Comparative Evaluation Of Osmotic Fragility Test And Hba2 Estimation As The Screening Tests For Beta Thalassemia Trait

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ABSTRACT

Aim: To evaluate sensitivity and specificity of osmotic fragility test in reference to HbA2 estimation in the screening of beta thalassemia trait.

Methodology: A total of 400 subjects were included in the study and divided into two groups, Group I with apparently healthy siblings of beta thalassemia major and group II with normal healthy controls. Their HbA2 levels were estimated and Group I subjects were further subdivided into two groups on the basis of HbA2 levels, Group IA comprises of normal siblings of beta thalassemia trait, Group IB comprises of BTT siblings of beta thalassemia major. Osmotic fragility test was done on all the subjects and MCF of all the three groups was estimated. The sensitivity and specificity of MCF in BTT siblings was estimated.

Results: Mean \pm SD values of MCF in groups IA, IB and III were 0.4 \pm 0.04, 0.34 \pm 0.04 and 0.42 \pm 0.03% respectively. In this study, sensitivity and specificity of MCF in BTT siblings is 98.6% and 91.7% respectively when HbA2 is used as a gold standard.

Conclusion: Osmotic fragility test is a sensitive, cost effective, and reliable screening test for detection of beta thalassemia trait in a population.

Key words: Beta thalassemia trait (BTT), Mean cell fragility (MCF),

INTRODUCTION

Osmotic fragility test can be very useful screening tool for beta thalassemia trait¹. Osmotic fragility test is sensitive, rapid and reliable screening test in under resourced laboratories². Over the vears. determination of osmotic fragility of red blood cells has emerged as single most effective, inexpensive and easily reproducible test for mass screening program for BTT³. A decreased tendency of osmotic fragility may also be found in cases of iron deficiency anaemia. It indicates the presence of unusually flattened red cells in which the volume to surface ratio is decreased. However osmotic fragility of red cells is frequently more markedly reduced in thalassemia major and minor than in iron deficiency anaemia⁴.

METHODOLOGY

This study was carried out at thalassemia centres Sir Ganga Ram Hospital Lahore, Institute of Blood Transfusion. A total of 400 subjects were included in the present study and divided into the three equal groups. Group I comprised of apparently asymptomatic siblings of beta thalassemia major patients and were further divided into two groups. IA

Correspondence to: Dr. Nasir Rashid, Assistant Professor Pathology, Amna Inayat Medical College, Sheikhupura are normal siblings of BTM patients and IB BTT siblings of BTM patients. Group II were normal healthy controls. Asymptomatic siblings of diagnosed cases of beta thalassemia major were selected. The patients with history or diagnosis of any acute or chronic illnesses were excluded. Blood was drawn in a EDTA containing tube / vial. This sample was used for haemoglobin electrophoresis and 3.0 ml venous blood was heparinized for osmotic fragility test.

RESULTS

The detail of results is given in tables 1 and 2

Table 1: MCF in Group IA, IB, II and III

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MCF (%)	IA	IB	III		
Mean±SD	0.40±0.04	0.34±0.04	0.42±0.03		
Ranges	0.35-0.45	0.20-0.45	0.40-0.45		
Total Subjects	44	156	200		

Table 2: Results of MCF In BTT siblings (HbA₂ as Gold Standard)

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Results of	HbA2		Total	
MCF(%)	Positive	Negative		
Positive	True + ve	False +ve	a+b	
	(a)(150)	(b)(04)	(154)	
Negative	False -ve	True-ve (d)	c + d	
_	(c)(02)	(44)	(46)	
Total	a + c	b + d	n =	
	(152)	(48)	(200)	

Sensitivity:a/(a+c) x $100 = \frac{150 \times 100}{152} = 98.6\%$

Specificity: $\frac{d \times 100}{d + b} = \frac{44 \times 100}{48} = 91.7\%$

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DISCUSSION

In this study, the sensitivity of MCF was 98.6 % and specificity is 91.7%. Gomber et al (1997)⁵ reported the sensitivity of mean cell fragility (MCF) as 95.59% and the specificity as 84.2%. Sirichotiyakul et al(2004)⁶ estimated the sensitivity and specificity of MCF in pregnant women. 1n this study, these values were reported to be 97.6% and 72.9% respectively. Raghavan et al (1991)⁷ also calculated the sensitivity of the test as 95.5% and specificity as 87%. Thomas et al(1996)8 reported that this test showed an overall sensitivity of 98.7%, specificity of 66.6%. Manglani et al (1997)⁹ reported the sensitivity of MCF as 94.4%, specificity as 64.2%. Chow et al(2005)10 estimated the sensitivity of MCF as 95%, specificity as 86%. Thool et al (1998)¹¹ translated their results of MCF into a specificity of 100% and sensitivity of 95.2%.

