Steroid Resistant Nephrotic Syndrome: Role of Histopathology

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This study was conducted to (1) see the histopathological distribution of different subtypes in steroid resistant nephrotic syndrome (SRNS) and (2) compare the clinical, biochemical parameters and outcome between Minimal Change Disease (MCD) with non-MCD subtypes in response to immunosuppressive therapy. A retrospective analysis was done of data on all biopsy proven children with idiopathic SRNS (no response to 4 weeks of standard prednisone therapy (60 mg/m2/day)) referred to our institute over last 12 years. They were treated with one of the following medications: oral or intravenous cyclophosphamide, cyclosporine or combination of dexamethasone and azathioprine. A comparison was done of the demographic clinical and biochemical features different histopathologies. We studied 136 children with SRNS (100M, 36F). They accounted for 15.1%(136/900) of all children with idiopathic nephrotic syndrome. Focal segmental glomerulosclerosis (FSGS) was the commonest 80/136 (59%), followed by MCD (17.6%). Children with non-MCD had a significantly greater prevalence of microhematuria as compared to MCD. The other baseline clinical and biochemical features including the glomerular filtration rate (GFR) were similar. After a mean follow up of 46 (8-148) months, a significantly greater children with non-MCD (65/112) continued to be proteinuric as compared to the MCD (3/24) (p=0.0001). FSGS was the commonest cause of SRNS in our patient population. Children with SRNS secondary to MCD are more likely to achieve remission as compared to non- MCD subtypes and have a better long-term prognosis. Hence kidney biopsy is of significant prognostic value in SRNS.

Key words: *Focal segmental glomerulosclerosis, Minimal change disease, Nephrotic syndrome, Steroid resistance.*

THE management of steroid resistant nephrotic syndrome (SRNS) continues to pose a therapeutic challenge to nephrologists. Although there is a consensus that all these children should undergo a kidney biopsy, there have been very few large studies that have evaluated the role of histopathology(1,2). Most of the other studies have included small number of children and have evaluated different therapeutic protocols for focal segmental glomerulosclerosis (FSGS). A recent meta-analysis failed to demonstrate any difference in the efficacy of immunosuppressive agents in inducing remission in SRNS children with minimal change disease (MCD) versus FSGS(3).

The International Study of Kidney Disease in Children (ISKDC) study demonstrated that clinical corticosteroid response had a high predictive accuracy for outcome than renal histological findings in children with idiopathic nephrotic syndrome(4). Hence, the

practice of performing kidney biopsy at presentation in all children was abandoned and over the years the role of kidney biopsy has become more and more restricted(5). The contribution of histology in the management of SRNS needs to be evaluated. Hence this study was conducted to compare: (1) the histopathological distribution of different subtypes in SRNS and (2) the clinical and biochemical parameters at the time of diagnosis and the outcome in MCD compared to non-MCD.

Subjects and Methods

A retrospective analysis was done of all children (with age of onset between 1 to 16 years) with SRNS referred to our institute over the last 12 years. These patients comprised a homogenous racial group representing the Northern and Eastern Indian populations. Inclusion criteria were: (1) steroid resistance either primary or secondary, (2) renal biopsy performed soon after a diagnosis of steroid resistance. We excluded children with (*i*) underlying secondary causes, (*ii*) hepatitis B surface antigen (HBsAg) seropositivity, human immunodeficiency virus (HIV) seropositivity or anti HCV seropositivity, (*iii*) biopsy failures.

At presentation, they were evaluated clinically for hypertension, hematuria, anthropometric parameters (height, weight and body surface area) and systemic involvement. They were investigated for confirmation of nephrotic syndrome, renal function status and evaluation of secondary causes of nephrotic syndrome. All cases fulfilled the International Study of Kidney Disease in Children criteria for the diagnosis of nephrotic syndrome(1).

A diagnosis of SRNS was made if they did not respond to a 4-week course of prednisone at a dose of 60 mg/m^2/day administered under our supervision, in the absence of evidence of underlying infection(3). After informed consent, kidney biopsy was performed in all patients. The biopsy specimens were reviewed and interpreted by the same pathologists and the histopathological diagnosis was made as per standard case definitions defined in our earlier study(6). Adequacy of biopsy was defined by the presence of at least 5 glomeruli in the specimen on light microscopy(7). For the purpose of this study, we analyzed only those children with SRNS who underwent kidney biopsy.

