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Treatment with tacrolimus and prednisolone is preferable to intravenous cyclophosphamide as the initial therapy for children with steroid-resistant nephrotic syndrome

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There are limited data on the relative efficacy and safety of calcineurin inhibitors and alkylating agents for idiopathic steroid-resistant nephrotic syndrome in children. To clarify this, we compared tacrolimus and intravenous cyclophosphamide therapy in a multicenter, randomized, controlled trial of 131 consecutive pediatric patients with minimal change disease, focal segmental glomerulosclerosis, or mesangioproliferative glomerulonephritis, stratified for initial or late steroid resistance. Patients were randomized to receive tacrolimus for 12 months or 6-monthly infusions of intravenous cyclophosphamide with both arms receiving equal amounts of alternate-day prednisolone. The primary outcome of complete or partial remission at 6 months, based on spot urine protein to creatinine ratios, was significantly higher in children receiving tacrolimus compared to cyclophosphamide (hazard ratio 2.64). Complete remission was significantly higher with tacrolimus (52.4%) than with cyclophosphamide (14.8%). The secondary outcome of sustained remission or steroid-sensitive relapse of nephrotic syndrome at 12 months was significantly higher with tacrolimus than cyclophosphamide. Treatment withdrawal was higher with cyclophosphamide, chiefly due to systemic infections. Compared to cyclophosphamide, 3 patients required treatment with tacrolimus to achieve 1 additional remission. Thus, tacrolimus and prednisolone are effective, safe, and preferable to cyclophosphamide as the initial therapy for patients with steroid-resistant nephrotic syndrome.

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The management of children with idiopathic steroid-resistant nephrotic syndrome is difficult, and there is no consensus on the most appropriate therapy.[1,2] The aim of treatment is complete or partial remission of proteinuria, the most important predictor of long-term outcome.[3,4] Although a report from the International Study of Kidney Diseases in Children showed no benefit with oral cyclophosphamide,[5] results from case series[6,7] and small trials[8,9] suggest that therapy with intravenous (i.v.) pulse cyclophosphamide is effective in 40–60% patients with steroid-resistant nephrotic syndrome. Although the use of i.v. cyclophosphamide carries risks of infections and gonadotoxicity, these results emphasize the need for a prospective evaluation of this agent.

Therapy with cyclosporine has shown higher efficacy, with remission rates of 70–80%,[10–12] but it is prolonged, expensive, and requires monitoring for nephrotoxicity and other adverse effects.[13,14] Although results from case series[15,16] and a randomized study[17] suggest that tacrolimus has comparable efficacy and less cosmetic side effects, this has not been examined in a large controlled study. In view of limited comparative data on the efficacy of various medications, we prospectively evaluated the efficacy and safety of therapy with tacrolimus and alternate-day prednisolone compared with i.v. cyclophosphamide and alternate-day prednisolone in patients with idiopathic steroid-resistant nephrotic syndrome.

RESULTS

Patient description

Of the 173 patients screened, 42 were excluded (Figure 1). Of those enrolled, 66 were randomized to the tacrolimus group

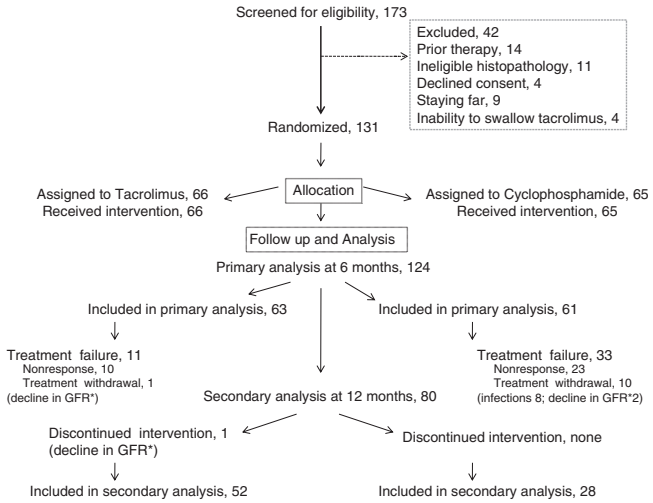


Figure 1 | Participant flow. Seven patients were lost to follow-up (tacrolimus group, three; cyclophosphamide group, four) after the first visit and were not included in the intention-to-treat analysis. Patients with treatment failure at 6 months were not included in the secondary analysis. *Decline in estimated glomerular filtration rate to <50 ml/min per 1.73 m^2.

