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ORIGINAL ARTICLE

Aberrant Heterosis In Hemoglobinopathies With Special Reference To β-Thalassemia And Structurally Abnormal Hemoglobins E And S In Orissa, India

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ABSTRACT

The population of India exhibits a wide range of genetic heterogeneity and, ecological and biological diversity including the reservoir for occurrence of a large number of abnormal hemoglobins and thalassemias in the world. Orissa state has nurtured ecological, cultural and genetic diversity of hemoglobinopathies, which has become a major genetic and public health problem in both tribal and nontribal inhabitants. Prospective studies are lacking in India. In Orissa, prospective studies of referral index cases of hemoglobinopathies provide most valuable data for analysis with respect to introspection and re-evaluation from research point of view. Intravenous blood samples were collected after obtaining informed consent from each index case of double heterozygosity for hemoglobinopathies. Background data of each individual were recorded like age, sex, caste, place of origin, reproductive history, consanguinity, etc. Twelve index cases of sickle cell-β-thalassemia and 9 index cases of hemoglobin E-β-thalassemia with anemia were subjected to detailed hematological and family genetic investigations. Routine standard hematological and biochemical investigations were carried out. This study highlights the rare occurrence of double heterozygosity of abnormal hemoglobins, i.e. Hb D, E and S with β-thalassemia mutation for the first time from Orissa. Study showed that index cases with Hb E-β-thalassemia and sickle cell-β-thalassemia manifest variable clinical, hematological and prognostic profile. High levels of fetal hemoglobin in patients reduce the severity of clinical symptoms in some but not in others. β-thalassemia or Hb E/S carrier mothers during their reproductive life either had spontaneous abortions or neonatal deaths. Aberrant heterosis for hemoglobinopathies such as occurrence of β-thalassemia mutation with structurally abnormal hemoglobins (Hb S and Hb E) is a rare entity, but occurs with severe clinical manifestations only in those areas or communities where abnormal hemoglobins and β-thalassemia are highly prevalent. This study provides for the first time a comprehensive database on the occurrence of double heterozygosity, testifying the genetic diversity and ethnic admixture in Orissa, India.

Key words: Hemoglobinopathies, Sickle cell- β -thalassemia, Hemoglobin E-β-thalassemia, Severe clinical & hematological profile, Endemic population.

Introduction

Hemoglobinopathies are the most commonly encountered monogenic disorders of blood posing a major genetic and public health problem in Southeast Asia and the Indian subcontinent [1]. Of the several abnormal hemoglobins so far identified [2], there are three variants – sickle cell (Hb S), hemoglobin E (Hb E) and hemoglobin D (Hb D),

which are predominantly prevalent in India. There are regional variations for these structural variants of hemoglobin; the cumulative allele frequency in different parts of India for these variants has been found to be 5.35% [3]. The average allele frequency of sickle cell and hemoglobin D has been observed to be 4.3% and 0.86%, respectively with hemoglobin E constituting 10.9% in North Eastern region of India [3]. The sickle cell disease is wide spread in tribal as well as nontribal communities especially in the Central-East India. With a prevalence range of 3-17%, the β-thalassemia is prevalent throughout India [2]. Thus, hemoglobinopathies are a huge genetic burden and pose a major health care challenge in India.

The sickle cell hemoglobin (Hb S) is structurally abnormal variant with glutamic acid residue replaced by valine at 6^{th} position of β-globin polypeptide chain of the molecule. Hemoglobin E is another structurally abnormal variant with a substitution of glutamic acid by lysine in the 26^{th} position of the β-globin polypeptide chain. Interaction of these structurally abnormal hemoglobins (D, E, S) with the disrupted synthesis of globin moiety leads to combination of two abnormalities, resulting in double heterozygosity of the disease. In view of the strict practice of caste endogamy and nonrandom (consanguinity) mating pattern, the β-thalassemia is seen in association with structural variants of hemoglobin in India. However, only a few reports on double heterozygosity are available in India [4],[5],[6],[7],[8].

Double heterozygosity for hemoglobinopathies is a rare entity. Due to the paucity of adequate literature available on the occurrence of heterosis for hemoglobinopathies in India and the hemoglobinopathies being a major genetic and public health problem in the state of Orissa, the present study was undertaken to evaluate the detailed family studies of double heterozygosity cases of sickle cell and hemoglobin E with β-thalassemia. This study will not only add to prognostic understanding of the heterosis phenomenon for hemoglobinopathies in the state but also be useful for genetic counseling, prenatal diagnosis and future molecular studies on the subject in India.

