUTIONS

Akram S: Main author & manuscript writing
Khattak SA: Review of manuscript
Khan MA: Script writing
Ali SA: Data analysis
Khan MA: Data analysis & review

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