Rituximab/Mycophenolate Combination Therapy in Children with Calcineurin Inhibitor-Resistant FSGS

Abstract

Introduction: There is a paucity of data and therapeutic options nationally and internationally on calcineurin inhibitor (CNI)-resistant forms of focal segmental glomerulosclerosis (FSGS) in children. CNI (tacrolimus or cyclosporine) are proven monotherapy in children with FSGS with a steroid-dependent (SD) or steroid-resistant (SR) course. We analyzed a novel therapeutic option in CNI-resistant FSGS by using the dual therapy of rituximab and mycophenolate to maintain remission. **Methods:** This is a retrospective analysis of clinical, therapeutic profile, and treatment outcomes (sustained remission versus no remission) in subjects with CNI-resistant FSGS who received dual rituximab therapy along with mycophenolate as maintenance therapy for a minimum of 1 year. **Results:** The median age of presentation of 13 recruited children was 7.8 years (range: 2.4–17.6 years); nine (69.2%) were males. Ten (76.9%) of them had an SD course and three (23.1%) had an SR course. Four (30.7%) had evidence of acute/chronic CNI toxicity, and the remaining nine (69.3%) showed no response to CNI therapy despite adequate trough levels. Post dual therapy, 11 (84.6%) had sustained remission for at 1 year and two (15.4%) children did not show remission. None reported adverse reactions or infections, and all had preserved renal functions. **Conclusion:** Dual combination therapy with rituximab and mycophenolate among children with CNI-resistant FSGS can emerge as a promising and efficacious treatment strategy to ensure sustained remission in this subset of patients.

Keywords: *Calcineurin inhibitor resistance, FSGS, mycophenolate, rituximab*

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Introduction

Idiopathic nephrotic syndrome affects 1–3 per 100,000 children per year. Approximately 85% are steroid sensitive.[1] About 30% of children with steroid-sensitive nephrotic syndrome may exhibit steroid dependence.[2] Kumar *et al*.[3] reported FSGS as the most common histologic subtype occurring in 38% of the children. Similarly, Arif *et al*.,[4] reported FSGS as the most common histologic subtype in 46.8% of cases of children who were biopsied for idiopathic NS. The estimated incidence of FSGS is approximately 7 per million with a prevalence of 4%.[5]

McGrogan *et al*.[6] reviewed published literature from around the world and reported that annual incidence rates is 0.2–1.8/100,000 population per year. FSGS is not a single disease, but rather a histological pattern of glomerular lesions caused by diverse clinicopathological entities with different mechanisms of injury with the podocyte as the principal target of the lesion, leading to characteristic lesions in part (i.e., focal) or some (i.e., segmental) glomeruli.[7] FSGS remains one of the most difficult-to-treat forms of nephrotic syndrome. Immunosuppressive therapy is the first treatment option for primary FSGS.[8]

Achievement of even partial remission is associated with better long-term survival. Among responders, some patients develop steroid dependence, but long-term use can lead to steroid toxicity. Calcineurin inhibitors are the most effective steroid-sparing agents used in children with nephrotic syndrome with FSGS.[8] However, treating children with FSGS who are resistant to CNIs may pose a challenge with limited therapeutic options.

In this retrospective study, we analyze the efficacy of rituximab and prolonged mycophenolate maintenance therapy for children with CNI-resistant FSGS.

Materials and Methods

We retrospectively reviewed the medical records of children with FSGS who were

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CNI resistant (had two or more relapses on CNI despite adequate trough levels or had acute/chronic CNI toxicity) and were subsequently treated with inj. rituximab and mycophenolate maintenance therapy for sustained remission. They were followed up at the Pediatric Nephrology Unit at our center between June 2018 and May 2022. The institutional ethical review board approved the study, and appropriate consent was taken from subjects.