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Material And Methods

This prospective family study is based on nine index cases of sickle cell-beta-thalassemia and seven index cases of hemoglobin-E-beta-thalassemia. They were selected out of 1500 routinely referred cases, sent for investigation for the cause of anemia and suspected to be suffering from hemoglobinopathies before first blood transfusion, from different peripheral Primary Health Centres (PHCs) and hospitals in the state of Orissa during the period between 2001 to April 2006. Details diagnosis and analysis of the referral cases in Orissa have already been reported elsewhere [9],[10]. All the above mentioned index cases and their other available family members such as parents, brothers and sisters were also subjected to clinical examination and investigation for the cause of anemia and genetic/marriage counseling after taking informed consent. In all, there were 30 subjects related with sickle cell-β-thalassemia and 22 for hemoglobin E-β-thalassemia. Background information for each family such as name, age, sex, caste, native place, reproductive history, family pedigree, and clinical signs and symptoms were recorded.

About 2-3 ml. intravenous blood samples were collected using ethylene diamine tetra acetic acid (EDTA) as anticoagulant by disposable syringes and needles from each individual after obtaining the informed consent. All the signs and symptoms related to hemoglobinopathy after clinical examination were recorded on the pre-designed proforma. Laboratory investigations were carried out following the standard procedures after cross checking for quality control from time to time. Hematological parameters were studied by using an automated Blood Cell Counter (Model- MS4, Melet Schloesing Laboratories, Cergy-Pontoise Cedex, France).

The sickling test was performed by using freshly prepared sodium metabisulphite solution as reducing agent for the presence or absence of sickle cell hemoglobin [11]. The routine hemoglobin lysate electrophoresis was carried out on cellulose acetate membrane (CAM) in Tris-EDTA-Borate buffer at pH 8.6 and quantification of A_2 fraction of adult hemoglobin was done by elution method [12]. The value more than 3.5% of A_2 fraction of adult hemoglobin was taken as cut off point for determining the β-thalassemia trait. Those individuals having the very high hemoglobin A_2 value, i.e. more than 10% were suspected to have Hb A_2 plus HbE, which was confirmed by the investigation of other family members. Estimation

of fetal hemoglobin was done as described by Weatherall [12].

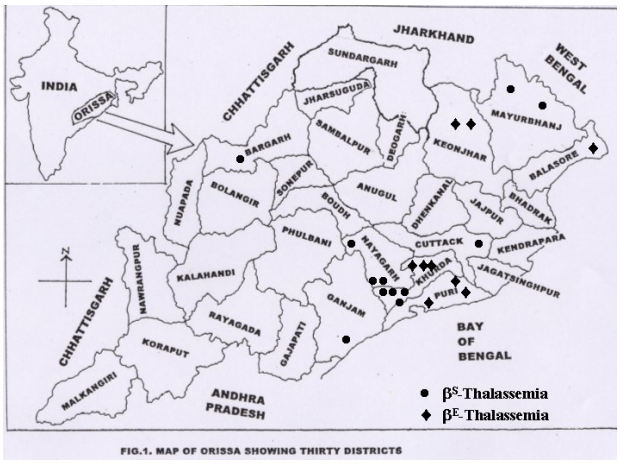
The diagnosis of sickle cell-β-thalassemia was based on the findings of hemoglobin (Hb) A, F, S/D and A_2 on electrophoresis under acidic and alkaline pH, elevated HbA_2 levels (>3.5%), and family studies. All the blood samples were further subjected to hemoglobin variant analysis for detecting any discrepancy (made for Bio-Rad Diagnostics, Hercules California, USA).

Results

The clinical, hematological and genetical findings of the 9 index cases of sickle cell-β-thalassemia and 5 index cases of hemoglobin E-β-thalassemia and their family members have been summarized in [Table/fig 2] and [Table/fig 3], respectively.

Out of twelve cases of sickle cell-β-thalassemia, it is apparent from table-1 that both parents who were obligatory heterozygotes for sickle cell and β-thalassemia were studied except one family (where the mother was not available) to confirm the diagnosis. Two cases each belonged to Khurda, Mayurbhanj and Nayagarh districts and one case each to Bargarh, Cuttack and Ganjam districts of Orissa. Fig-I is a map of Orissa showing distribution of cases studied. Three cases were from General (GC) Castes (1 Karan and 2 Khandayat), two cases each from Other Backward (OBC) Castes (Kulita and Mali), Scheduled (SC) Castes (Dhoba and Haddi) and Scheduled (ST) Tribes (Bhumij and Santhal). Both SC and ST cases belonged to Nayagarh and Mayurbhanj districts, respectively. It was evident that these cases were neither accentuated to a particular caste/community due to practice of consanguinity (inbreeding) nor to a particular area/region due to territorial endogamy but have random scatter in Orissa state. There were six cases each of age between 2 through 27 years and belonging to either sex with maximum number falling in adolescent age group [Table/fig 2].