Moinuddin & Ashraf (2004)¹² conducted a study in the Institute of Haematology, Bagai Medical University, Karachi evaluating a modification of MCF as a screening test for beta thalassemia trait. In this study, hemolysis was measured by recording the optical density after centrifugation. The osmotic fragility index was calculated to be the ratio of the optical density of the hemolysate in 0.36% saline to the optical density(OD) of the hemolysate in distilled water, multiplied by 100. 95% (57 out of 60) of the hematologically normal donors had OD higher than 70%. The superiority of MCF over other screening tests for beta thalassemia trait is not limited to these two factors. MCF is also a rapid test. The time required to perform the test did not exceed 45 minutes whereas performance of HbA2 estimation requires much longer than this. Moreover, unlike other screening tests for beta thalassemia trait, MCF does not require any calculation, equipment. Technically trained personnel and is much easier to interpret.

CONCLUSION

Mean cell fragility (MCF) is reduced in BTT siblings (IB) as compared to control group (II). The sensitivity of MCF is 98.6 % and specificity is 91.7%

REFERENCES

- 1. Bobhate SK, Gaikwad Sr, Bhaledrao I. NESTROFF as a screening test 101, detection of Beta-thalassemia trait. Indian .1 Pathol Microbiol 2002: 45: 265-7.
- Winichagoon P, Thitivichituilert A, Lebnak T, Piankijagum A, Fucharoen S. Screening for the carriers of thalassaemias and abnormal hemoglobins at the community level. Southeast Asian J Trop Med Public Health 2002:145-50.
- Mamtani M, Jawahirani A, Das K, Rughwani V, Kulkami H. Bias-corrected diagnostic performance of the naked-eye single-tube red-cell osmotic fragility test (NESTROFT): an effective screening tool for betathalassemia. Hematology 2006; 11:277-86.
- Roper D. and Layton M (eds). Dacie and Lewis Practice of Haematology, Churchill Livingston Edinburgh 2006; 205-16.
- Gomber S, Sanjeev, Madan N. Validity of Nestroft in screening and diagnosis of beta-thalassemia trait. J Trop Pediatr 1997; 43: 363-6.
- Sirichotiyakul S, Maneerat J, Sanquanscrinst I, Dhanaiijayanonda P, Tollgscnb T. Sensitivity and specificity of mean corpuscular volume testing for screening for alpha-thalassernia-1 and betatlialassecio traits.1 Obstet Gynaecol Res 2005: 31: 1: 198-201.
- Raghavan K, Lokeshwar MR, Birewar N, Nigam V, Manglani MV, Raju N. Evaluation of naked eye single tube red cell osmotic fragility test in detecting betathalassemia trait. Indian Pediatr 1991; 28: 469-72.
- Thomas S, Srivastava A, Jeyaseelan L, Dennison D, Chandy M. NESTROFT as a screening test for the detection of thalassaemia & common haemoglobinopathic evaluation against a high performance liquid chromatographic method. Inidan J Med Res 1996:,104: 194-7.
- Manglani M, Lokeshwar MR, Vani VG, Bhatia N, Mhaskar V. NESTROFF an effective screening test for beta thalassernia trait. Indian Pediatr1997; 34: 702-7.
- Chow J, Phelan L, Bain BJ. Evaluation of single-tube osmotic fragility as a screening test for thalassernia. Am J Hematol 2005; 79: 198-201.
- Thool AA, Walde MS, Shrikhande AV, Talib VH. A simple screening test for the detection of heterozygous beta thalassemia. Indian J Pathol Microbiol 1998; 41: 423-6.
- Moinuddin, Ashraf S. Osmotic fragility index as a screen for B-thalassemia trait. Proceedings of the 6th National conference of haematology: 2004 Feb 12-14: Lahore, Pakistan.