All patients included in this retrospective analysis met the following criteria: age 2–18 years with a follow-up of at least 1 year; idiopathic nephrotic syndrome with FSGS proven by renal histopathology; classified as CNI-resistant FSGS. It included children in a state of non-remission despite 6 months of continuous therapy with CNIs with adequate drug trough levels (100–150 ng/mL for Cyc A and 5–10 ng/mL for Tacrolimus) or two or more relapses while on CNIs while maintaining adequate drug trough levels; and those with CNI toxicities which mandated discontinuation of CNI to prevent further deterioration of renal functions. CNI toxicity was considered if an elevation of serum creatinine >30% of baseline despite maintaining adequate trough levels was documented or histopathological analysis revealed stripped fibrosis with isometric vacuolization of tubules along with other features of CNI toxicity. Those with **s**econdary causes of FSGS were excluded.

Standard definitions were used for nephrotic syndrome, remission, and relapse according to KDIGO guidelines.

Rituximab and mycophenolate were administered to all the children with CNI-resistant FSGS. Two doses of rituximab infusions were administered at an interval of 14 days at 375 mg/m^2. Mycophenolate was simultaneously started at 1200 mg/m^2 in two divided doses. Oral steroids were gradually tapered and stopped over 6–8 weeks. Cotrimoxazole was given to all children for at best 6 months for pneumocystis prophylaxis. All patients were followed up every 3 months to evaluate remission status and monitor the adverse effects of drugs.

Results

Table 1 presents the baseline characteristics of all 13 children. Nine (69.2%) were males. Ten (76.9%) of them had an SD course, and the remaining three (23.1%) had an SR course. For all the children recruited, CNIs were used as the first-line immunosuppressant after prednisolone. Later, they were classified as CNI resistant when there was a failure of induction of remission despite 6 months of continuous therapy with CNI with adequate trough levels or two or more relapses occurred while on CNI and maintaining adequate trough levels or showed features of acute or chronic toxicity mandating discontinuation of CNI. The median age of the institution of dual therapy of 13 recruited children was 7.8 years (range: 2.4–17.6 years). Among 13 children, three had AKI after initiation of CNI therapy which reversed after cessation of CNI and one had chronic toxicity to CNI after administration for 3 years as revealed by histopathology thus mandating discontinuation of CNI therapy. Among the other nine children, six had two or more relapses on CNI despite adequate drug levels and three children had no response to therapy with CNI at all despite 6 months of continuous therapy with adequate drug levels. Eleven (84.6%) were in complete remission at least for 1 year post-treatment [Table 2] and two (15.4%) remained in a state of non-remission. Out of 11 children who remained in sustained remission, two cases relapsed (cases 2 and 11; Table 2) after 2 years and 1 year, respectively. After induction of remission with oral prednisolone, both were given two more doses of rituximab, and mycophenolate was continued, following which the children are in sustained remission till the last date of follow-up.

There were no major adverse reactions during rituximab and mycophenolate combination therapy. The children

Table 1: Clinical profile of children with CNI-resistant FSGS

Case no.	Age	Sex	Course	Duration of CNI treatment	Response to CNI therapy	CNI status (inference)
1.	7 years 11 months	M	SD	1.5 years	2 R + AKI in 2nd R	Resistant
2.	9 years 10 months	F	SD	2.5 years	2 R	Resistant
3.	10 years 9 months	M	SD	2 years	2 R	Resistant
4.	6 years 4 months	M	SR	4 years	3 R	Resistant
5.	2 years 4 months	M	SD	6 months	No response + HTN	Resistant
6.	7 years 3 months	M	SD	2.5 years	2 R	Resistant
7.	4 years 5 months	F	SD	2 years	2 R + AKI in 2nd R	Resistant
8.	8 years 7 months	M	SD	2 years	3 R	Resistant
9.	7 years 8 months	M	SD	3 years CYC +2 years TAC	2+3 R	Resistant
10.	3 years 5 months	F	SR	1 year	2 episodes of AKI	Resistant
11.	15 years 10 months	M	SD	6 months	No response	Resistant
12.	17 years 10 months	M	SD	3 years	Chronic CNI toxicity	Resistant
13.	3 years 2 months	F	SR	6 months	No response	Resistant

Age: At the time of switching to the proposed combination therapy, Y- Years, m – months, M- male, F- female, SD- Steroid dependent, SR- Steroid resistant, CYC- Cyclosporin, TAC- Tacrolimus, R- relapses, AKI – Acute Kidney Injury, HTN- Hypertension