These cases had a large range of variation in hemoglobin levels (5.2-13.0%), but the majority had moderate to severe anemia. It is apparent from table 1 that the majority of the sickle cell-β-thalassemia cases showed reduced values of red cell indices like HCT, MCV, MCH and MCHC manifesting hematological aberrations before blood transfusion. The RBC values were either reduced or normal in 12 cases of sickle cell-β-thalassemia.



Map of Orissa showing thirty districts

The leucocytosis was observed in almost all the cases. Only one Khandayat family with three children showed the higher value of RDW, more than 12% in these patients [Table/fig 2]. The red cell morphology also showed hypochromia and microcytosis in this family. Almost in all the cases of sickle cell-β-thalassemia, the fetal hemoglobin was raised, ranging from 4.5% to 38.3% in the present study. Hb A_2 was also raised, i.e.>3.5%, in all the sickle cell-β-thalassemia cases.

All the sickle cell-β-thalassemia cases had splenomegaly. The enlarged spleen size varied between 5-18 cm below the left costal margin, whereas, the hepatomegaly ranged from 2-3 cm below the right costal margin in these patients. Three patients reported the history of multiple transfusions starting from the age of about one year ranging from 1-25 units. The clinical picture in all the cases was similar to that of homozygous sickle cell disease. One β-thalassemia carrier mother had two neonatal deaths [Table/fig 2].

Of the nine cases of hemoglobin E-β-thalassemia reported here, it is apparent from [Table/fig 3] and [Table/fig 4]. that both parents who were obligatory heterozygotes for hemoglobin E and β-thalassemia were studied to confirm the diagnosis. Except one case, which belonged to OBC (Thodia), all others were from General Castes (Brahmin, Khandayat and Muslim). There were six males and three females having this disease, with maximum number of them falling in the childhood and adolescent age group (1-18 years).

The hematological parameters of hemoglobin E-β-thalassemia cases are summarized in [Tables/Fig 3] and [Table/fig 4]. The hemoglobin levels of these

Table/Fig 1

cases varied between 2.8-9.2% and the majority had moderate to severe anemia. It is apparent [Tables/Fig 3] that the majority of the hemoglobin E-β-thalassemia cases before blood transfusion showed reduced values of red cell indices like RBC, MCV, MCH and MCHC representing abnormal hematological conditions. The leucocytosis was observed in two cases. Three children showed the higher value of RDW, more than 12% in these patients [Table/Fig 3] and [Table/Fig 4]. The red cell morphology also showed hypochromia, microcytosis and target cells in these children. Almost in all the cases of hemoglobin E-β-thalassemia, the fetal hemoglobin was raised, ranging from 2.4% to 42.4% in the present study.

All the hemoglobin E-β-thalassemia cases had splenomegaly. The enlarged spleen varied from 2-9 cm below the left costal margin, whereas, the hepatomegaly ranged from 2-4 cm below the right costal margin in these patients. All the patients reported the history of multiple transfusions ranging from 1-25 units from the age of about one year. The clinical picture in all cases was similar to that of hemoglobin E disease or homozygous β-thalassemia. It is to note that some β-thalassemia or Hb E carrier mothers in their reproductive life either had spontaneous abortions or neonatal deaths [Table/Fig 4] presents the summary sheet of hematological and clinical features of two index cases of hemoglobin E-beta-thalassemia with their mothers having double heterozygosity for two structural hemoglobin variants (abnormal hemoglobin DE and SE) in Orissa.

Discussion

Heterosis, also called hybrid vigor or boost in performance, is the increase in growth, size, fecundity , function , yield, or other characters in hybrids over those of the parents. In other words, heterosis is increased strength of different characteristics in hybrids, the possibility to obtain a "better" individual by combining the virtues of its parents. Aberrant heterosis is antagonistic to heterosis, i.e. combination of ill effects or abnormal qualities in an individual. Therefore, it is not always true that the heterosis increases the strength of different characteristics in hybrid. Aberrant heterosis may occur with severer ill effects or abnormal qualities, lethal for the survival of an individual.

The first case of Hb E-β-thalassemia was reported in a Kulita caste family from Western Orissa by Balgir [7], followed by Chhotray & coworkers [8] and other reports of the prevalence of hemoglobin E gene from the state of Orissa [13],[14].

