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STUDY OF RENAL PROFILE IN PATIENTS OF SICKLE CELL DISEASE

Nephrology

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ABSTRACT

Introduction: Sickle cell disease (SCD) or sickle cell anemia (SCA) is a hereditary genetic disease that is characterized by the presence of abnormal crescent-shaped red blood cells.[1] It is characterized by the presence of sickle hemoglobin (Hb S), caused by a single point mutation involving GAG - GTG transversion at codon 6 of the β-globin gene.[2-6]

Aim & Objective: The study was undertaken to study the renal profile in patients of SCD and to compare the renal profile in patients of SCD with traits.

Materials & Methods: It was a prospective cross sectional study, conducted in the department of Medicine on patients of sickle cell disease coming to medicine department or speciality OPD of sickle cell disease, of Acharya Vinoba Bhave Rural Hospital (AVBRH), a tertiary care hospital attached to Jawaharlal Nehru Medical College (JNMC), Sawangi, Wardha in Central India. It was conducted for 2 years from September 2014 – August 2016.

Results: Tubular dysfunction was more in patients with SS pattern than AS patttern. In our study, tubular dysfunction was found in 100% cases of SS pattern with crisis. Urine microscopy of SCD patients showed evidences of glomerular & tubular damage, papillary necrosis and tubulointerstitial nephritis. Hyposthenuria was a predominant finding in patients of SCD patients. >75% of SCD patients had hyposthenuria, suggesting that renal medulla in patients of crisis had developed ischemia, so passing large amount of diluted urine which further contribute to the development of dehydration leading to further renal ischemia and complications.

Conclusion: Renal involvement in the form of glomerular & tubular dysfunction occur in patients of Sickle Cell Disease (SCD).

KEYWORDS

Renal profile in patients of sickle cell disease, FENa, FE_{urea}

Introduction

Definition and Introduction:

Sickle cell disease (SCD) or sickle cell anemia (SCA) is a hereditary genetic disease (autosomal recessive) that is characterized by the presence of abnormal crescent-shaped red blood cells.[1]

It is characterized by the presence of sickle hemoglobin (Hb S), caused by a single point mutation involving GAG - GTG transversion at codon 6 of the β-globin gene.[2–6] It affects individuals of African, Mediterranean, Indian, Middle-Eastern, Caribbean, South and Central American ancestry.

Types of SCD:

Sickle Cell Anemia is mainly of 2 types:

- Homozygotes for Hb S have sickle cell anemia (Hb SS).
- Heterozygotes for Hb S have sickle trait (Hb AS).

Other variants include sickle C disease and sickle thalassemia.

Clinical symptoms or manifestations of SCD include hemolysis, anemia, pain crisis, infection, acute chest syndrome and vascular occlusion. All these manifestations potentially lead to repeated ischemic attacks and organ damage.[7,8]

Renal Involvement in SCD:

The kidney of the homozygous sickle cell anemia (Hb SS) patient is affected by the hemodynamic changes of chronic anemia and by the consequences of vaso-occlusion, especially in the renal medulla.[9,10] The disruption of distal nephron and medullary function leads to a reduction of renal concentrating capacity, urinary acidification defect and impaired potassium metabolism which are often observed in these patients.[11,12]

About 5% of the world population is the carriers of gene for sickle cell anemia. Percentage of the disease is high in Asia, the Mediterranean basis, the Middle East and Africa.[13] Few studies reported high mortality rate in children with sickle cell disease during the first five years of life due to infection and splenic sequestration.[13]

Early diagnosis and inclusive care can reduce morbidity and mortality rate significantly in patients with sickle cell disease.[13]

Central India, which is one of the high prevalence regions for the βs gene, has a prevalence rate of 9.4-22.2% for this mutation in various communities.[14,15]

Rationale for the study:

The Vidarbha region is known for sickle cell anemia, and very few studies have been done in our region on sickle cell anemia and renal status. As kidney is an end organ, any damage to it can cause fatal complications and death.

Early diagnosis of sickle cell nephropathy and effective management of vaso-occlusive crisis and measures to prevent crisis can certainly reduce the morbidity and mortality.

Very little data is available from Central India regarding the clinical profile of sickle cell nephropathy. Therefore the study is being undertaken.

Aim and Objective:

AIM

To study renal profile in the patients of sickle cell disease.

OBJECTIVE

To compare renal profiles of patients with sickle cell disease and traits.

Materials and Methods

This study was conducted in the department of Medicine on patients of sickle cell disease coming to medicine department or speciality OPD of sickle cell disease, of Acharya Vinoba Bhave Rural Hospital (AVBRH), a tertiary care hospital attached to Jawaharlal Nehru Medical College (JNMC), Sawangi, Wardha in Central India.

It was a prospective cross sectional study, and was conducted for 2 years from September 2014 – August 2016. Total 50 cases were studied. The protocol of this study was revised and approved by the Institutional Ethics Committee (I.E.C.) of the JNMC Sawangi (Annexure I).

Total 56 patients were selected. Among them 4 were having systemic illnesses and 2 were not willing for consenting, were excluded. Patients were included in the study after signing the written informed consent form (Annexure II).

Inclusion Criteria:

All the patients, irrespective of their age and sex, suffering from sickle cell disease proven on Hb electrophoresis with SS or AS pattern coming to medicine department.

Exclusion Criteria:

- Patients suffering from any systemic disease like Diabetes Mellitus (DM), systemic hypertension, Systemic Lupus Erythematosus (SLE), which could affect renal structure and function.
- Sickle cell patients on diuretics.
- Sickle cell patients having obstructive uropathy due to renal calculus disease.
- Not consenting

Data collection:

A detailed history of patients fulfilling the above mentioned inclusion criteria regarding symptomatology and clinical examination related to SCD and renal dysfunction, were recorded in proforma (Annexure III).

