Original Article

Urine Spot Protein Creatinine Ratio as a Predictor of Disease Severity and Adverse Outcome in Children with Dengue: A Cross-sectional Study

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ABSTRACT

Introduction: Dengue is a viral infection with different presentations, hence predicting the disease severity at admission is essential to triage patients needing meticulous monitoring. In severe dengue there is increase of urinary protein clearance due to the increase in systemic vascular permeability. Simple urine protein excretion screening test could guide the triage and monitor the patients with suspected dengue infection.

Aim: To evaluate the urine spot Protein Creatinine Ratio (PCR) as a tool in predicting the disease severity and adverse outcome in children with dengue.

Materials and Methods: The present cross-sectional study was conducted in Institute of Child Health, Egmore, Tamil Nadu, India, from October 2021 to December 2021. All children aged between 1 month to 12 years presenting with symptoms of fever, thrombocytopenia (less than 1,50,000/μL) with or without dengue non structural protein component (NS1)/Immunoglobulin M (IgM) Enzyme Linked Immunosorbent Assay (ELISA) positivity were recruited for the study. Study parameters included were

demographic factors, severity of dengue classification based on National Vector Borne Disease Control Programme (NVBDCP) guidelines, urine spot PCR. The Urine Protein Creatinine Ratio (UPCR) was compared between dengue and non dengue cases and within the different categories of dengue cases. Chi-square was used for comparing proportions.

Results: Among 150 children enrolled in present study, 134 (89.3%) were dengue positive, 16 (10.7%) were non dengue. Most predominant age group involved was >6-9 years followed by >3-6 years. Among 134 children with dengue, 98 (73.1%) had high UPCR and among 16 non dengue children, 2 (12.5%) had high UPCR which was statistically significant (p-value <0.0001). Mean spot UPCR was 0.32±0.12 in mild dengue, 0.77±0.40 in moderate dengue and 1.68±1.67 in severe dengue which was statistically significant (p-value=0.0017). Children with severe dengue had higher PCR values in comparison to mild and moderate dengue.

Conclusion: There was a statistically significant association of urine spot PCR with severity of dengue and this simple test can be used for triaging and monitoring children with suspected dengue.

INTRODUCTION

Dengue is an arboviral infection affecting especially humans and represents a major global public health issue. Its incidence is increasing steadily and, in many places, like in developing countries it has become an endemic problem [1]. Dengue mainly affects the paediatric age group and is associated with considerable morbidity and mortality [2]. Mortality in dengue is due to abnormal capillary permeability, abnormalities of haemostasis and in severe cases dengue shock syndrome. The annual incidence rate of dengue is 49.5 per 1000 child years among children with fever >3 days [3]. The risk factors for development of severe disease are poorly characterised and consequently uncomplicated cases are frequently hospitalised for observation during the critical phase for capillary leakage syndrome, thereby making the situation cumbersome to both patients and treating physician. Therefore, improvements in early diagnosis and risk prediction for severe disease are urgently needed. This would enable appropriate and early intervention. Ideally, the test should be cheap, fast, easy to perform, highly sensitive and specific [2].

The presence of microalbuminuria has been postulated as potential risk predictor for severe dengue [4,5], but there is little information on the magnitude, timing of onset, or evolution of urinary protein excretion during infection. Also 24-hour urinary albumin measurements are time consuming to perform. Both measurement of spot urine protein estimation as well as urine protein to creatinine

ratio is a less cumbersome and are more practical method. Measurement of spot urine protein to creatinine ratio is much easier approach and hence acceptable method [6].

The spot PCR is obtained by the ratio between urine protein excretion (measured by 24-hour protein excretion or spot urine sample) and creatinine excretion, expressed as mg/mmol or mg/mmol. Spot PCR represents a practical alternative to the 24-hour urine collection because it is easier to obtain and is not influenced by variations in water intake or diuresis [6]. While it has been studied in adult dengue cases [1], its usefulness as a predictor tool has not been well tested in paediatric population. Hence, present study emphasise at simple laboratory investigation of UPCR as a predictor of disease severity of dengue infection in children who are very crucial for monitoring and management of children at risk especially during dengue epidemics. Hence, this study was planned to evaluate the UPCR as a tool in predicting the disease severity and adverse outcome in children with dengue.

MATERIALS AND METHODS

The present cross-sectional study was conducted in a tertiary hospital, Institute of Child Health, Egmore, Madras Medical College, Tamil Nadu, India, from October 2021 to December 2021. Study was proceeded after obtaining consent from Institutional Ethics Committee (IEC) (No17112021) and written informed consent from parents.

