Original Article

Study of Gall Bladder Disease in Sickle Cell Patients at a Tertiary Care Hospital in Vidarbha

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ABSTRACT

The prevalence of sickle cell disease is high in central India and in certain localities of Maharashtra (specially Vidarbha). Sickle cell anemia can affect hepatobiliary system due to chronic ischemia or as a result of chronic haemolysis.

Objective : This case control study was carried out with aim of estimating proportion of different gall bladder disorders in sickle cell patients and to determine the association of various clinical, biochemical and functional factors, with gall bladder diseases in sickle cell patients.

Methods : 65 cases of sickle cell anemia (both AS and SS) were compared with age and sex matched healthy persons regarding gall stones, gall bladder sludge, gall bladder motility, haematological parameters, liver enzymes, cholesterol and BMI. Also sickle cell cases with and without gall bladder disorder were separately compared.

Results : We studied 30 cases of SS pattern and 35 cases of AS pattern. 23 (35.38%) cases had **gallbladder disease** in the form that 17 (26.15%) had gallstone and 6 (9.23%) had gall bladder sludge. The mean gallbladder motility was reduced significantly by 2% in sickle cell cases as compared with healthy controls. The cases with GB stone / sludge were having larger fasting gall bladder volume as compared to cases without gall stone / sludge. Fasting and post meal GB volume and gall bladder motility was significantly affected in sickle cell disease (SS) with gall stone / sludge as compared with cases without gall stones/sludge.

Conclusions : Fasting gallbladder volume and post meal gallbladder volume were increased and gallbladder motility was decreased in patients with sickle cell disease.

Key-words : sickle cell anemia, gall bladder motility, cholelithiasis

Introduction :

Sickle cell disease is the most common structural hemoglobinopathy (HbS). Sickle cell anemia is the commonest symptomatic hemoglobinopathy found in India with the frequency of sickle cell trait 4.3%[1], the frequency of sickle cell trait was much higher (upto 48%) in many tribal populations in India[2]. The prevalence of sickle cell disease is high in central India and in certain localities and certain castes of Vidarbha region of Maharashtra state[3].

Sickle cell disease can affect many parts of body and one of the common organ affected is gallbladder. Gall bladder is affected either directly by sickling causing chronic ischemia (rheological effect) and due to chronic haemolysis. Cholelithiasis is one of the common complication in sickle cell anemia, these pigment gall stones result from chronic hemolysis causing increased bilirubin production[4].

However, many SCD patients with marked hemolysis do not develop gallstones. Thus its not only the excess bilirubin but various other factors including abnormalities in gallbladder function / contraction may contribute to gallstone formation in sickle cell patients. Ultrasonography is a useful non-invasive technique to study changes in gallbladder structure and function. Gallstones are seen as the presence of mobile high-level echoes with acoustic shadows in the gallbladder, and sludge as the presence of mildly echogenic intraluminal sediment in the absence of acoustic shadows. Abnormalities in gallbladder function were assessed by calculating fasting and postmeal gallbladder volume and

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gallbladder motility / contractility. Present study was undertaken to determine the patterns of gallbladder disorders in sickle cell disease and compare them with healthy controls.

MATERIALS AND METHODS -

This hospital based case control study was carried out during the period of January 2011 to October 2012. The Institutional Ethics Board approved the protocol.

The study was camed out in 65 patients of sickle cell disease (30 sickle cell anemia (SS) and 35 Sickle cell trait (AS) patients.

Study Design : Case Control Study at a Tertiary Care Teaching Institute. **Setting** - Sickle Cell clinic (OPD), IGGMC Nagpur. **Sampling Population** - Cases were selected as random sampling from Sickle Cell OPD, Dept. of Medicine, IGGMC Nagpur. Control non probability convenient age and sex matched persons from Medicine OPD coming for minor complaints and healthy volunteers.

Inclusion Criteria for Cases : diagnosed sickle cell disease patients (SS and AS pattern) with age more than 12 years in stable state.