Methodology:

With all aseptic precautions, blood was collected from median cubital with a disposable syringe. 2 ml blood was required for CBC (EDTA bulb), 2 ml blood was required for KFT (plain bulb), and 1 ml blood was required for RBS (Fluoride bulb). Blood was transported to laboratory [Central Clinical Laboratory (CCL) of AVBRH] in sterile bulbs for investigations immediately.

Clean catch urine samples were collected from all the participants by the standard spontaneous voiding procedure. The collection of urine was done at noon time in order to avoid its contamination by contents of urethra or vagina (i.e. false results) and therefore its constituents more likely reflect kidney origin. First void samples were also taken as they are more informative and concentrated samples. All the samples of urine were collected in a sterile, labelled bottle containing 4% formaldehyde; which was then transported to the laboratory [Central Clinical Laboratory (CCL) of AVBRH] immediately.

- CBC was directly processed by coulter machine (Make : Beckman coulter AC-T 5 differential).
- For KFT & RBS, sample was first centrifuged by centrifuge machine REMI R8C (Make: REMI). Randox Daytona analyser (make/model: Randox clinical chemistry) was used for processing of KFT & RBS.
- Urine samples were first physically observed (physical examination of urine) for colour, turbidity, smell. Specific gravity was measured by urinometer (Make: Davinder Glass works, Delhi, India). pH was be measured by pH paper (Make: SD fine). For urine processing YD Uriscan Optima machine was also used, and mean of both results were noted. Albuminuria was measured by standard dip stick method.
- For routine microscopic examination of urine, urine samples were centrifuged by centrifuge machine REMI R8C (Make: REMI). Sediments of urine were taken on slide and covered by cover slip. Urine microscopic examination done by 10X and 40X (High Power Field) (HPF) and results were noted.
- Biochemical estimations such as estimation of potassium ion, sodium ion, urea and creatinine, were done Randox Daytona analyser (make/model: Randox clinical chemistry).

Following formulae were used to calculate GFR, FENa and FEurea. and results were noted in proforma.

GFR (ml/min) = [140-age] x body wt/72 x Sr. Cr. { x 0.85 for female}.[16]

FENa % = 100 x [urinary Na x Sr. Cr.]/[Sr. Na x urinary Cr.].[17]
Feurea % = 100 x [urinary urea x Sr. Cr.]/[blood urea x urinary Cr.].[18]

Normal Range of various parameters (According to AVBRH Central Laboratory Standard)

Parameters	Normal Range	Parameters	Normal Range
Hemoglobin (Hb)	(M- 13-15.5) (F-11-13.5) gm %	Random Blood Sugar (RBS)	70 - 140 mg%
Total Leukocyte count (TLC)	4000-11000 /cmm	Urinary pH	4.5-8
Platelet	1.5-4.5 L/cmm	Urinary Specific gravity	1.015-1.026
Blood Urea	18 - 40 mg%	Urinary Urea (U_{urea})	350-1000 mg%
Sr. Creatinine	0.7 - 1.5 mg%	Urinary Creatinine (U_{Cr})	20-28 mg%
Sr. Sodium (Na^+)	136 - 145 mEq/L	Urinary Sodium (U_{Na+})	40-220 mEq/L
Sr. Potassium (K^+)	3-5.5 mEq/L	Urinary Potassium (U_{K+})	25-125 mEq/L

Observations and Results

[Table 1 shows baseline characteristics of SCD patients.]

Study group comprised of total 50 cases of sickle cell disease patients. Out of which 30 had AS pattern and 20 had SS pattern. Mean age of total study population was 26.44±9.45 years, while that of AS pattern was 26.40±8.94 years and that of SS pattern was 26.50±10.42 years. p value (0.97) was not significant. Mean Hb concentration of total cases was 9.27±1.78 gm%. Patients with AS pattern had mean Hb concentration of 9.47±1.83 gm%, while the patients with SS pattern had mean Hb concentration of 8.98±1.70 gm%. p value (0.34) was not significant. Mean GFR of the study population was 81.34±22.49 ml/min, while that of AS pattern was 79.07±22.50 ml/min and of SS pattern was 84.76±22.61 ml/min. p value (0.38) was not significant. Mean urinary pH of total cases was 4.92±0.68. Patients with AS pattern had mean urinary pH of 4.98±0.72, while the patients with SS pattern had mean urinary pH of 4.83±0.62. p value (0.45) was not significant. Mean urinary specific gravity of total cases was 1.010±0.007. Patients with AS pattern had mean urinary specific gravity of 1.012±0.008, while the patients with SS pattern had mean urinary specific gravity of 1.008±0.006. p value (0.08) was not significant.

[Table 2 shows gender wise distribution of sickle cell patients.]

Total 50 patients were studied out of which males were 21 (42%) and females were 29 (58%). Out of 30 patients with AS pattern, maximum cases, i.e. 21 cases (70%) were females, while among 20 patients with SS pattern, maximum cases, i.e. 12 cases (60%) were males.

Male:Female ratio in total study population was 0.72:1, while that of AS pattern was 3:7 and that of SS pattern was 1.5:1.

The result was statistically significant. (18.18, p=0.0001, S).

[Table 3 shows age wise distribution of sickle cell patients.]

The study group comprised of age group of 13-56 years, with the mean age of total study population of 26.44±9.45 years. Maximum number of cases (22 cases i.e. 44%) were clustered under the age group of 21-30 years. There were only 3 (6%) cases > 40 years. Among them 2 cases (6.67%) were of AS pattern and 1 case (5%) was of of SS pattern. The result was not statistically significant. (2.96, p=0.39, NS).

[Table 4 shows age and gender wise distribution of sickle cell patients.]