and 65 to the cyclophosphamide group. Primary outcome was assessed in 124 patients, as 7 did not return after the first visit. One patient in the cyclophosphamide group was lost to follow-up after 5 months, and three did not return after the 8- to 10-month visits. Baseline characteristics were similar in the groups (Table 1); the cohort included 65.7% boys, and 61.8% had initial resistance. Minimal change disease and focal segmental glomerulosclerosis (FSGS) were present in 59.5% and 32.8% cases, respectively.

Table 1 | Baseline demographic and clinical characteristics

Characteristic	Tacrolimus, 66	Cyclophosphamide, 65	*P*-value
Age at enrollment, months	62.1 ± 39.1	74.5 ± 43.9	0.08
Boys	41	45	0.39
Weight, kg	18.74 ± 8.58	19.9 ± 9.41	0.46
Weight SDS[a]	−0.32 ± 1.57	−0.72 ± 1.39	0.33
Height, cm	101.69 ± 19.89	107.86 ± 23.44	0.16
Height SDS[a]	−1.53 ± 1.93	−1.49 ± 2.19	0.31
Illness before enrollment, months	13.9 ± 18.2	17.8 ± 29.9	0.36
Initial/late resistance	42[b]/24	39[b]/26	0.66
Histopathology			
Minimal change disease	44	34	0.09
Focal segmental glomerulosclerosis	17	26	0.08
Not otherwise specified/hilar	16/1	24/2	
Mesangioproliferative glomerulonephritis	5	5	0.9
Laboratory			
Serum creatinine, mg/dl	0.53 ± 0.12	0.51 ± 0.23	0.21
Serum albumin, g/dl	2.2 ± 0.7	2.3 ± 0.7	0.45
Serum cholesterol, mg/dl	376.0 ± 148.3	354.7 ± 139.8	0.41
eGFR[c], ml/min per 1.73 m^2	93.6 ± 33.2	92.1 ± 30.5	0.77
Urine protein/creatinine[d], mg/mg	6.50 ± 4.8	5.3 ± 2.9	0.27

[a]Weight standard deviation score (SDS) and height SDS were calculated using *WHO Antroplus version 1.0.4*.
[b]*NPHS2* gene sequencing in 21 patients with initial resistance showed 2 heterozygous (R229Q) mutations and 1 compound heterozygous mutation (p.R229Q/p.A308T). No mutations were detected in *WT-1* gene.
[c]Glomerular filtration rate estimated by the equation: 0.413 × (height/serum creatinine; see ref. 24).
[d]First morning spot urine sample.
Values are expressed as means ± s.d.
Groups were compared using *t*-test for continuous variables and χ^2 test for categorical variables. There were no significant differences between the two treatment groups with respect to any characteristic.

Therapy

All patients received the allocated intervention. The dose of tacrolimus and cyclophosphamide was 0.12 ± 0.03 mg/kg/day and 554.1 ± 98.2 mg/m^2/dose, respectively. The trough level of tacrolimus at 4 weeks was 5.8 ± 1.9 ng/ml. The dose of enalapril was 5.8 ± 2.1 and 5.5 ± 2.3 mg/day in tacrolimus and cyclophosphamide groups, respectively. The respective cumulative doses of prednisolone were 0.44 ± 0.19 and 0.39 ± 0.19 mg/kg/day for the first 6 months ($P = 0.18$), and 0.35 ± 0.15 and 0.34 ± 0.12 mg/kg/day for 12 months ($P = 0.74$).