It is apparent from the present study that the patients of hemoglobin E-β-thalassemia and sickle cell-β-thalassemia disease manifest heterogeneity in clinical manifestations, hematological picture, prognosis and management profile in the state of Orissa [Table/fig 2], [Table/fig 3], [Table/fig 4]. The patients with early onset and severe anemia have the disease course similar to homozygous β-thalassemia in the former and that of sickle cell disease in the latter case, while those patients with late onset and mild anemia, manage with occasional blood transfusions or remain completely asymptomatic without any hemolytic crisis in life. Similar observations were made for Hb E-β-thalassemia in Sri Lanka [15] and Thailand [1] and for sickle cell-β-thalassemia in India [16],[17],[18].

The severity of phenotype is, therefore, dependent on the type of β-thalassemia mutation, depending upon the levels of HbE and HbF and the number of β-globin genes, which tend to reduce the severity of the disease [16],[17]. The interaction of hemoglobin E and β-thalassemia results in a wide spectrum of clinical conditions, in some cases indistinguishable from β-thalassemia major, whereas, in others it has a milder course without dependent on transfusion. These findings get further support from the earlier studies [5],[7],[8],[13]. The Hb E is a mild structural variant of β-globin chain, being asymptomatic in homozygous state [19]. The clinical presentations of all the cases of Hb E-β-thalassemia were identical with those of β-thalassemia major patients in Orissa. The peripheral blood smear examination revealed a hypochromic and microcytic picture with predominance of target cells. All the cases in the present study were transfusion dependent from the age of one year onward and needed multiple blood transfusions, varying from 1 to 25 units.

The heterogeneous clinical and hematological profile of Hb E-β-thalassemia patients is reflected in [Table/fig 3] and [Table/fig 4]. However, the picture in seven cases in the present study was compatible with those patients of β-thalassemia major in this region. All the patients had pallor, fatigue, recurrent fever, joint pains, splenomegaly and hepatomegaly of varying degree or grades. The clinical picture is compatible with the studies reported from other parts especially from the North-Eastern region of India where its frequency is reported to be very high [4],[6],[14],[20]. Severely affected patients of Hb E-β-thalassemia had marked

anemia, jaundice, bossy maxillary bones and prominent hepatosplenomegaly.

Most of these Hb E-β-thalassemia patients die due to secondary infection in infancy, whereas, some of them survive upto 5th or 6th decade of life. As evident from the age distribution of the HbE-β-thalassemia patients (1-18 years) in the present study, the age of onset as well as the course of the disease is similar to that of β-thalassemia major.
High levels of fetal hemoglobin are known to reduce the severity of clinical symptoms (18). In the present study, Hb F values of Hb E-β-thalassemia cases ranging between 2.4-42.4% showed wide range of variations, suggesting some other factors like iron/folic acid deficiency, afflicted with nutritional disorders, parasitic infestations, malarial or viral infections, etc. which are very common in the state of Orissa, may be responsible for mild or severe course of the disease. However, all the Hb E-β-thalassemia patients in the present study were multiple transfusion dependants.
Female patients with sickle cell-β-thalassemia may have reduced fertility, but pregnancy occurs and is associated with high degree of maternal morbidity and fetal wastage [21]. In the present study, some β-thalassemia or Hb E or S carrier mothers in their reproductive life either had spontaneous abortions or neonatal deaths [Table/fig 2], [Table/fig 3]. These finding gets further support from our earlier study [22] that the infant and neonatal mortality is high among the carrier parents of Hb E/S disease or β-thalassemia in Orissa. Precipitating factors for onset of crisis include cold or wet weather, and winter season in the state of Orissa [23]. Jaundice is the most common feature. The patients with sickle cell- β -thalassemia had markedly higher values of HbA_2 and lower of MCV and MCH as per the normal standards.

The important determinants of clinical severity of sickle cell-β-thalassemia are the type of β-mutation, number of β-globin genes, and level of HbA_2 and of fetal hemoglobin that inhibits intracellular polymerization of sickle hemoglobin, and hence sickling, and reduces the severity of hemolysis and the occlusion of the microcirculation [18]. The molecular genetic nature of the determinant resulting in raised HbF levels (4.5% to 38.3%) in these Indian patients [Table/fig 2]. is not fully understood [20]. As a whole, the sickle cell-β-thalassemia in Orissa is indistinguishable from homozygous sickle cell disease except the raised HbA_2 level, and lower values of MCV and MCH [18]. The clinical manifestations are almost identitical to homozygous sickle cell disease in this Central-East Indian state.