Table 2: Outcome of dual therapy with rituximab and mycophenolate in children with CNI-resistant FSGS, follow-up, and adverse effects

Case no	Total doses of RTX	Response	Total duration of follow-up post RTX/MMF	No of relapses	Adverse effects/ infections during therapy
1.	2	CR	2 years 6 months	None	None
2.	4	CR	4 years	1 R after 2 years	None
3.	2	CR	1 year 9 months	None	None
4.	2	CR	2 years	None	None
5.	2	CR	1 year 9 months	None	None
6.	2	CR	1 year 6 months	None	None
7.	2	CR	1 year 6 months	None	Mild COVID
8.	2	CR	1 year 3 months	None	Mild COVID
9.	2	CR	1 year 8 months	None	None
10.	2	No response	1 year	-	None
11.	4	CR	1 year	1 R after 1 year	None
12.	2	CR	1 year	None	None
13.	2	No response	1 year	-	None

RTX- Rituximab, MMF- mycophenolate, CR- complete remission, R- relapse

have been followed up every third month from the initiation of the proposed regime to detect any evidence of relapse and to monitor any adverse drug effects. CD 19 levels were measured at 3 and 6 months for all the children after administration of rituximab. There was no increased incidence of infections during the period of follow-up with preserved renal functions. Two out of the 13 children were reported COVID positive but recovered uneventfully with conservative management on out-patient basis with no increased tendency of relapse.

Discussion

There is a paucity of data on treatment strategies for CNI-resistant FSGS among children; this greatly limits the treatment plans in such cases. There is ample data to suggest that monotherapy with the other available immunosuppressants such as mycophenolate, rituximab, and cyclophosphamide in FSGS is insufficient to maintain sustained and prolonged remission. Our analysis reveals a high rate of cumulative sustained remission for at least a year and beyond with combined therapy of rituximab and mycophenolate among pediatric patients with FSGS who were resistant to CNI.

The rationale of dual therapy is to stabilize the podocyte cytoskeleton by the dual pathway of action of both drugs. In view of unknown putative factors causing FSGS but amenable to immunosuppressive therapy, the rationale of using dual therapy was formulated. Rituximab is a monoclonal antibody directed against the CD 20-positive lymphocytes. However, rituximab has also been shown to play a role in B cell-independent mechanisms. Rituximab was demonstrated to regulate the activity of acid sphingomyelinase (ASMase), which is essential for signaling molecules on podocytes. Rituximab might cross-react with sphingomyelin phosphodiesterase acid-like 3b (SMPDL-3b). Rituximab thus prevents the actin cytoskeleton remodeling in podocytes by preserving the sphingolipid-related enzymes, SMPDL-3b, and ASMase. This might help rituximab to increase the susceptibility of the disease toward mycophenolate, thus synergistically contributing toward achieving a sustained and complete remission in children with FSGS.

Few studies evaluate mycophenolate as monotherapy alone for the treatment of FSGS.[9,10] Lau *et al*. published a systemic review in May 2013 on mycophenolate for primary FSGS.[11] It included three randomized controlled trials (RCTs) and 18 uncontrolled pre-post studies. Among them, only three uncontrolled pre-post studies used mycophenolate as monotherapy. The remaining three RCTs and 15 uncontrolled studies used MMF as add-on therapy. The majority consider mycophenolate as add-on therapy.[12-14] Cattran *et al*.[9] conducted a 6-month trial of mycophenolate in 18 patients of FSGS who were resistant to corticosteroid and CNI or cyclophosphamide. None of the patients achieved complete remission. Although a significant improvement in proteinuria was seen in 44% of patients by 6 months, this was sustained for up to 1 year post treatment in only 50% of the group, suggesting that mycophenolate monotherapy is inadequate and combination therapy with other immunosuppressants may be required for more prolonged and sustained remission. Senthil Nayagam *et al*.[12] in an RCT published in 2011 compared combination therapy of mycophenolate and prednisolone versus conventional therapy (prednisolone only) in FSGS. A 6-month treatment with combined mycophenolate and prednisolone was found as effective as the conventional treatment in short term but at the cost of prolonged steroid therapy.