Primary outcome. The primary outcome variable of the proportion of patients in complete or partial remission at 6 months was significantly higher with tacrolimus (82.5%) as compared with cyclophosphamide (45.9%; $P < 0.001$, Table 2). The proportion of patients showing complete remission was also higher with tacrolimus (52.4%) compared with cyclophosphamide (14.8%; $P < 0.001$).

The frequency of treatment failure was lower with tacrolimus (17.4%) than with cyclophosphamide (54.1%; $P < 0.001$). In all, 10 of 11 treatment failures with tacrolimus and 23 of 33 failures with cyclophosphamide were due to nonresponse to therapy at 6 months. Therapy was withdrawn in one patient in the tacrolimus group and two in the cyclophosphamide group owing to a decline in the glomerular filtration rate (GFR). Eight patients withdrew therapy with cyclophosphamide (one patient after 2 pulses; three each after 3 and 4 pulses; one after 5 pulses) because of recurrent serious infections.

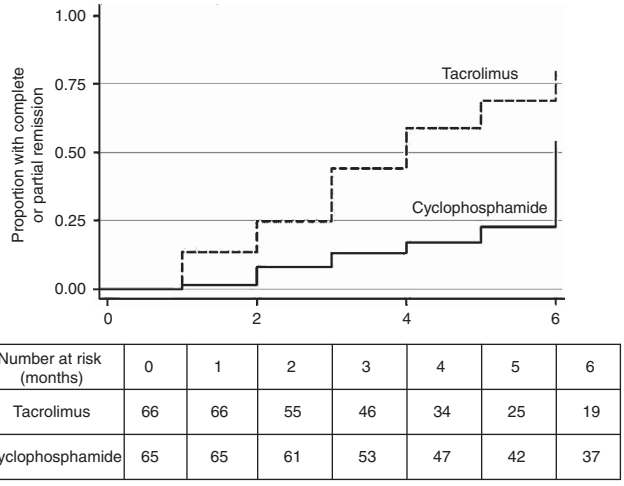
Survival analysis. Patients receiving tacrolimus showed a higher probability of complete or partial remission compared with cyclophosphamide (log-rank $P < 0.001$; hazard ratio (HR) 2.64; 95% confidence interval (95% CI) 1.67–4.19 ($P < 0.001$);Figure 2). The mean time to remission was 3.5 ± 1.7 months with tacrolimus and 4.5 ± 1.7 months with cyclophosphamide. The likelihood of remission was higher with minimal change disease (HR 1.74; 95% CI 1.08–2.80, $P = 0.02$) when compared with FSGS and mesangioproliferative glomerulonephritis. Other baseline characteristics did not affect the outcome. The benefit of tacrolimus in terms of attaining remission was maintained after adjustments for type of resistance and histopathology (adjusted HR 2.61; 95% CI 1.59–4.26, $P < 0.001$).

Subgroup analysis. For patients receiving tacrolimus, complete or partial remission within the subgroup of initial or late resistance was achieved in 33 (84.61%) versus 19

Table 2 | Primary and secondary outcomes

Primary (6-month) outcome	Tacrolimus, 63	Cyclophosphamide, 61	*P*-value
Remission	52 (82.5)	28 (45.9)	<0.001
Complete	33 (52.4)	9 (14.8)	<0.001
Partial	19 (30.1)	19 (31.1)	0.90
Treatment failure	11 (17.4)	33 (54.1)	<0.001
Nonresponse	10	23	
Withdrawal of therapy	1	10	
>1 serious infection	0	8	
Declining GFR[a]	1	2	
Secondary (12-month) outcome	**Tacrolimus, 52**	**Cyclophosphamide, 28**	***P*-value**
Sustained remission	38 (73.1)	12 (42.9)	0.002
Steroid-sensitive course	2	2	
Nonnephrotic proteinuria	11 (21.2)	11 (39.3)	0.08
Resistant nephrotic syndrome	0	3	
Withdrawal of therapy	1	0	

[a]Decline in estimated glomerular filtration rate (GFR) to <50 ml/min per 1.73 m^2.
Values in parentheses show percentages.