Inclusion criteria: All children aged between 1 month to 12 years, presenting with symptoms of fever, thrombocytopenia (<1,50,000/μL) with or without dengue non structural protein component (NS1/IgM) positivity were recruited for the study.

Exclusion criteria: Children who developed fever >48 hour after admission or following surgery were excluded from the study.

Initially 134 children turned to be dengue positive based on dengue NS1/IgM ELISA positivity and 16 were dengue negative. Positivity or negativity was based only on dengue NS1/IgM reports.

Sample size: A total of 150 neonates, who presented in the department with fever within the study period from October 2021 to December 2021 were enrolled in the study by purposive sampling.

Study parameters included in the study were demographic factors including age and sex, laboratory parameters such as urine protein and creatinine were evaluated. Urine spot protein was detected and quantified by pyrogallol red method. Creatinine by modified Jaffe's method. Urine spot protein creatinine ratio is calculated by dividing the level of protein (mg/dL) in a spot urine by the creatinine level (mg/dL) [7].

Procedure

Urine Protein Creatinine Ratio (UPCR) was done after admission. In children, normal range are [8]:

- Age <2 years, UPCR value <0.5
- Age ≥2 years, UPCR value <0.2

Small amount of protein in the urine is considered acceptable, proteinuria is defined as protein excretion greater than 100 mg/m^2 per day or more than 0.2 mg protein/mg creatinine (also known as a urine protein/creatinine ratio ([U p/c] >0.2) on a single spot urine collection; in neonates and infants, higher amount of protein excretion, upto 300 mg/m^2 is permissible [9]. Urine creatinine (24-hour urine collection) values can range from 500 to 2000 mg/day (4,420 to 17,680 mmol/day) [10,11]. The UPCR was compared between dengue and non dengue cases based on dengue NS1/IgM positivity. After dengue NS1/IgM positivity further UPCR was assessed in mild, moderate and severe categories of dengue cases. Severity of dengue classification was based on NVBDCP guidelines [12] and World Health Organisation (WHO) 1997 classification scheme, which comprises three categories.

- Dengue Fever (DF)- Mild dengue
- Dengue Haemorrhagic Fever (DHF)- Moderate dengue,
- Dengue Shock Syndrome (DSS)- Severe dengue

based on clinical signs, it was used to classify the degree of severity of the disease [13].

STATISTICAL ANALYSIS

Analyses were performed using the Statistical Package for the Social Sciences (SPSS) version 20.0 (SPSS Inc, Chicago, IL). Descriptive data was presented as mean±SD or percentages. Chi-square was used for comparing proportions. The p-value of <0.05 was considered statistically significant.

RESULTS

Among 150 children admitted with fever and thrombocytopenia 134 (89.3%) were dengue serology positive, 16 (10.7%) were cases of fever with thrombocytopenia who were dengue negative. Maximum patients 52 (38.8%) in dengue positive group were aged between >6-9 years followed by the 47 (35.1%) children in age group >3-6 years and 21 (15.7%) in age group 1-3 years. In dengue negative group maximum children 6 (37.5%) aged between >6-9 years followed by 5 (31.2%) in >3-6 years. In dengue positive group there were 73 (54.5%) males and 61 (45.5%) females while in dengue negative group 11 (68.7%) were males and 5 (31.3%) were females [Table/Fig-1].

Categories	Variables	Dengue positive (n=134)	Dengue negative (n=16)	p-value
Sex	Male	73 (54.5%)	11 (68.7%)	0.276
	Female	61 (45.5%)	5 (31.3%)	
Age group (years)	<1	10 (7.5%)	1 (6.3%)	0.489
	1-3	21 (15.7%)	2 (12.5%)	
	>3-6	47 (35.1%)	5 (31.2%)	
	>6-9	52 (38.8%)	6 (37.5%)	
	>9-12	4 (2.9%)	2 (12.5%)	
Urine protein	Mean±SD	34.1±1.6	10±0.7	0.0389
Creatinine	Mean±SD	72.3±1.3	30±0.8	0.0001
Urine spot PCR	Mean	0.47	0.33	0.3799

[Table/Fig-1]: Demographic characters.
p-value <0.05 considered significant

Among 134 children with dengue, 98 (73.1%) had high urine PCR and among the 16 non dengue children, 2 (12.5%) had high UPCR which was statistically significant (p-value <0.0001) [Table/Fig-2].