Conclusion

The present study highlights the co-inheritance of β-thalassemia and hemoglobin E or sickle cell gene, which is wide spread in Southern and Western Orissa [24],[25]. It is a pity that a large number of such double heterozygosity cases remain mostly undiagnosed or misdiagnosed, wrongly interpreted and mismanaged leading to premature death without proper treatment in the state of Orissa [26]. For bringing awareness, motivation for carrier detection to reduce the genetic burden, and intervention in affected families and communities need to be launched vigorously in Orissa.
Molecular diagnosis of Hb D, HbE or Hb S gene is required in the first trimester of the antenatal period [27] of pregnancy along with characterization of β-thalassemia mutations in the region. The establishment of the prenatal diagnostic facilities and services, genetic/marriage counseling are the ultimate aims to be achieved in the state of Orissa.

Conflict of Interest: None declared

Table/Fig 2. Summary sheet of Hematological and Clinical Features of Nine Families of Sickle Cell-Beta-Thalassemia in Orissa.

Subjects	Age in yrs	Sex	Hb (g/dl)	RBC ($x10^6/\mu l$)	HCT (%)	MCV (fl)	MCH (pg)	MCHC (g/dl)	RDW (%)	WBC ($x10^3/\mu l$)	Electro-phoresis	Sick-ling	HbA_2 (%)	HbF (%)	HbS (%)	HbA (%)	Spleen (cm)	Liver (cm)	Clin
Father	72	M	08.7	3.8	29	77.1	22.9	29.2	8.6	6.3	AS	+ve	2.0	1.2	24.9	71.9	-	-	-
Mother	65	F	11.3	5.8	39	68.3	19.5	28.6	9.9	15.2	AA_2	-	4.1	0.4	-	95.5	-	-	-
Son	27	M	13.0	5.9	41	69.4	22.0	30.3	7.6	8.5	ASF	+ve	3.8	4.5	25.0	66.7	8	3	1*
(Kulita, OBC/Bargarh)																			
Father	38	M	11.0	5.9	37	62.9	18.7	29.8	8.9	7.5	AA_2	-	4.3	1.3	-	94.4	-	-	-
Mother	30	F	11.0	3.9	35	88.5	28.2	31.8	9.2	7.8	AS	+ve	4.5	1.8	31.9	61.8	-	-	-
Dau	6	F	5.2	2.6	19	74.3	20.0	27.0	11.2	14.3	ASF	+ve	6.6	22.2	64.6	6.6	9	2	2*
Dau	2	F	8.8	5.4	31	57.9	16.3	28.2	10.4	18.7	AA_2	-	3.6	6.0	-	90.4	-	-	-
(Bhumij, ST/Mayurbhanj)																			
Father	48	M	16.0	6.0	49	81.3	26.5	32.5	8.2	6.9	AS	+ve	2.3	0.7	36.6	60.4	-	-	-
Mother	40	F	10.5	6.1	36	59.4	17.1	28.9	9.2	12.7	AA_2	-	4.1	1.9	-	94.0	-	-	-
Son	15	M	6.5	3.7	24	63.8	17.2	27.2	10.5	17.5	ASF	+ve	4.8	19.7	69.0	6.5	10	-	3*
(Mali, OBC/Ganjam)																			
Father	53	M	12.9	5.0	40	80.5	26.1	32.1	11.2	5.1	AA_2	-	4.4	0.6	-	95.0	-	-	-
Mother	40	F	11.2	4.9	36	74.4	23.5	31.6	11.8	3.8	AS	+ve	1.7	2.5	32.2	63.6	-	-	-
Son	14	M	10.9	5.3	36	67.8	20.4	30.1	12.3	7.6	ASF	+ve	8.0	11.6	73.4	7.0	7	3	4*
(Santhal, ST/Mayurbhanj)																			
Father	36	M	14.5	5.0	46	92.0	29.2	31.7	7.9	8.4	AS	+ve	1.6	1.2	23.6	73.6	-	-	-
Mother	27	F	11.0	5.9	39	85.5	18.6	28.5	9.2	11.6	AA_2	-	4.5	0.6	-	94.9	-	-	-
Dau	3	F	7.7	3.7	28	63.9	20.6	29.6	8.6	17.9	ASF	+ve	3.9	38.1	51.7	5.5	-	-	5*
(Khandayat, GC/Cuttack)																			
Father	32	M	12.6	6.8	49	71.5	18.4	25.8	8.6	6.9	AA_2	-	5.8	0.5	-	93.7	-	-	-
Mother	29	F	12.9	4.9	45	91.2	26.2	28.7	8.7	5.7	AS	+ve	1.2	1.3	71.2	26.3	-	-	-
Dau	8	F	10.2	4.6	33	72.0	21.9	30.5	14.8	7.0	ASF	+ve	4.3	6.7	69.1	19.9	6	2	-
Son	6	M	8.1	3.8	29	76.2	21.4	28.1	16.3	11.9	ASF	+ve	3.7	18.2	70.1	8.0	5	3	6*
Dau	2	F	9.9	5.8	37	64.8	17.1	26.4	13.0	15.9	ASF	+ve	4.4	16.6	65.9	13.1	7	2	7*
(Khandayat, GC/Khurda)																			
Father	46	M	13.4	5.9	51	86.0	22.8	26.5	8.9	9.0	AS	+ve	2.1	2.3	27.3	70.4	-	-	-
Mother	35	F	9.2	5.3	36	68.2	17.2	25.4	9.8	7.3	AA_2	-	4.8	0.3	79.6	15.3	-	-	8*
Dau	8	F	6.4	4.0	26	64.4	16.1	25.1	10.8	11.4	ASF	+ve	4.9	6.3	76.7	12.1	8	2	9*
(Dhoba, SC/Nayagarh)																			
Father	32	M	13.4	5.7	41	71.0	21.9	30.9	9.6	8.6	AA_2	-	5.7	0.9	-	93.4	-	-	-
Mother	26	F	12.6	4.3	32	74.2	31.6	32.6	9.5	11.5	AS	+ve	2.6	1.7	34.0	61.7	-	-	-
Dau	12	F	8.6	4.9	43	68.9	28.5	32.2	7.2	8.4	ASF	+ve	3.8	38.3	44.5	13.4	7	3	10*
Son	7	M	8.8	4.3	35	68.0	25.7	31.5	9.3	8.6	ASF	+ve	3.8	18.2	50.7	27.3	18	2	-
(Haddi, SC/Nayagarh)																			
Father	41	M	11.9	6.4	44	68.8	18.7	27.2	11.7	7.2	AA_2	-	4.6	0.7	-	94.7	-	-	-
Son	16	M	6.2	3.7	24	65.7	16.7	25.5	11.1	11.6	ASF	+ve	3.8	12.2	72.8	11.2	9	3	11*
(Karan, GC/Khurda)																			

