Correlation of Liver Function Test with Different Age and Sex Group and with Ferritin Level in Thalassemia

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Abstract

Thalassemia is the most common genetic blood disease in the world and varies in different population group in the world. In thalassemic patients liver damage is augmented when age of the patients are increase. These are due to increasing age with advancement of disease progression, repeated blood transfusion, less use/ intolerance of iron chelating agent, decreased activity of hepatocyte to rescue them in such excess billirubin and iron flood. Secondary to hypersplenism.

Iron overload due to frequent transfusions in β-thalassemia results in abnormal organ function tests. Proper and timely screening of these parameters can help in early diagnosis & prevention of iron overload. Iron overload is a main leading cause of elevated liver enzymes, and presence of HCV infection is significantly related to the increased iron overload[14]. Liver injury whether acute or chronic, eventually results in an increase in serum concentrations of Alanine transaminase (ALT) and Aspartate transaminase (AST). In transfusion dependent thalassemia patients iron overload is often inevitable and exposed to transfusion-associated infections. Apart from these when the age is more, the disease progresses with their complication like hepatic injury. The thalassemia patient develops liver fibrosis as a result of iron overload due to excessive blood transfusion and also from excess intestinal absorption. N Sultana et al.[6] showed that serum ferritin and serum bilirubin parameter of iron over load and jaundice are correlated.

Key Words: *Thalassemia, liver function test, ferritin.*

Introduction

Thalassemia, the most common hereditary disorder worldwide[1].Approximately 7% of the global population is carrier for hemoglobin disorder[2].Beta-thalassemia syndromes are a group of hereditary blood disorders characterized by reduced or absent beta globin chain synthesis, resulting in reduced Hb in red blood cells (RBC), decreased RBC production and anemia. It is mostlyinherited as recessive traits [3]. Hemoglobinopathies are characterized by the production of structurally defective hemoglobin due to abnormalities in the formation of globin moiety of the molecule [4].Beta-thalassaemias are heterogeneousgroup of disorders and has three types, beta-thalassemia minor or beta-thalassemiatrait, beta-thalassemia intermedia and beta-thalassemia major. Beta thalassemia occurwidely in a broad belt, ranging from theMediterranean and parts of north and West Africa through the Middle East and Indian subcontinent to South East Asia[4].Hb-E beta-thalassemiais the commonest severe form of thalassemia in South East Asia and part ofIndian subcontinent.Hb-E isineffectively synthesized and hence, when itinherited together with beta-thalassemia there ismarked deficiency of beta-chain production [5].It is also divided into mild, moderateand severe form clinically. Clinical presentation of severe form is similar with beta-thalassemia major. These patients can survive long with the bettertreatment [6]. They need alifelong repeated blood transfusion to

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maintaintheir hemoglobin level around 12g/dl, but multiple transfusion will cause an accumulationof iron in various tissue accompanied by anincrease serum iron level[7].In thalassemia, iron over load occurswhen iron intake is increased over a sustainedperiod of time, either from the transfusion of redcells or because there is increased absorption ofiron from digestive tract. As there is nomechanism in human to excrete the excess iron, it has to be removed by chelating agent[8]. Iron can deposit in different visceral organs mainly in heart, liver and endocrine glands causing tissuedamage and most of the mortality and morbidity associate with thalassemia. Iron is stored in the body as ferritin mainly within the reticuloendothelial system and its overload may promote hepatic injury and fibrogenesis [9]. Furthermore blood transfusion dependent thalassemia are also liable to infected with Hepatitis B and Hepatitis C if proper screening is not done which can cause hepatic fibrosis and cirrhosis[10]. So thalassemia patients must be routinely checked for liver function, cardiac function, endocrine function and also serum ferritin and they should be treated accordingly for the maintenance of healthy life.