Of total 21 males, maximum number of male cases, 8 cases (38.10%) were in the age group of 11-20 years, and of total 29 females, maximum number of female cases, 15 cases (51.72%) were in the age group of 21-30 years.

The result was statistically significant. (12.25, p=0.0066, S).

[Table 5 shows gender wise distribution of patients with sickle cell crisis.]

Of total 50 cases of SCD patients, 12 patients with sickle cell crisis were studied. Male:Female ratio in total study population with crisis was 1:1. i.e. 6 cases (50%) were males and 6 cases (50%) were females. All 4 cases (100%) of AS pattern with crisis were females, while maximum number of cases, 6 cases (75%) were males out of 8 cases of SS pattern with crisis.

The result was statistically significant. (22.47, p=0.0001, S).

[Table 6 shows age and gender wise distribution of SCD patients in crisis.]

Of 12 patients of sickle cell crisis, maximum number of patients, 5 patients (41.67%) were in the age group of 21-30 years followed by 4

patients (33.33%) in the age group of 11-20 years. All the 4 patients of AS pattern with crisis were females. Among them 2 (50%) were in the age group of 21-30 years.There were no patients with sickle cell crisis in the age group of above 40 years in the present study.

The result was statistically significant. (29.45, p-value=0.0001, S).

[Table 7 shows anemia in patients of SCD.]

According to WHO criteria, patients were categorized into different classes of Anemia.

41 cases (82%) had anemia, among them 4 (8%) were severely anemic. In the present study the lowest Hb% was 5.5 gm% and the highest Hb% was 13.9 gm%; with the mean Hb concentration of 9.27±1.78 gm%. Most of the cases, 31 cases (62%) of total 50 patients in our study had moderate anemia. Of those 31 cases 16 cases (53.33%) were of AS pattern, and 15 cases (75%) were of SS pattern.

The result was statistically significant. (12.71, P=0.0053, S).

[Table 8 shows gender wise distributions of anemia in patients of SCD.]

12 males (57.14%) out of 21 males, 19 females (65.52%) out of 29 females had moderate anemia. 4 males (19.05%) out of 21 males, 2 females (6.90%) out of 29 females had mild anemia. There were total 4 (8%) cases who had severe anemia, among them 1 (4.76%) was male and 3 (10.34%) were females. Moderate to severe anemia was more in females than males, whereas males mostly had mild to moderate anemia.

The result was statistically significant. (8.80, P=0.031, S).

[Table 9 shows FE_{Na} in patients of SCD.]

Abnormal fractional excretion of sodium (FE_{Na}) was found in 36 (72%) out of 50 patients of SCD. Of those 36 patients, 16 (80%) were of SS pattern and 20 (66.67%) were of AS pattern.

The result was statistically significant. (4.33, P=0.037,S).

The above mentioned observations suggest that tubular dysfunctions occured in patients of SCD with both AS as well as SS pattern. It was more in patients with SS pattern than AS pattern.

[Table 10 shows FE_{Na} in patients of sickle cell crisis.]

7 (58.33%) out of 12 patients of sickle cell crisis had abnormal FE_{Na}. Among the 4 patients with AS pattern with crisis, 2 cases (50%) had abnormal FE_{Na}. Similarly out of 8 patients with SS pattern with crisis, 5 (62.5%) had abnormal FE_{Na}.

The result was not statistically significant. (3.43, P=0.06, NS).

In our study, it was observed that there was no significant difference in tubular dysfunction in patients with AS & SS pattern in crisis.

[Table 11 shows FE_{urea} in patients of SCD.]

43 cases (86%) of total 50 cases had abnormal FE_{urea} (Fractional excretion of urea).

6 cases (20%) of total 30 cases of AS pattern had normal FE_{urea}. On the other hand only 1 case (5%) out of 20 cases of SS pattern had normal Fe_{urea}.

Remaining patients i.e. 24 cases (80%) of AS pattern , and 19 cases (95%) of SS pattern had abnormal FE_{urea} indicating tubular damage.

In our study, tubular dysfunction was found in 80-95% of cases. Renal involvement was more in SS pattern than AS pattern.

The result was statistically significant. (10.29, P=0.0013, S).

[Table 12 shows FE_{urea} in patients of sickle cell crisis.]

Out of 12 patients of sickle cell crisis 10 patients (83.33%) had abnormal Fe_{urea}.

2 cases (50%) of AS pattern in crisis, and all the 8 (100%) cases of SS pattern in crisis had abnormal Fe_{urea}.

The result was statistically significant. (66.67, P=0.0001, S).

In our study, it was observed that tubular dysfunction occured in patients of SCD with both AS as well as SS pattern in crisis. It was more in patients with SS pattern in crisis than AS pattern in crisis. 100% cases of SS pattern with crisis had renal dysfunction in the form of tubular damage.

[Table 13 shows physical and microscopic urinary findings in patients of SCD.]

12 cases (24%) of total 50 cases had significant microscopic albuminuria. Among them 6 cases (20%) were of AS pattern and remaining 6 cases (30%) were of SS pattern. There was no significant difference between the two groups.

Microscopic hematuria was a predominant finding in patients of SS pattern i.e. 5 cases (25%) of total 20 cases. Similarly cast and crystals were found in 5 cases (25%) of SS pattern. On the other hand, among the patients with AS patern 5 cases (16.67%) had hematuria and 7 cases (23.33%) had cast and crystals in their urine microscopy. Of total population, 5 patients (10%) had RBC casts, while 4 patients (8%) had granular casts. 3 cases (15%) of SS pattern had significant number of pus cells in their urine. 6 cases (30%) of SS pattern and 4 cases (13.33%) of AS pattern had epithelial cells in urine.

Above mentioned findings in SCD patients might be the result of glomerular and tubular damage with papillary necrosis. Also these findings could be secondary to interstitial nephritis.