Dengue	Urine spot PCR		Chi-square test	p-value
	Normal Age (<2 years) <0.5 Age (≥2 years) <0.2	High Age (<2 years) >0.5 (≥2 years) >0.2		
Yes (134)	36 (26.9%)	98 (73.1%)	3.6474	<0.0001
No (16)	14 (87.5%)	2 (12.5%)		

[Table/Fig-2]: Urine spot protein creatinine ratio in dengue and non dengue children.
p-value <0.05 considered significant

Among 134 dengue children, 93 children with mild dengue had mean spot PCR 0.32±0.12, 33 children with moderate dengue had mean spot PCR 0.77±0.40, 8 children with severe dengue had mean urine spot PCR 1.68±1.67 which showed statistical significance (p-value=0.0017) [Table/Fig-3].

Study group	n (%)	Mean UPCR	Chi-square test	p-value
Dengue fever	93 (69.4)	0.32±0.12	12.746	0.0017
Dengue haemorraghic fever	33 (24.6)	0.77±0.40		
Dengue shock syndrome	8 (6.0)	1.68±1.67		

[Table/Fig-3]: Showing association of UPCR values in dengue children.
p-value <0.05 considered significant

Total 16 cases were in age group <2 years and 118 cases in ≥2 years and urine spot PCR was high in age group ≥2 years with 93 (78.8%) and thus children ≥2 years were more likely to have high UPCR [Table/Fig-4].

Age group	Urine spot protein creatinine ratio		Chi-square tests	p-value
	Normal	High		
<2 years (16)	11 (68.7%)	5 (31.3%)	17.7785	0.000138
≥2 years (118)	25 (21.2%)	93 (78.8%)		

[Table/Fig-4]: Comparison of urine spot PCR in dengue positive children among different age group.
p-value <0.05 considered significant

Out of the 134 dengue positive children male: female ratio was 1.2:1. Among 73 male, 50 (42.4%) children and among 61 female, 43(36.4%) children had high urine PCR. There was no statistical association between gender and UPCR [Table/Fig-5]. Out of 134 dengue children studied only one child died.

Age group (Total)	Sex	Urine spot PCR		Chi-square test	p-value
		Normal N (%)	High N (%)		
<2 (16)	F	4 (25)	4 (25)	2.618	0.106
	M	7 (43.8)	1 (6.2)		
≥2 (118)	F	10 (8.5)	43 (36.4)	0.7622	0.360
	M	15 (12.7)	50 (42.4)		

[Table/Fig-5]: Showing comparison of proteinuria between gender groups.

DISCUSSION

Dengue fever is now a widely distributed viral illness causing varied range of presentation and carries its own mortality rate. Mortality in dengue is due to abnormal capillary permeability, abnormalities of haemostasis and in severe cases, dengue shock syndrome. In the paediatric age group with severe dengue, outcome depends on the high suspicion level and scrutiny in monitoring the children for complications. This study was undertaken to study the significance of UPCR as an early predictor of severity of illness in children with dengue fever. This study revealed a statistically significant increased incidence of proteinuria among children with dengue in comparison to children with non dengue thrombocytopenia. These findings were similar to the observation of Graham RR et al., [14]. The study revealed that among the children with dengue positivity, the degree of proteinuria was high in severe dengue children than when compared with mild and moderate dengue cases which was statistically significant (p-value=0.0017) which concludes that there exists a positive association between the degree of proteinuria and the disease severity. Similar findings were consistent with study done by Vasanwala FF et al., patients with DHF had significantly higher median peak proteinuria levels (0.56 versus 0.08 g/day; p-value <0.001) compared to dengue fever [15].

The study revealed a high UPCR value in children ≥2 years when compared with infants which was statistically significant (p-value <0.0001) which reveals that children ≥2 years may manifest severe dengue illness whereas in study done by Datla P et al., age did not show any association with UPCR values [1]. The comparison of degree of proteinuria among male and female gender did not show any statistical significance which concludes that severity of dengue does not have a gender predisposition according to the study which was coincident with the study done by Datla P et al., [1]. Higher incidence in males were noted in the study done by Wali JP et al., [16].

Since out of 134 dengue children studied only one child died, the association of degree of proteinuria with adverse outcome like mortality cannot be commented on unlike study done by Datla P et al., where the association of raised UPCR with mortality showed a positive correlation and was statistically significant [1]. This study establishes a simple laboratory investigation of urine spot PCR as a predictor of disease severity of dengue infection in children which is very crucial for monitoring and management of children at risk especially during dengue epidemics.