Exclusion Criteria :

1. Hospitalized patients of sickle cell disease in acute crisis and those who had received recent blood transfusion (1 month prior).
2. Diagnosed cases of Diabetes mellitus, hypercholesterolemia and hypertriglyceridemia, Other Hemolytic anemia (thalassemia, hereditary spherocytosis, sickle α / β thalassemia), subjects with BMI more than 25 kg/m^2.
3. Previously or recently diagnosed congenital anatomical anomalies and abnormalities of biliary system.

Inclusion Criteria for Control group a healthy individual. (No family history of sickle cell disease)

STUDY PROTOCOL -

65 cases of sickle cell disease recruited from sickle cell OPD under Department of Medicine and Regional Hemoglobinopathy Detection and Management Centre, IGGMC, Nagpur and 65 controls each satisfying the above said inclusion and exclusion criteria for study were enrolled in the study. Written and informed consent was obtained from subjects. Detailed history regarding general health and gallbladder symptoms was noted. Past history regarding hospitalization, vaso-occlusive crisis, blood transfusions, jaundice were inquired in detail, treatment history and family history was taken. Blood investigations CBC, Hb electrophoresis / HPLC, Liver function test, lipid profile were done in both cases and controls. Statistical analysis was done.

ULTRASONOGRAPHY OF GALLBLADDER was done in all cases. The gallbladder was studied for content, wall, shape, and motility with standardized methodology. Ultrasonographic evaluation of fasting gallbladder volume and post meal gallbladder volume was done in all the subjects using a 3.5 MHz transducer. The probe was placed on the right sub-costal area while the patients were in supine position and angled to obtain images showing largest longitudinal diameter of the gall bladder. A single, experienced observer on PHILIPS HD-11 ultrasound machine at Department of Radiology, IGGMC, and Nagpur recorded all the measurements. Gallbladder motility was assessed by monitoring gallbladder volumes with 8 hrs. of fasting and 1 hour after ingestion of a fixed liquid test meal formulation (post meal) consisted of 200 ml fixed liquid formula (Amul cool badam with high fat 3.1 g, protein 3.2 g and carbohydrate 12 gm in 100 ml). Fat meal being a very good powerful stimulus for GB contraction.[6,7] Gallstones were diagnosed by the presence of mobile high-level echoes with acoustic shadows in the gallbladder, and sludge by the presence of mildly echogenic intraluminal sediment in the absence of acoustic shadows.

Gallbladder motility (in percentage) was calculated by :

$$\frac{\textit{(Fasting gallblader volume - Post meal gallblader volume) x 100}}{\textit{fasting gallbladder volume}}$$

RESULTS -

In this study we included 65 cases with sickle cell disease and 65 age (± 5 years) and sex matched healthy subjects. In cases 33 (50.77%) were male

and 32 (49.23%) were female. The mean age of cases 24.21 ± 7.54 & in control 24.07 ± 7.55 years.

Age wise the number of cases were 21, 28, 13 and 3 in the age group of 12-19, 20-29, 30-39 and above 40 years respectively.

There was significant difference in hemoglobin concentration, liver enzymes, bilirubin and body mass index in sickle cell cases as compared to healthy controls. The difference in total cholesterol, gall bladder wall thickness and MCV was not highly significant. In present study statistically significant difference ($p < .05$) was found in mean fasting gallbladder and mean postmeal gallbladder volume in cases as compared to controls. The mean gallbladder motility was significantly reduced in sickle cell disease than healthy controls. ***(Table 1)***

In this study gallbladder disorder in the form of gall stone and sludge was found in 23 (35.38%) cases.14 (46.66%) of SS and 3 (8.57%) of AS. Occurrence of gall stones was higher in SS cases. Gall bladder sludge was found in 10% SS and 8.5% in AS cases. 13 (43.33%) cases with SS pattern had not developed gall stones/sludge. ***(Table 2)***

The difference in total bilirubin, indirect bilirubin, AST, ALT, alkaline phosphatase, fasting and post meal gall bladder volume were highly significant in cases with and without gall stone / sludge. Gall bladder wall thickness was found significantly less in cases with gall stone / sludge.