The result was statistically significant. (8.56, P=0.003, S).

[Table 14 shows physical and microscopic urinary findings in patients of sickle cell crisis.]

Out of 12 patients with sickle cell crisis, 9 cases (75%) had significant albuminuria, among which 3 cases (75%) were of AS pattern with crisis and 6 cases (75%) were of SS pattern with crisis.

3 cases (75%) of AS pattern with crisis had hematuria, while 4 cases (50%) of SS pattern with crisis had hematuria. Suggesting that hematuria occured in both sickle cell trait and disease. Suggesting that patients with sickle cell crisis had glomerular and tubular dysfunction along with papillary necrosis.

The result was statistically significant. (13.33,P=0.0003,S).

All the 4 cases (100%) of AS pattern with crisis had casts and crystals. There were 5 cases (62.5%) of SS pattern with crisis who had casts and crystals. Casts were mostly RBC cast, followed by granular casts. Suggesting that patients with sickle cell crisis had nephritis or glomerular or tubular damage.

The result was statistically significant. (45.40, P=0.0001,S).

There were 4 cases (75%) of SS pattern with crisis who had tubular epithelium or casts in their urine microscopy, suggesting tubular damage; while none of the patients of AS pattern had tubular epithelium in their urine microscopy.

Renal complications in sickle cell disease result from occlusion of the vasa recta in the renal medulla. Sequelae include papillary necrosis, nephritis and glomerular and tubular damage leading to hematuria, proteinuria, hyposthenuria, acidification defect, and increased chances of UTI complicating to pyelonephritis.

The result was statistically significant. (66.67, P=0.0001, S).

[Table 15 shows specific gravity of urine in patients of SCD.]

Of 50 SCD patients 12 patients (24%) had normal urinary specific gravity, while 38 patients (76%) had abnormal urinary specific gravity. Hyposthenuria was a predominant finding in patients of sickle cell

disease. Among 36 patients of hyposthenuria, 19 cases (63.33%) were of AS pattern and 17 cases (85%) were of of SS pattern. These findings suggestive of renal medullary ischemia. The result was statistically significant. (12.58,P=0.0004,S).

Only 2 cases (66.67%) had hyperosthenuria. Both of them had AS pattern.

The result was statistically significant. (100.8, P=0.0001, S).

[Table 16 shows specific gravity of urine in patients of sickle cell crisis.]

In 12 patients with sickle cell crisis 9 (75%) had developed hyposthenuria. Out of which 3 cases (75%) were out of 4 cases of AS pattern and 6 cases (75%) were out of 9 cases of SS pattern.

None of the patients of sickle cell crisis had hyperosthenuria, suggesting that renal medulla in patients of crisis had developed ischemia, so they were passing large amount of diluted urine, which further contribute to the development of dehydration, leading to further renal ischemia and complications.

Discussion

The present study was conducted in the department of Medicine on patients of sickle cell disease coming to medicine department or speciality OPD of sickle cell disease, of Acharya Vinoba Bhave Rural Hospital (AVBRH), a tertiary care hospital attached to Jawaharlal Nehru Medical College (JNMC), Sawangi, Wardha in Central India. It was a prospective cross sectional study. The study was conducted for 2 years from September 2014 – August 2016. Total 56 patients were selected. Among them, 4 were having systemic illnesses and 2 were not willing for consenting were excluded. Remaining 50 cases who satisfied the inclusion criteria were included in study and their complete clinical profile was studied.

In our study total 50 cases were studied, among them 30 (60%) had AS pattern, while 20 (40%) had SS pattern. The study group comprised of age group of 13-56 years, with the mean age of total study population of 26.44±9.45 years. This correspond well with the demographic pattern of population in the country. The reasons for the low prevalence in higher age was small sample size of the study, and the fact that, most of the sickle cell patients succumbed due to the disease in early ages. Maximum number of cases (22 cases i.e. 44%) were clustered under the age group of 21-30 years. Maximum number of cases of individual groups, i.e. 14 cases (46.67%) out of 30 patients of AS pattern and 8 cases (40%) out of 20 patients of SS pattern, both were in the age group of 21-30 years.

There were only 3 (6%) cases >40 years. Among them 2 cases (6.67%) were of AS pattern and 1 case (5%) was of of SS pattern.

These findings of the present study correlate more or less with the findings of Umesh L Dhumne, et al. (2011)[19]; Arun U Deore, et al. (2013)[20] ; Andhale RB, et al. (2014)[13]; Arun U Deore, et al. (2014)[21]; Shafeer VP, et al. (2014)[22] and Alkhunaizi AM, et al. (2014)[23].

Of the 50 cases, males were 21 (42%) and females were 29 (58%). Out of 30 patients with AS pattern, maximum cases, i.e. 21 cases (70%) were females, while among 20 patients with SS pattern, maximum cases, i.e. 12 cases (60%) were males. Male:Female ratio in total study population was 0.72:1, while that of AS pattern was 3:7 and that of SS pattern was 1.5:1.

Of total 21 males, maximum number of male cases, 8 cases (38.10%) were in the age group of 11-20 years, and of total 29 females, maximum number of female cases, 15 cases (51.72%) were in the age group of 21-30 years.

These findings of the present study correlate more or less with the findings of Sesso, et al. (1998)[24]; Abdu A, et al. (2011)[25] ; Umesh L Dhumne, et al. (2011)[19]; Mpalampa, et al. (2012)[26] and Shafeer VP, et al. (2014)[22].

Out of 50 cases of SCD patients, 12 patients with sickle cell crisis were studied. Male:Female ratio in total study population with crisis was 1:1. i.e. 6 cases (50%) were males and 6 cases (50%) were females. All 4 cases (100%) of AS pattern with crisis were females, while maximum number of cases, 6 cases (75%) were males out of 8 cases of SS pattern with crisis.