Children with MCD, FSGS and Mesangial Proliferative Glomerulonephritis (MesPGN) subtypes were treated with one or more of the following regimens: (1) Cyclophosphamide given orally at a dose of 2.5 mg/kg/day for 3 months or in monthly pulses of 500 mg/m^2 administered intravenously for 6 months after informed consent(8), (2) Cyclosporine at a dose of 5-8 mg/kg/day, (3) Intravenous dexamethasone at a dose of 5 mg/kg alternate day for 2 weeks, weekly for 2 months and thereafter oral prednisone 2 mg/kg on alternate day and oral azathioprine at a dose of 2.5 mg/kg/day. Children with membrano-proliferative GN (MPGN) were administered steroids as per the ISKDC protocol(9) while those with membranous nephropathy (MGN) were treated with Ponticelli's regimen(10). If the protein-uria persisted beyond 6 months after stoppage of immunosuppressive agents, an ACEI was added. These children were followed up monthly initially and thereafter every 3 months. On each visit, the child was evaluated clinically for evidence of disease activity and complications (infections and drug side-effects).

A diagnosis of primary steroid resistance was made if the proteinuria persisted after 4 weeks of daily prednisone therapy in a dose of 60 mg/m^2/day in the absence of any evidence of underlying infection(3). Secondary steroid

resistance was defined as no response to 4 weeks of daily prednisone therapy at a dose of 60 mg/m^2/day in a child previously known to have a steroid sensitive course. Hypertension was defined as systolic or diastolic blood pressure >95th percentile for age, gender and height on 3 separate occasions(11). Microscopic hematuria was defined as the presence of more than 3 RBC per high power field in an unspun fresh urine specimen(6). The proteinuria was evaluated in terms of: (*i*) complete remission *i.e.*, a reduction in urinary protein excretion in spot sample (urine protein/creatinine ratio) <0.2 for at least 3 consecutive days, (*ii*) partial remission *i.e.*, a reduction in urinary protein excretion in spot sample (urine protein/creatinine ratio) between 0.2 and 2.0 for at least 3 consecutive days and (*iii*) persistent proteinuria *i.e.*, persistent spot protein/ creatinine >2.0. GFR was estimated by the Schwartz formula(12,13).

Statistical analysis

All values are given as mean ± standard deviation. The statistical analysis was performed by the SPSS 9.0 statistical analysis software (Chicago, Ill, USA). Outcome was evaluated in terms of partial/complete remission versus no remission. The results were analyzed for their statistical significance using Student's test for continuous variables and chi-square test for discrete variables; $P < 0.05$ was considered significant.

Results

The study group comprised of 136 children with SRNS. They comprised of 15.1% (136/900) children referred to our institute over the last 12 years. There were 100 males, 36 females. Mean age of onset of nephrotic syndrome was 8.72 yr (1-18 yr). Facial edema was found in all the 136 children, microhematuria in 67 (41.3%) and hypertension in 36 children.

In patients with SRNS, FSGS was the commonest histopathologic subtype occurring in 80 out of 136 patients (58.8%). Other lesions included MCD in 24 patients (17.6%), MesPGN in 24 (17.6%), MGN in 6 (4.4%) and MPGN in two. After a mean follow-up of 46 months (8-148 months), 69 continued to have proteinuria while 67 children attained remission.

The demographic and clinical features of different histopathological subtypes are depicted in *Table I.* We observed that FSGS was the commonest cause of SRNS in children with onset of nephrotic syndrome at all age groups. Of the various clinical and biochemical features, only microhematuria was significantly more common in non- MCD subgroups (*Table II*). The distribution of

TABLE I–*Demographic and Clinical Features in Different Histopathological Groups*

	MCD (n=24)	FSGS (n=80)	MesPGN (n=24)	MPGN (n= 2)	MGN (n= 6)	Total (n=136)
Age of onset (yr) (mean ± SD)	6.8±5.5	9.1±5.40	10.8±3.9	10±2.8	11.3±3.5	8.7±5.1
Gender (M/F)	19/5	56/24	18/6	2/0	5/1	100/36
Microhematuria	4	40	19	2	2	67
Hypertension	3	20	9	1	0	33