Hb%, BMI and total cholesterol was not significantly associated.

Total Bilirubin was 2.70 ± 0.93 mg % in cases with gall stones & sludge and 1.02 ± 0.92 mg % in those without gall stones & sludge. The difference in means was also statistically highly significant. ($p < 0.05$)

Table 1 : **Comparison of various factors associated with gall stone / sludge formation in cases and controls**

	Cases (n = 65)		Controls (n = 65)		p-value
	Mean	SD	Mean	SD	
Hb (gm%)	8.27	1.13	10.76	0.94	**p=0.001***
MCV	81.09	6.81	83.40	5.53	**P=0.03**
AST (U/L)	28.27	10.61	22.09	6.44	**p=0.001***
ALT (U/L)	29.84	10.36	22.98	5.85	**p=0.001***
ALP (U/L)	162.75	51.14	131.01	12.94	**p=0.001***
Total Bilirubin (mg%)	1.61	0.93	0.77	0.17	**p=0.001***
T Cholesterol	153.53	14.92	160.46	13.01	p=0.05
GB wall thickness (mm)	1.29	0.45	1.45	0.5	p=0.05
BMI	21.67	1.69	23.2	1.38	p=0.0001*
Fasting gall bladder cc	27.43	6.43	19.2	1.71	**p=0.001***
Post meal gall bladder Volume	4.29	1.11	2.66	0.64	**p=0.001***
Gall bladder motility %	84.22	2.83	86.17	2.87	**p=0.001***

Table 2 : **Distribution of different gallbladder diseases in sickle cell cases**

Gallbladder Disorders in cases (n=65)	Sickle cell disease (SS pattern) (n-30)	Sickle cell trait (AS pattern) (n-35)
Gallstone = 17 (26.1%)	14 (46.66%)	3 (8.57%)
Gall bladder Sludge = 6 (9.2%)	3 (10%)	3 (8.57%)
Total = 23 (35.38%)	17 (56.66%)	6 (17.14%)

The fasting gall bladder volume in cases with gall stone & sludge was 34.76 ± 1.98 cc as compared to 23.21 ± 3.15 cc in cases without gall stones & sludge. The difference in the means was highly statistically significant. The post meal gall bladder volume in cases with gall stone & sludge was 5.05 ± 0.74 cc as compared to 3.80 ± 0.89 cc in cases without gall stones & sludge. The difference in the means was statistically highly significant.

The motility of gall bladder in cases with gall stone & sludge was significantly high as compared to cases without gall stones & sludge. The difference in the means was statistically significant 0.02. (P < 0.05) ***(Table 3)***

Sickle cell disease (SS) pattern cases were separately analysed as follows. ***(Table 4)***

Table 3 : **Comparison of various factors in sickle cell cases with gall stones / sludge and without gallstone / sludge**

	Cases with gall stones / sludge (n=23) Mean ± SD	Cases without gall stones / sludge (n=42) Mean ± SD	P value	T value
Age (years)	26.78 ± 7.54	23.5 ± 5.79	0.05	1.99
BMI	21.52 ± 1.69	21.75 ± 1.71	0.6	
Hb%	7.80 ± 1.13	8.53 ± 1.14	0.01	2.47
T Cholesterol	155.43 ± 14.92	150.95 ± 13.87	0.22	
T Bilirubin	2.70 ± 0.93	1.02 ± 0.92	0.0001	t=7.01
I Bilirubin	1.28±0.39	0.65±0.38	0.0001	t=6.35
AST	38.6±10.61	22.61±9.8	0.0001	t=6.1
ALT	39±19.74	24.83±10.2	0.0003	t=3.82
Alk Phos mg/dl	212.86±51.14	135.3±51.63	0.0001	t=5.84
GB wall thickness (mm)	1.0	1.45	0.0001	t=4.3
Fasting volume	35.13 ± 6.43 cc	23.21 ± 6.44 cc	0.0001	T=7.13 (CI 8.58-15.25)
Post meal volume	5.17 ± 1.11 cc	3.80 ± 1.10 cc	0.0001	T=4.78 (CI=0.79-1.94)
GB motility %	85.28 ± 2.83	83.63 ± 2.87	0.02 (<0.05)	T=2.22 (CI=0.16-3.13)