Number at risk (months)	0	1	2	3	4	5	6
Tacrolimus	66	66	55	46	34	25	19
Cyclophosphamide	65	65	61	53	47	42	37

Figure 2 | Kaplan–Meier curve showing the probability of attaining complete or partial remission. The intention-to-treat population, comprising 124 patients, showed a significantly higher probability of complete or partial remission with tacrolimus and alternate-day prednisolone. *P*-value (log rank) <0.001. Data were censored at 5 months in one patient lost to follow-up thereafter.

(79.16%) patients, respectively (*P*-value = 0.58). The likelihood of remission was significantly higher with tacrolimus within the subgroups of initial resistance (HR 2.78, 95% CI 1.54–5.03, $P = 0.001$), late resistance (HR 2.35, 95% CI 1.11–4.97, $P = 0.02$), minimal change disease (HR 2.37, 95% CI 1.32–4.23, $P = 0.004$), and FSGS (HR 2.54, 95% CI 1.09–5.93, $P = 0.03$; Figure 3).

Secondary outcome. Of the 80 patients with complete or partial remission at 6 months, remission was sustained in more patients receiving tacrolimus (73.1%) compared with cyclophosphamide (42.9%; $P = 0.002$; Table 2). All patients attaining complete remission (tacrolimus, 33; cyclophosphamide, 9) showed favorable outcome (sustained remission

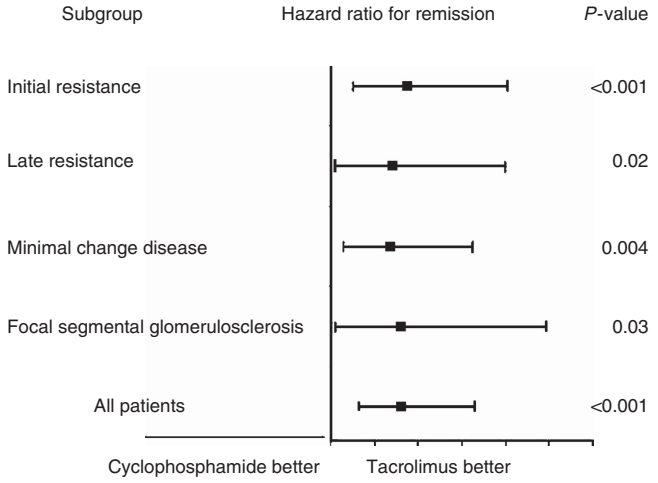


Figure 3 | Forest plot showing the comparative efficacy of tacrolimus to intravenous (i.v.) cyclophosphamide, as ascertained by complete or partial remission at 6 months, in subgroups of patients with steroid-resistant nephrotic syndrome. The horizontal bars represent 95% confidence intervals.

or steroid-sensitive illness) at 12 months. Of the 38 patients in partial remission (tacrolimus, 19; cyclophosphamide, 19), 22 had nonnephrotic proteinuria, 12 showed favorable outcome, 3 had recurrent steroid resistance, and 1 showed decline in GFR. The odds of a 12-month favorable outcome were higher with tacrolimus (OR 3.33; 95% CI 1.24–8.90, $P = 0.016$).

Change in proteinuria. Urine protein to creatinine ratio (Up/Uc, mg/mg) in tacrolimus and cyclophosphamide groups was 3.1 ± 3.8 versus 3.2 ± 2.4 ($P = 0.87$) at 2 months, 1.4 ± 2.9 versus 6.5 ± 8.8 ($P = 0.01$) at 4 months, and 0.9 ± 1.5 versus 2.5 ± 4.1 ($P = 0.01$) at 6 months, respectively. The former showed higher decline in protein excretion from baseline until 6 months (tacrolimus 6.3 ± 8.7 vs. cyclophosphamide 2.3 ± 4.7; $P = 0.01$).

Estimated GFR. The decline in GFR <50 ml/min per 1.73 m^2 occurred in two patients each with tacrolimus and cyclophosphamide. At 12 months, the GFR had declined by 9 and −1.5 ml/min per 1.73 m^2, respectively; the differences were not significant either from baseline ($P = 0.24$) or between groups ($P = 0.64$).