Martinelli *et al*.[15] in 2004 published a long-term trial comprising 54 patients comparing cyclophosphamide

in combination with prednisone, as compared with prednisone alone for the treatment of nephrotic syndrome due to FSGS. Complete remission occurred in 20.4% and partial remission in 14.8% of the patients treated with steroids as compared to 26.7 and 20.0% of the patients treated with cyclophosphamide + prednisone, respectively, thus providing no overwhelming advantage in the face of increased adverse effects associated with cyclophosphamide such as bone marrow suppression and hemorrhagic cystitis.

Although there are many reports of the excellent steroid-sparing effect of rituximab in idiopathic complicated childhood nephrotic syndrome, most patients are likely to relapse after recovery of B lymphocytes. Ravani *et al*.[16] in an RCT demonstrated that the relapse rate at 6 and 12 months of follow-up with rituximab treatment alone in cases of SDNS or CNI-dependent NS were 50% and 75%, respectively. Hansrivijit *et al*.[17] in a systemic review and meta-analysis published in 2020 reviewed the effects of rituximab therapy in FSGS in adults. The overall remission rate and relapse rate of rituximab therapy in FSGS were 53.6% and 47.3%, respectively. Complete remission occurred in 42.9% of cases. Despite the good initial clinical response, rituximab responders remain prone to further relapses, necessitating a repeat course of rituximab or the addition of another steroid-sparing immunosuppressant.

From the above discussion, it is evident that mycophenolate or rituximab when used as a monotherapy in children with nephrotic syndrome with FSGS may not be sufficient alone to maintain sustained and prolonged remission. Thus, the continuation of a maintenance immunosuppressive agent after induction with rituximab may be recommended for the prevention of relapse. Basu *et al*.[18] published a retrospective analysis of 24 children of SRNS treated with rituximab. Although the analysis was done majorly on subjects with minimal change disease, the study population also comprised 11 FSGS patients, out of which six showed no response and five children showed partial or complete remission. Subsequently, these five children were put on maintenance mycophenolate therapy. At the end of 1 year, three out of the five children had sustained complete remission and two were in partial remission. However, by the end of 24 months, all children with FSGS relapsed, necessitating subsequent courses of rituximab. This strategy significantly prolonged the relapse-free period and reduced the relapse rate and daily dose of steroids as compared to studies of rituximab monotherapy. A recent study has been done by Sinha *et al*.[19] on the use of rituximab in CNI refractory SRNS. Post infusion of rituximab, steroids were tapered but CNI or mycophenolate was continued. Among 31 children with CNI refractory SRNS, 58% of children achieved CR or PR. Even the achievement of PR or CR is considered beneficial because it delays/prevents the onset of end-stage renal disease.

In our study, rituximab and mycophenolate were administered after induction of remission with oral prednisolone at a dose of 2 mg/kg/day or at least in partial remission. It is believed that the effect of rituximab is lower during the phase of nephrotic range proteinuria as a significant amount of rituximab is lost in the urine, resulting in lower rituximab levels and faster recovery of B lymphocytes. In addition, during the phase of nephrotic range proteinuria, there is a loss of valuable immunoglobulins and other protective factors simultaneously; rituximab administration might invite potentially dangerous infective complications.

Two children showed no response to two doses of rituximab and mycophenolate therapy beyond 6 months. One of them had C1q nephropathy with FSGS pattern on histopathologic examination.

Considering the promising results demonstrated by this single-center study with a limited number of subjects, further randomized multicentric trials should be conducted to explore the benefits of this combination therapy.

Conclusion

Dual combination therapy with rituximab and mycophenolate among children with CNI-resistant FSGS can be a promising and efficacious treatment strategy to ensure sustained remission in this subset of patients.

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Conflicts of interest

There are no conflicts of interest.