1*Recurrent Fever, Joint pains.
2* Weakness, loss of appetite, recurrent jaundice, joint pains, abdominal pains.
3* Pallor, weakness, joint pains.
4* No problem
5* Retarded Growth.
6* No problem
7* Jaundice, Pallor
8* Two Neonatal deaths
9* Weakness, pallor, recurrent fever, epistaxis, joint pains
10* Pallor, jaundice, retarded growth, history of hospitalization.
11* Pallor, jaundice, abdominal pains, Hyperbilirubinemia

Table/Fig 3. Summary sheet of Hematological and Clinical Features of Five Families of Hemoglobin –E-Beta-Thalassemia in Orissa.

Subjects	Age in yrs	Sex	Hb (g/dl)	RBC ($x10^6/\mu l$)	HCT (%)	MCV (fl)	MCH (pg)	MCHC (g/dl)	RDW (%)	WBC ($x10^3/\mu l$)	Electro-phoresis	HbA_2 (%)	HbF (%)	HbA (%)	Spleen (cm)	Liver (cm)	Clinical
Father	54	M	11.2	4.5	41	89.8	24.6	27.5	8.6	6.3	AE	26.6	0.4	73.0	-	-	-
Mother	45	F	11.3	5.2	38	67.4	22.4	33.4	7.5	7.0	AA_2	4.0	0.5	95.5	-	-	1*
Son	20	M	12.8	5.7	38	67.4	22.4	33.4	9.0	4.4	AA_2	3.9	0.5	95.6	-	-	-
Dau	18	F	9.2	5.0	29	58.1	18.1	31.1	10.3	4.9	AEF	83.9	2.4	13.7	8	3	2*
Son	16	M	3.8	1.6	13	87.5	24.5	28.1	11.3	0.9	AEF	84.5	3.2	12.3	6	2	3*
(Khandayat, GC/Puri)																	
Father	35	M	12.9	5.6	41	74.1	23.1	31.2	7.9	8.8	AE	38.8	0.9	83.3	-	-	-
Mother	32	F	10.4	5.9	34	58.2	17.6	30.3	12.6	6.3	AA_2	5.6	1.2	93.2	-	-	-
Son	9	M	7.9	5.2	28	53.5	15.0	28.2	23.0	10.9	AEF	41.3	10.0	49.7	7	2	4*
Son	7	M	11.5	5.3	35	66.2	21.5	32.6	9.7	8.4	AE	27.8	1.5	70.7	-	-	-
(Muslim, Khurda)																	
Father	40	M	13.5	5.9	44	75.0	22.8	30.4	7.9	8.6	AE	22.8	0.8	76.4	-	-	-
Mother	35	F	12.0	4.5	37	83.6	26.8	32.1	8.4	7.8	AA_2	3.8	0.5	95.7	-	-	5*
Dau	16	F	4.9	2.9	18	62.7	16.8	27.0	11.4	3.5	AEF	76.7	3.5	19.8	9	3	6*
Dau	13	F	12.7	5.3	41	77.1	23.8	30.9	6.7	7.6	AA_2	8.0	0.6	71.4	-	-	-
Son	11	M	12.1	5.3	40	74.3	22.7	30.5	7.4	13.3	AA	2.0	0.4	97.6	-	-	-
Son	8	M	11.3	5.3	36	67.4	21.2	31.5	8.7	11.5	AE	33.4	0.7	65.9	-	-	-
(Thodia, OBC/Khurda)																	
Father	40	M	12.9	7.4	47	63.2	17.4	27.5	10.9	8.4	AA_2	5.6	0.6	93.8	-	-	-
Mother	36	F	10.4	5.3	37	70.6	19.8	28.0	10.1	13.9	AE	27.9	0.6	71.5	-	-	7*
Son	1	M	5.6	4.5	22	49.1	12.4	25.2	18.5	31.7	AEF	56.8	23.2	20.0	6	2	8*
(Khandayat, GC/Puri)																	
Father	41	M	11.9	6.6	42	63.0	18.0	28.6	10.6	6.7	AA_2	5.4	0.5	94.1	-	-	-
Mother	32	F	10.4	5.1	37	71.7	20.2	28.2	9.0	8.8	AE	26.3	0.5	73.2	-	-	9*
Dau	8	F	5.1	2.5	19	77.5	20.6	26.6	9.5	3.8	AEF	77.6	10.4	12.0	9	4	10*
Son	4	M	2.8	1.9	11	61.3	14.7	24.1	19.0	5.1	AEF	41.0	42.4	16.6	7	3	11*
(Brahmin, GC/Keonjhar)																	