Of 12 patients of sickle cell crisis, maximum number of patients, 5 patients (41.67%) were in the age group of 21-30 years followed by 4 patients (33.33%) in the age group of 11-20 years.

All the 4 patients of AS pattern with crisis were females. Among them 2 (50%) were in the age group of 21-30 years. Among 8 patients of SS pattern with crisis, 6 (75%) were males and 2 (25%) were females. Of those 8 cases, maximum number of cases (3 cases - 37.5%) were males and they were in the age group of 11-20 years.

There were no patients with sickle cell crisis in the age group of above 40 years in the present study.

These findings of our study correlate more or less with the findings of Arun U Deore, et al. (2013)[20]; Andhale RB, et al. (2014)[13] ; Alkhunaizi AM, et al. (2014)[23] and Arun U Deore, et al. (2014)[21].

According to WHO criteria, patients were categorized into different classes of Anemia.

41 cases (82%) had anemia, among them 4 (8%) were severely anemic. In the present study the lowest Hb% was 5.5 gm% and the highest Hb% was 13.9 gm%; with the mean Hb concentration of 9.27±1.78 gm%.

Most of the cases, 31 cases (62%) of total 50 patients in our study had moderate anemia. Of those 31 cases 16 cases (53.33%) were of AS pattern, and 15 cases (75%) were of SS pattern.

12 males (57.14%) out of 21 males, 19 females (65.52%) out of 29 females had moderate anemia. 4 males (19.05%) out of 21 males, 2 females (6.90%) out of 29 females had mild anemia. There were total 4 (8%) cases who had severe anemia, among them 1 (4.76%) was male and 3 (10.34%) were females. Moderate to severe anemia was more in females than males, whereas males mostly had mild to moderate anemia.

Patients with SCD had chronic hemolytic anemia. Reason for more severity & prevalence of anemia in females include pregnancy, menstrual blood loss and nutritional factors. Patients with SS pattern & patients in crisis had more anemia than patients with AS pattern & patients without crisis repectively, due to amount of increased sickle hemoglobin.

These findings of our study correlate more or less with the findings of Mapp, et al. (1987)[10]; Serjeant GR (1992)[27]; Mohanty, et al. (2002)[28]; Montalembert (2008)[29]; Parmar Deepankar, et al. (2013)[30]; Michael Bursey, et al. (2013)[31]; Shafeer VP, et al. (2014)[22] and Lepira FB, et al. (2016)[32] .

To assess the tubular functions we had calculated fractional excretion of sodium (FE_{Na}), and to support the values of FE_{Na} we had also calculated fractional excretion of urea (Fe_{urea}).

Abnormal fractional excretion of sodium (FE_{Na}) was found in 36 (72%) out of 50 patients of SCD.

Of those 36 patients, 16 (80%) were of SS pattern and 20 (66.67%) were of AS pattern.

There was no evidence of significant serum electrolyte abnormality, however significant decrease in urinary potassium excretion was noted. This decreased potassium excretion in future might precipitate hyperkalemia and its consequences.

The above mentioned observations suggest that tubular dysfunctions occured in patients of SCD with both AS as well as SS pattern. It was more in patients with SS pattern than AS pattern.

43 cases (86%) of total 50 cases had abnormal FEurea. 6 cases (20%) of total 30 cases of AS pattern had normal FEurea. On the other hand only 1 case (5%) out of 20 cases of SS pattern had normal FEurea. Remaining patients i.e. 24 cases (80%) of AS pattern , and 19 cases (95%) of SS pattern had abnormal FEurea indicating tubular damage. In our study, tubular dysfunction was found in 80-95% of cases. Renal involvement was more in SS pattern than AS pattern.

Sickled RBC cause occlusion of the vasa recta in the renal medulla, causing disturbance in counter current exchange mechanism leading to urinary concentration & distal acidification defect and tubular dysfunction. Number of crises were more in patients with SS pattern than AS; that is why glomerular and tubular dysfunctions were more in SS pattern than AS pattern.

These findings of the present study correlate more or less with the findings of De Jong, et al. (1981)[11]; Falk RJ, et al. (1994)[12]; Scheinman, et al. (1994)[33]; Mohanty, et al. (2002)[28]; K. López, et al. (2011)[34]; Geraldo B. Silva Junior, et al. (2013)[35]; Varsha Wankhade, et al. (2013)[36] and Andhale RB, et al. (2014)[13].

7 (58.33%) out of 12 patients of sickle cell crisis had abnormal FE_{Na}.

Among the 4 patients with AS pattern with crisis, 2 cases (50%) had abnormal FE_{Na}. Similarly out of 8 patients with SS pattern with crisis, 5 (62.5%) had abnormal FE_{Na}.

In our study, it was observed that there was no significant difference in tubular dysfunction in patients with AS & SS pattern in crisis by using FE_{Na} as a marker of tubular dysfunction.

Out of 12 patients of sickle cell crisis 10 patients (83.33%) had abnormal Fe_{urea}.
2 cases (50%) of AS pattern in crisis and all the 8 (100%) cases of SS pattern in crisis had abnormal Fe_{urea}.

In our study, it was observed that tubular dysfunction occured in patients of SCD with both AS as well as SS pattern in crisis. It was more in patients with SS pattern in crisis than AS pattern in crisis. 100% cases of SS pattern with crisis had renal dysfunction in the form of tubular damage by using FE_{urea} as a marker of tubular dysfunction.

Sickled RBC cause occlusion of the vasa recta in the renal medulla, causing disturbance in counter current exchange mechanism leading to urinary concentration & distal acidification defect and tubular dysfunction. Number of crises were more in patients with SS pattern than AS; that is why glomerular and tubular dysfunctions were more in SS pattern than AS pattern.