Limitation(s)

The limitation of present study was that it did not take into account the day of illness when the proteinuria was assessed and also the day of peak proteinuria in dengue illness. Since out of 134 dengue children studied only one child died, the association of degree of urine spot PCR with adverse outcome like mortality cannot be commented.

CONCLUSION(S)

Given the increase in occurrence of dengue fever in children and its associated complications, the need for early predictors of disease severity is important for easy and apt monitoring of children at risk. Urine spot protein creatinine ratio assessment is easy to perform and inexpensive test. There was a statistically significant association of urine spot PCR with severity of dengue and this simple test can be used for triaging and monitoring children with suspected dengue. This study found higher urine spot protein creatinine ratio to be a non invasive marker in predicting the severity of dengue in children with dengue fever. Urine spot protein creatinine ratio in children with dengue fever can be used as a screening test in predicting the severity of disease at admission.

REFERENCES

[1] Datla P, Raju U, Reddy P, Srikrishna S, Deshmukh T. Study establishing correlation of proteinuria and urine protein/creatinine ratio with disease severity in paediatric dengue fever. Int J Med Paedia Oncol. 2017;3(1):24-28.

[2] Dengue: Guidelines for diagnosis, treatment, prevention and control. World Health Organization. Special Programme for Research and Training in Tropical Diseases, World Health Organization. Dengue: guidelines for diagnosis, treatment, prevention and control. New ed. Geneva: TDR: World Health Organization; 2009. Accessed date: September 17, 2021.

[3] Rose W, Sindhu KN, Abraham AM, Kang G, John J. Incidence of dengue illness among children in an urban setting in South India: A population-based study. Int J Infect Dis. 2019;84S:S15-S18.

[4] Chen Y, Maguire T, Hileman RE, Fromm JR, Esko JD, Linhardt RJ, et al. Dengue virus infectivity depends on envelope protein binding to target cell heparan sulfate. Nat Med. 1997;3(8):866-71.

[5] Lumpaopong A, Kaewplang P, Watanaveeradej V, Thirakhupt P, Chamnanvanakij S, Srisuwan K. Electrolyte disturbances and abnormal urine analysis in children with dengue infection. Southeast Asian J Trop Med Public Health. 2010;41:7276.

[6] Bakker AJ. Detection of microalbuminuria. Receiver operating characteristic curve analysis favours albumin-to creatinine ratio over albumin concentration. Diabetes Care. 1999;22:307-13.

[7] Kaminska J, Dymicka-Piekarska I, Tomaszewska J, Matowicka-Karna J, Koper-Lenkiewicz OM. Diagnostic utility of protein to creatinine ratio (P/C ratio) in spot urine sample with in routine clinical practice. Crit Rev Clin Lab Sci. 2020;57(5):345-64.

[8] Woroniecki R. Proteinuria: Signs and symptoms in paediatrics. American Academy of Paediatrics. 2015;709-17.

[9] Viteri B, Reid-Adam JH. Hematuria and proteinuria in children. Pediatr Rev. 2018;39(12):573-87.

[10] Landry DW, Bazari H. Approach to the patient with renal disease. In: Goldman L, Schafer AI, eds. Goldman-Cecil Medicine. 26th ed. Philadelphia, PA: Elsevier; 2020: chap 106.

[11] Oh MS, Briefel G. Evaluation of renal function, water, electrolytes, and acid-base balance. In: McPherson RA, Pincus MR, eds. Henry's Clinical Diagnosis and Management by Laboratory Methods. 23rd ed. St Louis, MO: Elsevier; 2017: chap 14.

[12] National guidelines for clinical management of dengue fever 2015.page 11. Accessed date: September 17, 2021.

[13] World Health Organization. Dengue Haemorrhagic Fever: Diagnosis, Treatment, Prevention and Control, 2nd edn. Geneva, Switzerland: WHO, 1997.

[14] Graham RR, Juffrie M, Tan RA. prospective sero-epidemiologic study on dengue in children four to nine years of age in Yogyakarta, Indonesia 1995-1996. Am J Trop Hyg. 1999;61(3):412-19.

[15] Vasanwala FF, Puvanendran R, Ng JM, Suhail SM. Two cases of self-limiting nephropathies secondary to dengue haemorrhagic fever Singapore Med J. 2009;50(7):e253.

[16] Wali JP, Biswas A, Handa R, Aggarwal P, Wig N. Dengue hemorrhagic fever in adults: A prospective study of 110 cases. Top Doct. 1999,29:27-30.

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