1* Had neonatal deaths of 3 sons and one daughter.
2* Recurrent fever, joint pains, transfusion dependent.
3* Pallor, weakness, transfusion dependent.
4*History of jaundice, transfusion dependent.
5*Had one spontaneous abortion.
6* Abdominal pains, growth retardation, transfusion dependent.
7* Had one abortion
8* Pallor, weakness, transfusion dependent.
9*Had neonatal deaths of one son and one daughter
10* Pallor, recurrent fever, protuberated abdomen , transfusion dependent
11* Pallor, recurrent fever.

Table/Fig 4. Summary sheet of Hematological and Clinical Features of Two index cases of Hemoglobin E-Beta-Thalassemia with mothers having double heterozygosity for two structural hemoglobin variants (abnormal hemoglobin DE and SE) in Orissa.

Subjects	Age in yrs	Sex	Hb (g/dl)	RBC ($x10^6/\mu l$)	HCT (%)	MCV (fl)	MCH (pg)	MCHC (g/dl)	RDW (%)	WBC ($x10^3/\mu l$)	Electro-phoresis	Sick-ling	HbA_2 (%)	HbF (%)	HbS (%)	HbA (%)	Spleen (cm)	Liver (cm)	Clin
Father	27	M	11.2	4.5	35	69.0	24.6	27.5	8.6	6.3	AA_2	-ve	6.6	1.4	-	92.0	-	-	-
Mother	23	F	12.6	2.4	15	61.9	31.0	19.3	13.4	12.9	SE	+ve	26.2	4.8	69.0	-	-	-	1*
Son	1	M	9.4	3.6	28	60.9	30.0	19.3	11.4	4.7	AEF	-ve	65.7	7.3	-	27.0	2	-	2*
(Khandayat, GC/Khurda)																			
Father	35	M	9.9	4.9	28	64.0	20.0	21.0	15.4	6.5	AA_2	-ve	5.6	0.8	-	93.6	-	-	-
Mother	27	F	12.3	4.7	29	73.6	25.9	25.6	15.1	8.6	DEA_2	-ve	32.3	1.0	66.7	-	3	2	3*
Son	3	M	6.7	3.7	23	62.5	18.1	19.3	32.1	9.3	AEF	-ve	67.7	25.8	-	6.5	2	-	4*
(Brahmin, GC/Balasore)																			

1* History of jaundice.
2* Pallor, recurrent fever, transfusion dependent
3* Pallor, Microcytosis, splenomegaly, hepatomegaly.
4* Pallor, Hypochromia, Microcytosis, recurrent fever, splenomegaly, transfusion dependent.

References

[1] Fucharoen S, Winichagoon P. Hemoglobinopathies in Southeast Asia: molecular biology and clinical medicine. Hemoglobin 1997; 21:299-319.