12 cases (24%) of total 50 cases had significant microscopic albuminuria. Among them 6 cases (20%) were of AS pattern and remaining 6 cases (30%) were of SS pattern. There was no significant difference between the two groups. In our study, mean urinary pH of total cases was 4.92±0.68. Patients with AS pattern had mean urinary pH of 4.98±0.72, while the patients with SS pattern had mean urinary pH of 4.83±0.62. In our study, patients had not lost the acidification ability to far extent, that might be the possible reason that study cases had decreased number of patients with UTI and its complications.

Microscopic hematuria was a predominant finding in patients of SS pattern i.e. 5 cases (25%) of total 20 cases. Similarly cast and crystals were found in 5 cases (25%) of SS pattern. On the other hand, among the patients with AS patern 5 cases (16.67%) had hematuria and 7 cases (23.33%) had cast and crystals in their urine microscopy. Of total population, 5 patients (10%) had RBC casts, while 4 patients (8%) had granular casts. 3 cases (15%) of SS pattern had significant number of pus cells in their urine. 6 cases (30%) of SS pattern and 4 cases (13.33%) of AS pattern had epithelial cells in urine.

The above mentioned findings in SCD patients might be the result of glomerular and tubular damage with papillary necrosis. Also these findings could be secondary to interstitial nephritis.

Out of 12 patients with sickle cell crisis, 9 cases (75%) had significant albuminuria, among which 3 cases (75%) were of AS pattern with crisis and 6 cases (75%) were of SS pattern crisis.

3 cases (75%) of AS pattern with crisis had hematuria, while 4 cases (50%) of SS pattern with crisis had hematuria. Suggesting that hematuria occured in both sickle cell trait and disease. Suggesting that patients with sickle cell crisis had glomerular and tubular dysfunction along with papillary necrosis.

All the 4 cases (100%) of AS pattern with crisis had casts and crystals. There were 5 cases (62.5%) of SS pattern with crisis who had casts and crystals. Casts were mostly RBC cast, followed by granular casts. Suggesting that patients with sickle cell crisis had nephritis or glomerular or tubular damage.

There were 4 cases (75%) of SS pattern with crisis who had tubular epithelium or casts in their urine microscopy, suggesting tubular damage; while none of the patients of AS pattern had tubular epithelium in their urine microscopy.

Renal complications in sickle cell disease result from occlusion of the vasa recta in the renal medulla. The low partial pressure of oxygen and high osmolarity predispose to hemoglobin S polymerization and erythrocyte sickling. Sequelae include papillary necrosis, nephritis and glomerular and tubular damage leading to hematuria, proteinuria, hyposthenuria, acidification defect, and increased chances of UTI complicating to pyelonephritis.

These findings of present study correlate more or less with the findings of Strauss, et al. (1986)[37]; Mapp, et al. (1987)[10]; Nissenson, et al. (1989)[38]; Sklar, et al. (1990)[39]; Scheinman, et al. (1994)[33] ; Wong, et al. (1996)[40]; Van Eps, et al. (1997)[41]; Guasch, et al. (1997)[42]; Sesso, et al. (1998)[24]; Pablo, et al. (1999)[44]; Pham, et al. (2000)[45]; Powars, et al. (2005)[46]; Bray, et al. (2006)[47]; McKie, et al. (2007)[48]; Alvarez, et al. (2008)[49]; Abdu A, et al. (2011)[25]; K. López, et al. (2011)[34]; Pandey S, et al. (2012)[50] and Geraldo B. Silva Junior, et al. (2013)[35].

Of 50 SCD patients 12 patients (24%) had normal urinary specific gravity, while 38 patients (76%) had abnormal urinary specific gravity. Mean urinary specific gravity of total cases was 1.010±0.007. Patients with AS pattern had mean urinary specific gravity of 1.012±0.008, while the patients with SS pattern had mean urinary specific gravity of 1.008±0.006.

Hyposthenuria was a predominant finding in patients of sickle cell disease.

Among 36 patients of hyposthenuria, 19 cases (63.33%) were of AS pattern and 17 cases (85%) were of of SS pattern. These findings suggestive of renal medullary ischemia.

Only 2 cases (66.67%) had hyperosthenuria. Both of them had AS pattern.

None of the patients of SS pattern had hyperosthenuria, suggesting that renal involvement was more in patients of SS pattern than AS pattern. And so they were passing large amount of diluted urine, which further contribute to the development of dehydration, leading to further renal ischemia and complications.

In 12 patients with sickle cell crisis 9 (75%) had developed hyposthenuria. Out of which 3 cases (75%) were out of 4 cases of AS pattern and 6 cases (75%) were out of 9 cases of SS pattern.

None of the patients of sickle cell crisis had hyperosthenuria, suggesting that renal medulla in patients of crisis had developed ischemia, so they were passing large amount of diluted urine, which further contribute to the development of dehydration, leading to further renal ischemia and complications.

These findings of our study correlate more or less with the findings of De Jong, et al. (1981)[11]; Tejani, et al. (1985)[51]; Strauss, et al. (1986)[37]; Mapp, et al. (1987)[10]; Aoki, et al. (1990)[52]; Gupta, et al. (1991)[53]; Serjeant GR (1992)[27]; Falk RJ, et al. (1994)[12]; Guasch, et al. (1996)[43]; Sesso, et al. (1998)[24] ; Pablo, et al. (1999)44; K. López, et al. (2011)[34]; Kaur M, et al. (2013)[54]; Geraldo B. Silva Junior, et al. (2013)[35]; Varsha Wankhade, et al. (2013)[36] and Andhale RB, et al. (2014)[13].