Materials and Method

The study was conducted in the Hematology Department, Calcutta school of Tropical medicine,from September 2018 to May 2019. This study was designed on the basis of retrospective observational type of study.This study was conducted on consecutive 64 transfusion dependent thalassemia patient. Thalassemia diagnosis was confirmed by examining hemoglobin electrophoresis. A pre-designed Proforma was used to collect information from the hospital records. By using the proforma which included sex, age at presentation, age at diagnosis, ferritin level, body weight, pre-hemoglobin and clinical symptom at presentation.

Inclusion criteria

Patient over 2 years to 30 years of age with confirmed thalassemia by examining hemoglobin electrophoresis. Those having symptom of clinical jaundice with or without history of chronic blood transfusion.

Exclusion criteria

Age more than 30 years

Statistical Analysis

Data were analyzed using the statistical software SPSS (version 21). All the variables were tested for normality, so that suitable parametric statistical tools could be used. Analysis of variance (ANOVA) of the data was used to detect overall difference in group means.

Observation and Results

Clinico-hematological study of Thalassemia was done on 64 patients during the period of 2018-2019.

AGE and SEX:-

Mean (±SD) age in total 64 patients between the age group 1 to 10 years was 5.70 ± 2.084, between 11 and 20 years mean was 15.76 ±3.562 and between age 21 to 30 years was 25 ± 2.8982 were include in the study. Our study based on 64 Thalassemia patients. Sex distribution in different patients shown in Age and sex distribution in different patients shown in **Table 1:**

Table1: Group statistics of LFT with AGE and SEX distribution

subject		Frequency	Percent (%)	Valid Percent	Cumulative Percent
Age group	1-10	17	26.6	26.6	26.6
	11-20	25	39.1	39.1	65.6
	21-30	22	34.4	34.4	100.0
	Total	64	100.0	100.0	
Valid	F M Total	35 29 64	54.7 45.3 100.0	54.7 45.3 100.0	54.7 100.0

Corelation Between Lft and Age:-

A Pearson product- moment correlation is run to determine the relationship between Total Billirubin and AGE. There is low correlation is found between TBill and AGE, which is statistically insignificant (r=.111, n=64, p=0.384).

SGPT-

The descriptive table show below some useful descriptive statistics like Mean, std. Deviation, and 95% confidence Interval for dependent variable (SGPT) for different age group (1-10,11-20,21-30) .**Table 2:**

Table 2:The descriptive table show below the SGPT Level of different age group

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
1-10	17	50.335	46.4452	11.2646	26.455	74.215	10.0	159.0
11-20	25	54.160	41.2530	8.2506	37.132	71.188	9.0	169.0
21-30	22	45.182	29.8530	6.3647	31.946	58.418	5.0	121.0
Total	64	50.058	38.8359	4.8545	40.357	59.759	5.0	169.0

The table shows the output of the ANOVA analysis and whether there is a statistically significant difference between our group means. We can see that the P-value (Sig value) is .737, which is above 0.05. So there is no Significant difference found in SGPT with three age groups (1-10,11-20,21-30). Similar analysis was done for SGOT of different age group, and shows that the p-value(.479), that was also insignificant.

Independent samples t-test of SGPT on different sex

The table below shows independent samples t-test. Our level of significance is 0.05.and equal variance assumed. Our null hypothesis is mean SGPT for male and female are equal, and alternative hypothesis is mean SGPT of male and female is not equal.

		(1) Levene's Test for Equality of Variances		(2) t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
SGPT	Equal variances assumed	.082	.776	.043	62	.966	.4210	9.8301	-19.2291	20.0711
	Equal variances not assumed			.043	60.372	.966	.4210	9.8301	-19.1887	20.0306

Table 4: FERITINCAT * AGECAT Cross tabulation

			AGECATEGORY			Total
			0-10	11-20	21-30	
FERRITIN CATEGORY	0-1000	Count	12	19	14	45
		Expected Count	12.0	17.6	15.5	45.0
	1000-2000	Count	3	4	5	12
		Expected Count	3.2	4.7	4.1	12.0
	2000-3000	Count	2	2	3	7
		Expected Count	1.9	2.7	2.4	7.0
Total		Count	17	25	22	64
		Expected Count	17.0	25.0	22.0	64.0