MCD: minimal change disease; FSGS: focal segmental glomerulosclerosis; MesPGN: mesangioproliferative GN; MPGN: membranoproliferative GN; MGN: membranous nephropathy.

children with primary steroid resistance (13/24 in MCD *vs* 80/112 in non-MCD) was not significantly different in the two groups (P = 0.16). On comparing the outcome in response to immunosuppressive therapy, we found that children who had MCD were significantly more likely to attain remission as compared to non-MCD (P = <0.00001).

Discussion

There is a paucity of recent data about the histopathological spectrum of SRNS. Although there have been previous studies on this aspect(1,2), there is recent evidence to suggest that there is a changing trend in histopathology of idiopathic nephrotic syndrome(14). The underlying histopathologic characteristics in nephrotic syndrome are of significance in determining the outcome(4). MCD, MesPGN and FSGS together account for the majority of children with idiopathic nephrotic syndrome(14,15) and are also treated with a common steroid protocol(16).

In the landmark study by ISKDC, MCD, FSGS and MPGN each accounted for about a quarter of children with SRNS(1). In contrast in the current study, FSGS was the commonest lesion accounting for 59% of all patients followed by MesPGN which accounted for 18%. We have also previously reported an increasing incidence of FSGS in our patient population, although(14) MPGN accounted for only 2 of the 136 children. This figure is much lower than the ISKDC study(1) and may thus be an underestimate. This is because only these 2 children had a nephrotic presentation and had normal serum complement levels. Hence both of them ended up with 4 weeks of prednisone therapy before they were labeled as SRNS and underwent a biopsy. Children who had a nephritic-nephrotic presentation and low serum complement levels were biopsied at the onset. Since these children did not receive the conventional 4-week course of daily prednisone, and hence were not labeled as SRNS and not included in this analysis. All these patients were HBsAg and HCV negative.

TABLE II–*Comparison of Demographic, Clinical and Biochemical Features Between MCD and non-MCD Subtypes*

	MCD (n = 24)	Non-MCD (n = 112)	P
Age of onset (yr)	6.8 + 5.4	9.0 + 5.0	0.06
Hypertension	3	30	0.22
Microhematuria	4	63	0.001
Infections	2	15	0. 89
INR:SNR	13:11	80:32	0.16
Urea nitrogen (mg/dL)	13.15 ± 4.7	15.6 ±12.5	0.39
Creatinine (mg/dL)	0.83 ± 0.43	0.92 ± 0.52	0.43
Protein (g/dL)	5.05 ± 0.88	5.2 ± 0.94	0.49
Albumin (g/dL)	2.2 ± 0.77	2.4 ± 0.60	0.15
Outcome (Persistent proteinuria)	3	65	0.0001
Duration follow up (months)	52.1± 29.1	44.8 ± 27.4	0.25

Data expressed as mean ±SD; INR=Initial non responder (primary steroid resistant).
SNR=Subsequent non-responder (secondary steroid resistant).

In our earlier study of the distribution of histopathology in all patients with INS, we had observed that in children with onset under 8 years age, MCD was the most commonly encountered entity(6). In contrast in the current study with SRNS, FSGS was the commonest lesion in children under 8 years as well as beyond 8 years. On comparing the various clinical features in MCD *vs* non-MCD groups, we observed that only micro-hematuria was significantly more common in non-MCD group. This is in contrast to our observations in our earlier study in children with INS, where we observed that children in MCD group had a significantly younger age of onset and a lower incidence of hypertension and microhematuria(6). In contrast to the study by White, *et al.* we did not observe any difference in the histopathological distribution in boys or girls.

Following immunosuppressive therapy, we found that MCD had significantly greater remission rates compared to those with non-MCD. This is perhaps the largest study to demonstrate this in SRNS patients(1,2). The distribution of children with primary and secondary steroid resistance was similar in the two groups and hence was unlikely to influence the outcome in terms of remission. Thus kidney biopsy has a significant prognostic value in children with SRNS, in contrast to steroid sensitive nephrotic syndrome.