Strength &Limitations

Strength :

In our study, SCD patients who have other systemic diseases are excluded. So renal involvement in such patients reflect its cause to be the sickle cell disease only.

To predict the glomerular & tubular dysfunction, combination of FE_{Na} & FE_{urea} are used. Combination of these indices have greater sensitivity & specificity than any single index of tubular dysfunction or AKI.

In our study, we include cases of SCA with & without crisis. No study was available in literature searched for renal involvement in sickle cell crisis.

Limitations:
Small sample size is a limitation of our study. Further studies can facilitate knowledge about the various types of renal involvement in sickle cell disease and their effect on morbidity, mortality and life expectancy. Awareness about the various risk factors will help in reduction of the morbidity and mortality associated with SCD.

Renal biopsy was not possible due to IEC objections.

Only the patients coming to the medicine department or speciality OPD are included. Thus the findings do not represent renal involvement in general population. So the results can not be generalized.

This was only a observational study and possible effect of treatment given to patients with proteinuria could not be studied.

Recommendations
Prevalence of sickle cell anemia is high in central India. Renal involvement is a known manifestation of SCD. To comment on renal status of sickle cell patients we have to assess different kidney function tests.

Microscopic examination of urine is the cheapest and the easiest technique to determine the physiology of the kidney, and urinary tract system, and its abnormalities. Physiologic status of the kidney can also be known by simple indices like FE_{Na}, FE_{urea} & specific gravity of urine. Correlation of these investigations with clinical, hematological, biochemical and urinary findings in patients of SCD can help in reducing the burden of UTI, sepsis, cystitis and the complications like ATN, glomerular and tubular dysfunctions, tubulointerstitial nephritis, pyelonephritis, renal failure and ESRD.

Rrenal profile and serum electrolytes should be done and should be regularly monitored before starting any diuretics to the SCD patients with systemic disease requiring diuretics (e.g. Congestive Cardiac Failure).

SCD patients should be taught about the regular medical check up and measures to avoid crisis.

These measures should be taken to prevent crisis, renal dysfunction and their complications leading to ESRD ultimately causing great morbidity and mortality.

Table 1 : Baseline characteristics of SCD patients

Baseline characters	Total (n=50)	AS (n=30)	SS (n=20)	p-value
Mean Age (S.D.)	26.44±9.45	26.40±8.94	26.50±10.42	0.03 p=0.97,NS
Mean Hb (S.D.)	9.27±1.78	9.47±1.83	8.98±1.70	0.96 p=0.34,NS
Mean Creatinine (S.D.)	0.91±0.21	0.95±0.21	0.84±0.19	1.91 p=0.06,NS
Mean RBS (S.D.)	102.58±18.88	101.57±18.84	104.1±19.32	0.46 p=0.64,NS
Mean GFR (S.D.)	81.34±22.49	79.07±22.50	84.76±22.61	0.87 p=0.38,NS
Mean Urinary pH (S.D.)	4.92±0.68	4.98±0.72	4.83±0.62	0.75 p=0.45,NS
Mean Urinary Specific Gravity (S.D.)	1.010±0.007	1.012±0.008	1.008±0.006	1.74 p=0.08,NS
Mean Urinary Na+ loss (S.D.)	73.06±38.42	63.13±21.76	87.95±51.92	2.33 p=0.024,S
Mean Urinary K+ loss (S.D.)	17.34±9.06	17.69±8.84	16.82±9.59	0.33 p=0.74,NS
Mean FE_{Na} (S.D.)	1.73±1.08	1.47±0.72	2.11±1.39	2.11 p=0.044,S
Mean Fe_{urea} (S.D.)	76.80±53.68	72.92±54.42	82.61±53.40	0.62 p=0.53,NS

Table 2 : Gender wise distribution of sickle cell patients

Sex	Total (n=50)	AS (n=30)	SS (n=20)	⨯2-value
Male	21 (42%)	9 (30%)	12 (60%)	18.18
Female	29 (58%)	21 (70%)	8 (40%)	p=0.0001,S
Total	50 (100%)	30 (100%)	20 (100%)	

Table 3: Age wise distribution of sickle cell patients

Age (years)	Total (n=50)	AS (n=30)	SS (n=20)	⨯2-value
11-20 years	13 (26%)	8 (26.66%)	5(25%)	2.96
21-30 years	22 (44%)	14(46.67%)	8(40%)	p=0.39,NS
31-40 years	12 (24%)	6(20%)	6(30%)	
≥41 years	3 (6%)	2(6.67%)	1(5%)	
Total	50 (100%)	30(100%)	20 (100%)	

Table 4:Age and gender wise distribution of sickle cell patients

Age (years)	Total (n=50)	Male (n=21)	Female (n=29)
11-20 years	13 (26%)	8 (38.10%)	5 (17.24%)
21-30 years	22 (44%)	7 (33.33%)	15 (51.72%)
31-40 years	12 (24%)	5 (23.81)	7 (24.14%)
≥41 years	3 (6%)	1 (4.76%)	2 (6.90%)
Total	50 (100%)	21 (100%)	29 (100%)
⨯2-value	12.25,p=0.0066,S		

Table 5: Gender wise distribution of patients with sickle cell crisis

Sex	Total patients in crisis (n=12)	AS in crisis (n=4)	SS in crisis (n=8)	⨯2-value
Male	6 (50%)	0(0%)	6 (75%)	22.47
Female	6 (50%)	4 (100%)	2 (25%)	p=0.0001,S
Total	12 (100%)	4 (100%)	8 (100%)	

Table 6: Age and gender wise distribution of patients with sickle cellcrisis cell crisis