References

1. Trautmann A, Vivarelli M, Samuel S, Gipson D, Sinha A, Schaefer F, *et al*. IPNA clinical practice recommendations for the diagnosis and management of children with steroid-resistant nephrotic syndrome. Pediatr Nephrol 2020;35:1529-61.
2. Alharthi AA. Patterns of childhood steroid-sensitive and steroid resistant nephrotic syndrome in saudi children. Clin Pediatr (Phila) 2017;56:177-83.
3. Kumar J, Gulati S, Sharma AP, Sharma RK, Gupta RK. Histopathological spectrum of childhood nephrotic syndrome in Indian children. Pediatr Nephrol 2003;18:657-60.
4. Arif MK, Arif M, Amjad N. A histopathological outlook on nephrotic syndrome: A pediatric perspective. Indian J Nephrol 2016;26:188-91.
5. Kitiyakara C, Eggers P, Kopp JB. Twentyone-year trend in ESRD due to focal segmental glomerulosclerosis in the United States. Am J Kidney Dis 2004;44:815-25.
6. McGrogan A, Franssen CF, de Vries CS. The incidence of primary glomerulonephritis worldwide: A systematic review of the literature. Nephrol Dial Transplant 2011;26:414-30.
7. Shabaka A, Tato Ribera A, Fernández-Juárez G. Focal segmental glomerulosclerosis: State-of-the-Art and clinical perspective. Nephron 2020;144:413-27.
8. Han KH, Kim SH. Recent advances in treatments of primary

focal segmental glomerulosclerosis in children. Biomed Res Int 2016;2016:3053706.
9. Cattran DC, Wang MM, Appel G, Matalon A, Briggs W. Mycophenolate mofetil in the treatment of focal segmental glomerulosclerosis. Clin Nephrol 2004;62:405-11.
10. Choi MJ, Eustace JA, Gimenez LF, Atta MG, Scheel PJ, Sothinathan R, *et al*. Mycophenolate mofetil treatment for primary glomerular diseases. Kidney Int 2002;61:1098-114.
11. Lau EW, Ma PH, Wu X, Chung VC, Wong SY. Mycophenolate mofetil for primary focal segmental glomerulosclerosis: Systematic review. Renal Fail 2013;35:914-29.
12. Senthil Nayagam L, Ganguli A, Rathi M, Kohli HS, Gupta KL, Joshi K, *et al*. Mycophenolate mofetil or standard therapy for membranous nephropathy and focal segmental glomerulosclerosis: A pilot study. Nephrol Dial Transplant 2008;23:1926-30.
13. Gipson DS, Trachtman H, Kaskel FJ, Greene TH, Radeva MK, Gassman JJ, *et al*. Clinical trial of focal segmental glomerulosclerosis in children and young adults. Kidney Int 2011;80:868-78.
14. El-Reshaid K, El-Reshaid W, Madda J. Combination of immunosuppressive agents in treatment of steroid-resistant minimal change disease and primary focal segmental glomerulosclerosis. Ren Fail 2005;27:523-30.
15. Martinelli R, Pereira LJ, Silva OM, Okumura AS, Rocha H. Cyclophosphamide in the treatment of focal segmental glomerulosclerosis. Braz J Med Biol Res 2004;37:1365-72.
16. Ravani P, Magnasco A, Edefonti A, Murer L, Rossi R, Ghio L, *et al*. Short-term effects of rituximab in children with steroid-and calcineurin-dependent nephrotic syndrome: A randomized controlled trial. Clin J Am Soc Nephrol 2011;6:1308-15.
17. Hansrivijit P, Cheungpasitporn W, Thongprayoon C, Ghahramani N. Rituximab therapy for focal segmental glomerulosclerosis and minimal change disease in adults: A systematic review and meta-analysis. BMC Nephrol 2020;21:1-11.
18. Basu B, Mahapatra TKS, Mondal N. Mycophenolate mofetil following rituximab in children with steroid-resistant nephrotic syndrome. Pediatrics 2015;136:e132-9.
19. Sinha R, Banerjee S, Mukherjee A, Pradhan S, Akhtar S. Early use of rituximab in calcineurin inhibitor–refractory and steroid-resistant nephrotic syndrome. Kidney Int Rep 2020;5:2354-7.