Number needed to treat. As compared with i.v. cyclophosphamide, three patients were required to be treated with tacrolimus and alternate-day prednisolone in order to achieve one additional complete or partial remission.

Safety and tolerability

Ten patients receiving cyclophosphamide had adverse events, resulting in withdrawal of treatment. Persistent nephrotoxicity necessitating tacrolimus discontinuation was seen in two cases and reversible toxicity in seven. Serious infections were higher with cyclophosphamide (16 episodes in 8 patients, including 1 death) compared with tacrolimus (4 episodes in 4 patients; $P = 0.004$; Table 3). No patients had hyperglycemia, neutropenia, or alopecia.

Table 3 | Adverse events and serious adverse events

Event	Tacrolimus, 66	Cyclophosphamide, 65
Any adverse event	45	54
Any serious adverse event	9	19
Persistent nephrotoxicity	2	—
Reversible nephrotoxicity	7	—
Serious infections[a]	4	16
Lower respiratory tract	3	8[b]
Peritonitis	1	8
Anasarca; hospitalization	3	3
Minor infections[c]	27	24
Seizures[d]	1	—
Headache[d]	1	—
Cystitis	—	2
Vomiting	—	9

[a]All managed during hospital stay.
[b]One patient died.
[c]Includes upper respiratory tract infections and gastroenteritis.
[d]Neuroimaging (magnetic resonance imaging) was normal.

DISCUSSION

The results of this trial show that treatment with tacrolimus and alternate-day prednisolone was effective and safe in inducing and sustaining remission in patients with newly diagnosed, steroid-resistant nephrotic syndrome. A higher proportion of patients treated with tacrolimus (82.5%) achieved complete or partial remission at 6 months compared with i.v. cyclophosphamide (45.9%); a majority of the former had favorable outcome at 12 months. Compared with therapy with i.v. cyclophosphamide, one additional remission was achieved for every three patients who received tacrolimus. Although this study provides the first prospective comparison between tacrolimus and i.v. cyclophosphamide, a recent report showed that therapy with the former resulted in remission in 11 of 17 adults with steroid- and cyclophosphamide-resistant nephrotic syndrome.[18] Another report on 19 children with diverse renal histologies showed that tacrolimus achieved complete remission in 11, including those unresponsive to cyclophosphamide.[19]

Although the efficacy of cyclosporine for resistant nephrotic syndrome has been shown previously,[10–12] the need for prolonged therapy and risk of nephrotoxicity has prompted evaluation of other agents. A National Institutes of Health (NIH) study compared the efficacy of 12 months of treatment with cyclosporine with a combination of dexamethasone and mycophenolate mofetil in 138 patients with FSGS.[20] Both groups showed similar benefits in attaining at least partial remission (cyclosporine 46%; combination 33%), and the study failed to demonstrate an unambiguous benefit with cyclosporine. The Arbeitsgemeinschaft für Pädiatrische Nephrologie (APN) group compared cyclosporine with i.v. cyclophosphamide in 32 children with steroid resistance.[21] Although cyclosporine induced partial remission in 60% patients compared with 17% with the latter, inference was limited as patients who did not respond by 12 weeks were excluded. A randomized study, on children with steroid-resistant nephrotic syndrome, showed that tacrolimus and cyclosporine had similar efficacy (remission in 85.7% and 80% patients, respectively).[17] Therapy with tacrolimus was associated with fewer relapses, less cosmetic effects, and lower incidence of hypertension.