Table 5: **Chi-Square Tests**

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	.907a	4	.924
Likelihood Ratio	.911	4	.923
Linear-by-Linear Association	.215	1	.643
N of Valid Cases	64		

When reading this table we are interested in the results of the "**Pearson Chi-Square**" row. We can see here that $\chi(4) = .907$, $p = .924$.This tells us that there is no statistically significant association between FERRITIN CATEGORY and AGE CATEGORY. Here we can see that ,The strength of association between two variables is very weak.

Table 6 : ANOVA table showing SGPT of different ferritin group					
	Sum of Squares	**df**	**Mean Square**	**F**	**Sig.**
Between Groups	2432.310	2	1216.155	.801	.453
Within Groups	92585.967	61	1517.803		
Total	95018.276	63			

From the above table we can see that the significance value is 0.453(p-value), which is above 0.05, and therefore we can conclude that there is no significant difference found in SGPT with three ferritin group.

Discussion

Regular blood transfusion and iron chelation therapy have improved the life expectancy in patients with transfusion dependent thalassemia and now they survive beyond the third decade of life. Liver derangement is becoming an important cause of morbidity and mortality in these patients. Viral infections (mainly hepatitis B and C) causing chronic hepatitis and/or severe iron overload are both important causes of liver pathology. Iron chelation with Deferasirox (DFX), an oral single dose therapy, has improved the compliance to chelation which reduces excessive body iron [11].

Liver enzymes are raised and indicative of liver injury in transfusion dependant β-thalassemia major patients [12]. In thalassemia, abnormal liver function

appears to be related to the high ferritin levels and the age when transfusions was initiated.[13-17]In thalassemia, liver is the earliest organ affected by iron and serum SGOT and SGPT are raised due to peroxidative injury and direct toxic effect of iron on liver cells[12]. So this study was conducted to know the derangement of liver enzymes and their correlation with the age and serum ferritin levels.

In our study on 64 thalassemic patient, we found raised SGOT and SGPT only in 33 and 31 patients respectively. They had no correlations with age with p value0.479 and0.737 respectively.

Reduction of serum ferritin concentration was associated with a significant decrease in serum ALT, AST and ALP concentration[11]. In a study by Soliman A et al, they found a significant correlations between serum ferritin concentrations and ALT and AST levels ($p < 0.01$)[11].

Another study by Suman RL et al showed as Iron deposition in liver takes place, its functions are affected which are predicted by raised SGOT and SGPT. SGOT and SGPT were raised significantly (p Value <0.05) and continue to rise as ferritin crosses 1000 ng/ml. There was a positive correlation between serum ferritin and liver enzymes (Pearson's bivariate correlation coefficient r=+0.87±84)[12].

But in our study we found raised ferritin level(1000- 2000 ng/ml) in 12 patients and 7 patients in 2000 -3000 ng/ml. Still we could not found any correlation of it with LFT (Mainly transaminases). The p value was 0.453 for SGPT.

In those two studies, they found raised SGOT and SGPT as their ferritin level were very high in all patient, whereas we found raised ferritin level in very few patient. It may be due to our all patients are routinely checked for serum ferritin and they received chelation therapy when ferritin level goes above 500 ng/ml.

Conclusion:

Iron overload can cause liver injury which causes raised liver enzymes. We failed to show this. Like us, some investigator also found such weak correlation [18] as exact mechanisms are still unclear. So, further detailed studies needed to find out the promising correlations in transfusion dependent thalassemic patients.