Particulars	Total patients in crisis (n=12)		AS in crisis (n=4)		SS in crisis (n=8)	
AGE / SEX	M	F	M	F	M	F
11-20 years	3(25%)	1(8.33%)	0 (0%)	1 (25%)	3(37.5%)	0(0%)
21-30 years	2(16.67%)	3(25%)	0 (0%)	2 (50%)	2(25%)	1(12.5%)
31-40 years	1(8.33%)	2(16.67%)	0 (0%)	1 (25%)	1(12.5%)	1(12.5%)
≥41 years	0(0%)	0(0%)	0 (0%)	0 (0%)	0(0%)	0(0%)
Total	6(50%)	6(50%)	0 (0%)	4(100%)	6(75%)	2(25%)
⨯2-value	29.45,p-value=0.0001,S					

Table 7: Anemia in patients of SCD

Anemia	Total (n=50)	AS (n=30)	SS (n=20)	⨯2-value
Non Anemic	9 (18%)	6 (20%)	3 (15%)	12.71
Mild Anemia	6 (12%)	5 (16.67%)	1 (5%)	P=0.0053,S
Moderate Anemia	31 (62%)	16 (53.33%)	15 (75%)	
Severe Anemia	4 (8%)	3 (10%)	1 (5%)	

Table 8: Gender wise distributions of anemia in patients of SCD

Anemia	Total (n=50)	Male (n=21)	Female (n=29)	⨯2-value	Overall
Non Anemic	9 (18%)	4 (19.05%)	5 (17.24%)	0.13 P=0.71,NS	8.80, P= 0.031,S
Mild Anemia	6 (12%)	4 (19.05%)	2 (6.90%)	6.36 P=0.011,S	
Moderate Anemia	31 (62%)	12 (57.14%)	19 (65.52%)	1.71 P=0.19,NS	
Severe Anemia	4 (8%)	1 (4.76%)	3 (10.34%)	1.80 P=0.17,NS	

Table 9: FE_{Na} in patients of SCD

Particulars	Total (n=50)	AS (n=30)	SS (n=20)	⨯2-value
FE_{Na}< 1% (Normal)	14(28%)	10(33.33%)	4(20%)	4.33 P=0.037,S
FE_{Na}> 1% (Abnormal)	36(72%)	20(66.67%)	16(80%)	4.33 P=0.037,S

Table 10: FENa in patients of sickle cell crisis FE_{Na}

Particulars	Total patients in crisis (n=12)	AS in crisis (n=4)	SS in crisis (n=8)	⨯2-value
FE_{Na}< 1% (Normal)	5 (41.67%)	2 (50%)	3 (37.5%)	2.92 P=0.08,NS
FE_{Na} > 1% (Abnormal)	7 (58.33%)	2 (50%)	5 (62.5%)	3.43 P=0.06,NS

Table 11: FEurea in patients of SCD Fe_{urea}

Particulars	Total (n=50)	AS (n=30)	SS (n=20)	⨯2-value
Fe_{urea}< 35% (Normal)	7 (14%)	6(20%)	1 (5%)	10.29 P=0.0013,S
FE_{urea}> 35% (Abnormal)	43 (86%)	24(80%)	19(95%)	10.29 P=0.0013,S

Table 12: FEurea in patients of sickle cell crisis Fe_{urea}

Particulars	Total pstients in crisis (n=12)	AS in crisis (n=4)	SS in crisis (n=8)	ϰ2-value
Fe_{urea}< 35% (Normal)	2 (16.67%)	2 (50%)	0 (0%)	**66.67 P=0.0001,S**
FE_{urea}> 35% (Abnormal)	10 (83.33%)	2 (50%)	8 (100%)	**66.67 P=0.0001,S**

Table 13: Physical and microscopic urinary findings in patients of SCD

Particulars	Total (n=50)	AS (n=30)	SS (n=20)	ϰ2-value
Significant Albuminuria	12 (24%)	6 (20%)	6 (30%)	2.66 P=0.10,NS
Hematuria	10 (20%)	5 (16.67%)	5 (25%)	1.92 P=0.16,NS
Cast and crystals	12 (24%)	7 (23.33%)	5 (25%)	0.10 P=0.74,NS
Pus cells	5 (10%)	2 (6.67%)	3 (15%)	3.26 P=0.07,NS
Epithelial cells	10 (20%)	4 (13.33%)	6 (30%)	**8.56 P=0.003,S**

Table 14: Physical and microscopic urinary findings in patients of sickle cell crisis

Particulars	Total patients in crisis (n=12)	AS in crisis (n=4)	SS in crisis (n=8)	ϰ2-value
Significant Albuminuria	9 (75%)	3 (75%)	6 (75%)	0.00 P=1.00,NS
Hematuria	7 (58.33%)	3 (75%)	4 (50%)	**13.33 P=0.0003,S**
Cast and crystals	9 (75%)	4 (100%)	5 (62.5%)	**45.40 P=0.0001,S**
Pus cells	2 (16.67%)	2 (50%)	3 (37.5%)	2.92 P=0.08,NS
Epithelial cells	4 (33.33%)	0 (0%)	4 (50%)	**66.67 P=0.0001,S**

Table 15: Specific gravity of urine in patients of SCD

Particulars	Total (n=50)	AS (n=30)	SS (n=20)	ϰ2-value
Hyposthenuria (< 1.015)	36 (72%)	19 (63.33%)	17 (85%)	**12.58 P=0.0004,S**
Hyperosthenuria (> 1.026)	2 (4%)	2(66.67%)	0(0%)	**100.8 P=0.0001,S**

Table 16: Specific gravity of urine in patients of sickle cell crisis

Particulars	Total patients in crisis (n=12)	AS in crisis (n=4)	SS in crisis (n=8)	ϰ2-value
Hyposthenuria (< 1.015)	9 (75%)	3 (75%)	6 (75%)	-
Hyperosthenuria (> 1.026)	0 (0%)	0 (0%)	0 (0%)	-

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