In conclusion, FSGS remains the most common histopathological subtype in children with SRNS. It is difficult to clinically differentiate MCD from non-MCD. Children with MCD had a significantly greater incidence of remission with immuno-suppression as compared to non-MCD subgroup. The prognosis of children with SRNS with MCD is much better than non-MCD. Kidney biopsy is of prognostic value in these children.

Contributors: SG was involved in the concept, study design and preparation of the manuscript. DS was involved in data acquisition and analysis. RKS, AS and AG assisted with drafting of the manuscript and critical review. RKG contributed to the review and analysis of the histological data. US assisted with the statistical analysis.

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REFERENCES

1. International Study of Kidney Disease in Children. The primary nephrotic syndrome in children: Identification of patients with minimal change nephrotic syndrome from initial response to prednisone. J Pediatr 1981; 98: 561-564.
2. White RH, Glasgow EF, Mills RJ. Clinico-pathological study of nephrotic syndrome in childhood. Lancet 1970; 1: 1353-1359.
3. Habashy D, Hodson EM, Craig JC. Interventions for steroid-resistant nephrotic syndrome: A systematic review. Pediatr Nephrol 2003; 18: 906-912.
4. International Study of Kidney Disease in Children. Nephrotic syndrome in children: Prediction of histopathology from clinical and laboratory characteristics at time of diagnosis. Kidney Int 1978; 13: 159-165.
5. Gulati S, Sharma AP, Sharma RK, Gupta A, Gupta RK. Do current recommendations of kidney biopsy in nephrotic syndrome need modifications? Pediatr Nephrol 2002; 7: 404-408.
6. Kumar J, Gulati S, Sharma AP, Sharma RK, Gupta RK. Histopathological spectrum of childhood nephrotic syndrome in Indian children. Pediatr Nephrol 2003; 18: 657-660.
7. Oberholzer M, Torhorst J, Perret E. Mihatsch MJ. Minimum sample size of kidney biopsies for semi-quantitative and quantitative evaluation. Nephron 1983; 34: 192-195.
8. Elhence R, Gulati S, Kher V, Gupta A, Sharma RK. IV pulse cyclophosphamide-A new regime for steroid-resistant minimal change nephrotic syndrome. Pediatr Nephrol 1994; 8: 1-3.

9. Tarshish P, Bernstein J, Tobin JN, Edelmann CM. Treatment of mesangiocapillary glomerulo-nephritis with alternate-day prednisone: A report of the International Study of Kidney Disease in Children. Pediatr Nephrol 1992; 6: 123-130.

10. Ponticelli C, Passerini P. Treatment of membranous nephropathy. Nephrol Dial Transplant 2001; 16: 8-10.

11. National High Blood Pressure Education Program Working Group on Hypertension Control in Children and Adolescents. Update on the 1987 task force report on high blood pressure in children and adolescents: a working group report from the National High Blood Pressure Education Program. Pediatrics 1996; 98: 649-658.

12. Schwartz G J, Haycock GB, Spitzer A . Plasma creatinine and urea in children: Normal values for age and sex. J Pediatr 1976; 88: 828-830.

13. Schwartz GJ, Brion LP, Spitzer A . The use of plasma creatinine concentration in for estimating glomerular filtration rate in infants, children and adolescents. Pediatr Clin North Am 1987; 34: 571-590.

14. Gulati S, Sharma AP, Sharma RK, Gupta A. Changing trends of histopathology in childhood nephrotic syndrome. Am J Kid Dis 1999; 34: 646-650.

15. Broyer M, Meyrier A, Niaudet P, Habib R. Minimal change and focal segmental glomerulosclerosis, *In:* Davison AM, Cameron JS, Grunfeld JP, Kerr DNS, Ritz E, Winerals CS (eds). Oxford Textbook of Clinical Nephrology. Oxford, Oxford University Press, 1998; pp 493-535.

16. Gulati S, Kher V, Sharma RK, Gupta A. Steroid response pattern in Indian children with nephrotic syndrome. Acta Pediatr Scand 1994; 83: 530-533.