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References

1. Weatherall DJ, Clegg JB, Thalassaemia - a global public health problem. Nat Med, 1996:2,847-9.
2. Panda PK,SharmaY.Ferritinlevel:Predictor of thalassemia cardiomyopathy.Heart India 2018;6: 18-21.
3. Galanello R, Origa R. Beta-thalassemia. Orphanet J Rare Dis. 2010 May 21;5:11.
4. Firkin F, Chesterman C, Penington D, Rush B.(eds) 1996, de Gruchy,s Clinical Hematology in Medical Practice.5th ed, Blackwell, Australia,137.
5. Hoffbrand AV, Cohen A , Hershko C. Role of deferiprone in chelation therapy for transfusion iron overload. Blood 2003; 102: 17- 24
6. N Sultana1, S Sadiya2, MH Rahman1. Correlation between serum bilirubin and serum ferritin Level in thalassaemia patients. Bangladesh J Med Biochem 2011; 4(2): 6-12
7. Khan WA, Banu B, Amin SK et al. prevalence of beta thalassaemia trait and HB-E trait in Bangladeshi school children and health burden of thalassaemia in our population.2005; 21(1): 1-7.
8. Voskaridou E, Douskou M, Terpose E, et al. magnetic resonance imaging in the evaluation of iron overload inpatients with ? - thalassaemia and sickle cell disease. BrJ Haematol 2004; 126: 736 - 742.
9. Hershko C, Konijn AM, Link G. Iron chelators for thalassaemia. Br J Haematol 1998; 101: 399 - 406.
10. Olivieri NF, Brittenham Gm: Iron- chelating therapy and the treatment of thalassaemia. Blood 1997; 89: 739.

11. 1.Soliman A, Yassin M, Al Yafei F, Al-Naimi L, Almarri N, Sabt A, De Sanctis V. Longitudinal Study on Liver Functions in Patients with Thalassemia Major before and after Deferasirox (DFX) Therapy.Mediterr J Hematol Infect Dis. 2014 Apr 7;6(1):e2014025. doi: 10.4084/MJHID.2014.025. eCollection 2014.
12. Suman RL, Sanadhya A, Meena P, Goyal S. Correlation of liver enzymes with serum ferritin levels in β-thalassemia major. Int J Res Med Sci 2016;4:3271-4.
13. [Ref. of. 13-17] Larson AM, Taylor SL, Bauermeister D, Rosoff L, Jr, Kowdley KV. Pilot Study of the Relationship Between Histologic Progression and Hepatic Iron Concentration in Chronic Hepatitis C. J ClinGastroenterol. 2003;37:406–11. doi: 10.1097/00004836-200311000-00012.
14. Kaddah A, El-Shabrawi M, Saleh A, Al Gaithy A, Basalama A. Liver iron content and histological findings versus serum ferritin in detection of different grades of hemosiderosis in hepatic diseases in thalassemia major. J A C. 2005;6:475–84.
15. de Virgiliis S, Sanna G, Cornacchia G, Argiolu F, Murgia V, Porcu M, Cao A. Serum ferritin, liver iron stores, and liver histology in children with thalassaemia. Arch Dis Child. 1980;55:43–5. doi: 10.1136/adc.55.1.43.
16. Li CK, Chik KW, Lam CW, To KF, Yu SC, Lee V, Shing MM, Cheung AY, Yuen PM. Liver disease in transfusion dependent thalassaemia major. Arch Dis Child. 2002;86:344–7. doi: 10.1136/adc.86.5.344.
17. Vogel E, Lebensburger JD, Bai S, Fineberg N, Hilliard L, Vadlamud N, Dimmit R, Kelly D, Thomas H, Howard TH. Liver histology, liver iron concentration (LIC), and serum ferritin in a large cohort of chronically transfused children with sickle cell anemia: Limitations of LIC as a marker for hepatic injury and ferritin as an indicator for chelation initiation. 53rd ASH annual meeting; 2011. (abstract)https://ash.confex.com/ash/2011/webprogram/Paper42892.html.
18. Asif M, Manzoor Z, Farooq MS, Kanwal A, Shaheen U, Munawar SH, Khan IA, Aziz A. Correlation between serum ferritin level and liver function tests in thalassemic patients receiving multiple blood transfusions. Int J Res Med Sci 2014;2:988-94.