[2] Balgir RS. The burden of hemoglobinopathies in India and the challenges ahead. Curr Sci 2000; 79:1536-1547.

[3] Balgir RS. Genetic epidemiology of the three predominant abnormal hemoglobins in India. J Assocn Phys India 1996; 44:25-28.

[4] Chatterjea JB. Some aspects of Hb E and its genetic interaction with thalassemia. Indian J Med Res 1965; 53:377-383.

[5] Agarwal S, Gulati R, Singh K. Hemoglobin E-beta-thalassemia in Uttar Pradesh. Indian Pediatr 1997; 34:287-292.

[6] De M, Das SK, Bhattarcharya DK, Talukder G. The occurrence of Beta-thalassemia mutation and its interaction with hemoglobin E in the Eastern India. Int J Hemat 1997; 66: 31-34.

[7] Balgir RS. Double heterozygosity for abnormal hemoglobins in six families of Orissa. Curr. Med. Trends 1998; 2:378-385.

[8] Chhotray GP, Dash BP, Ranjit MR, Colha, RB, Mohanty D. Hemoglobin E/ß-thalassemia – an experience in the Eastern Indian state of Orissa. Acta Hemat 2003; 109:214-216.

[9] Balgir RS. Spectrum of hemoglobinopathies in the state of Orissa, India: A ten years cohort study. J Assoc Phys India 2005; 53:1021-1026.

[10] Balgir RS. Scenario of hemoglobin variants in Central-East coast of India. Curr Sci 2006; 90:1651-1657.

[11] Dacie JV, Lewis SM. Practical Hematology. 7th Edn. Edinburgh: Churchill Livingstone 1991,pp.227-258.

[12] Weatherall DJ. Hematologic Methods. In: DJ Weatherall (Ed.) Methods in Hematology: Thalassemias.Vol.6.New York: Churchill Livingstone. 1983. pp. 27-53.

[13] Balgir RS, Mishra RK, Murmu B. Clinical and hematological profile of hemoglobino- pathies in two tribal communities of Sundargarh district in Orissa, India. Int J Hum Genet 2003; 3:209-216.

[14] Balgir RS. Prevalence of abnormal hemoglobin E gene in the Dhelki Kharia tribal population. Curr Sci 2003; 85:1604-1608.

[15] Premawardhene A, Fisher CA, Olivieri NF, de Silva S, Arambepola M, Perera W, O'Donnell A, Peto TEA, Viprakasit V, Merson L, Muraca G, Weatherall DJ. Haemoglobin E ß thalassaemia in Sri Lanka. Lancet 2005; 366:1467-1470.

[16] Serjeant GR. Sickle cell disease. New York: Oxford University Press.1992.

[17] Weatherall DJ, Clegg JB. Inherited hemoglobin disorders: an increasing global health problem. Bull World Health Organization 2001; 79: 704-712.

[18] Kulozik AE, Bail S, Kar BC, Serjeant BE, Serjeant GE. Sickle cell- ß thalassemia in Orissa state, India. Br J Hemat 1991; 77: 215-220.

[19] Balgir RS. Fertility and clinical profile of hemoglobin-E afflicted mothers in North Eastern India. Med Surg 1993; 10:57-59.

[20] Balgir RS. Emerging trends in genetic epidemiology of hemoglobinopathy in seven sister states of North Eastern India. In: North East India in Perspectives: Biology, Social Formation and Contemporary Problems. Das, Rajat Kumar, Basu, Debashish. Eds. New Delhi: Akansha Publishing House, 2005,pp.17-37.

[21] Balgir RS, Dash BP, Das RK. Fetal outcome and childhood mortality in offspring of mothers with sickle cell trait and disease. Indian J Pediatr 1997; 64:79-84.

[22] Balgir RS. Infant mortality and reproductive wastage associated with different genotypes of haemoglobinopathies in Orissa, India. Ann Hum Biol 2007; 34:16-25.

[23] Balgir RS. Epidemiological study of sickle cell disease in Western Orissa. Man in India 1993; 73:363-370.

[24] Balgir RS, Dash BP, Murmu B. Blood groups, hemoglobinopathy and G-6-PD deficiency investigations among fifteen major scheduled tribes of Orissa, India. Anthropologist 2004; 6:69-75.

[25] Balgir RS. Clinical genetics and hematological profile of sickle cell cases in twenty families of Orissa. Indian J Hemat Blood Transfus 2006; 22:45-52.

[26] Balgir RS. Genetic epidemiology of the sickle cell anemia in India. Indian Practr 2001; 54:1-6.

[27] Balgir RS. Control and prevention of genetic load of hemoglobinopathies in India. Natl Med J India 1999; 12: 234-238.