Hb :Hemoglobin, MCV : Mean corpuscular volume, TLC : Total leucocyte count, AST : Aspartate transaminase, ALT : Alanin transaminase, ALP : Alkaline phosphatase, BMI : body mass index)

** indicates statistically significant p value*

Table 4 : **Intra group comparison of Sickle cell disease SS cases with or without gallstone / sludge**

	Sickle cell disease SS with GB Stone / sludge (n=17) mean ± SD	Sickle cell disease SS without gall stone / sludge (n=13) mean ± SD	Healthy controls (n=65) mean ± SD	p value
Fasting gall bladder volume	35.23±5.71cc	25.46±5.66cc	19.2±1.71 cc	0.0001
Post meal gall bladder volume	7.88±1.69cc	5.15±1.72 cc	2.66±0.64 cc	0.0001
Gall bladder contractility/motility	77.6±2.58	79.86±2.52	86.17±2.87	0.023 (<0.05)
GB wall thickness	1±0.25 mm	1.25±0.27 mm	1.45±0.5 mm	0.12
Total bilirubin	2.8±0.93mg/dl	1.3±0.96mg/dl	0.77±0.17 mg/dl	0.0002

Sickle cell cases with gall stone / sludge had significantly higher fasting and post meal gall bladder volume as compared to those cases without gall stones / sludge and healthy controls. Total bilirubin was also significantly high in sickle cell disease with GB stone / sludge as compared to those SS cases who had not developed gall stones and healthy controls.

DISCUSSION :

There is very frequent association of gall bladder disease in sickle cell patients.[8,9,10]. Eversion GT[11] in his study showed that there was increased prevalence of gall bladder dysfunction among sickle cell patients and they tend to have larger gallbladder and reduced responsiveness to meals affecting gall bladder contractility / motility which might lead to development of stasis of bile and development of complications like sludge, cholelithiasis and cholecystitis. We also included AS (Sickle cell trait) cases which were not included in other studies.

Gall bladder motility / contractility was decreased in sickle cell cases as compared with controls. This might be of possible mechanism causing stasis of bile and precipitation of biliary pigments into gallstone. Gall bladder stores and concentrate bile salt during fasting and release them into duodenum in response to gastric emptying of meal. The flow of bile into duodenum is controlled by gall bladder muscle contraction, gall bladder tone in fasting, hepatic secretary pressure and relaxation of sphincter Oddi. There may be problem with gall bladder smooth muscle contractility which may be due to diminished response or impaired secretion of a gut peptide CCK-PZ (Cholecystokinin pancreozymin).[11,12] Defective smooth muscle contractility and / or relaxation are found in cholesterol stone-containing gallbladders, featuring a type of gallbladder leiomyopathy; defects of CCK receptors.[13]

Study observed that some sickle cell cases particularly having lesser fasting and post meal gall bladder volume and bilirubin did not developed gall stone / sludge hence we compared these two groups separately.

Those sickle cell SS cases having fasting gall bladder volume 35.23 ± 5.71 cc, post meal gall bladder volume 7.88 ± 1.69 cc developed gall stones / sludge. Bilirubin concentration was also significantly high 2.8 ± 0.93 mg / dl in these SS cases with gall stone / sludge.

CONCLUSIONS :

1. Sickle cell cases had larger fasting gall bladder volume. (GB muscle tone decreased)
2. Sickle cell cases had increased post meal gall bladder volume. (impaired GB contraction)
3. Sickle cell cases had decreased gall bladder motility in response to meals.
4. Those sickle cell cases with lower Hb%, higher bilirubin, higher fasting and post meal gall bladder volume developed gall stones / sludge.

Limitations :

1. Factors responsible for gall bladder contractility like gut hormone CCK-PZ needed to be studied.
2. Study had small sample size, larger studies will confirm these findings.

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