This study shows that almost one-half of the patients treated with tacrolimus have complete remission compared with 14.8% with cyclophosphamide. These findings are contrary to the low rates of complete remission with cyclosporine in the NIH-FSGS trial (19%) and APN study (13.3%), but similar to those previously reported at this center (cyclosporine 50%; tacrolimus 42.8%).[17] All patients with complete remission at 6 months had a favorable outcome at 12 months, confirming that complete remission was valuable in the long-term management of patients.[3,4] The 12-month outcome was also satisfactory in patients with partial remission, with most showing complete remission or nonnephrotic proteinuria. A similar benefit of partial response in slowing the rate of progression and improving renal survival was reported previously in 281 patients with FSGS from the Toronto Glomerulonephritis Registry.[22]

The chief adverse events were acute nephrotoxicity with tacrolimus and serious infections with cyclophosphamide. Most episodes of nephrotoxicity responded to dose reduction, and tacrolimus discontinuation was rarely required. In the APN study, four patients not responding to therapy with cyclosporine showed decline in renal functions.[21] The NIH-FSGS study reported significantly lower median GFR at 26 weeks in patients receiving cyclosporine compared with those receiving mycophenolate mofetil. Although in this study data at 12 months were available only for those in remission, patients receiving tacrolimus showed a 10% reduction in GFR compared with the baseline. Future research should examine the role of biomarkers for detecting early toxicity and whether alternative agents can sustain tacrolimus-induced remission.

This study has certain limitations. It was not stratified based on renal histology, although there was equal distribution of histopathologies in both arms. However, although the NIH trial was limited to patients with FSGS,[20] we also included patients with minimal change disease. Although not powered for subgroup analysis, we found that therapy with tacrolimus was effective in patients with minimal change disease and FSGS. Second, although detailed genetic evaluation before enrollment would have been appropriate, this was carried out only in few patients with initial resistance. Experience from this center suggests that the proportion of patients with mutations in *NPHS2* and *WT1* genes is relatively low (Sinha *et al.*, abstract presented at ISN Forefronts Symposium, 22–25 September 2011 in Aarhus, Denmark). The strengths of this adequately sized trial were that randomization, data collection, and analysis were performed centrally. The baseline characteristics were well balanced, and a widely accepted definition of steroid resistance was used. Safety monitoring was ensured and nonresponders were managed appropriately. The results of this study on children with initial and late resistance and

major biopsy diagnoses are therefore generalizable. Therapy with tacrolimus and low-dose prednisolone should be preferred to cyclophosphamide as the initial therapy for patients with steroid-resistant nephrotic syndrome, as it is effective and safe in inducing and maintaining remission of proteinuria.

MATERIALS AND METHODS

Study design

This prospective, investigator-initiated, open-label, randomized, controlled trial compared the efficacy of tacrolimus with i.v. cyclophosphamide in combination with prednisolone for inducing remission in patients with steroid-resistant nephrotic syndrome. Patients were enrolled from March 2008 to September 2010 in pediatric nephrology units of five centers. An Ethics Committee at each site approved the study protocol. Either the parent or guardian provided written informed consent. The study was conducted in accordance with the original protocol and had no amendments. This study report complies with the Consolidated Standards of Reporting Trials statement.[23]

The coordinating center (All India Institute of Medical Sciences, New Delhi) conceived and designed the study protocol. Investigators at the coordinating center vouch for the accuracy and completeness in collation and analysis of data. The manuscript was reviewed, edited, and approved by all authors.

Study participants

Patients, 2–16 years old, with newly diagnosed initial or late steroid-resistant nephrotic syndrome and renal histological features suggestive of minimal change disease, FSGS, or mesangioproliferative glomerulonephritis were eligible. Initial resistance was the absence of remission despite therapy with prednisolone at 2 mg/kg/day (maximum 60 mg) for 4 weeks; patients with remission at onset but steroid resistance in a subsequent relapse were defined as late resistance. We included patients with an early-morning Up/Uc of >2.0 or the presence of $3+/4+$ proteinuria by dipstick test.

Patients with any of the following were excluded: (1) impaired renal function (estimated GFR <60 ml/min per 1.73 m^2);[24] (2) intake of immunosuppressive medications other than prednisolone in the preceding 6 months in patients with late resistance; (3) prior therapy with cyclophosphamide or calcineurin inhibitors; (4) infection with hepatitis B or C, or HIV; (5) IgA nephropathy or collapsing glomerulopathy; and (6) inability to swallow tacrolimus capsules.

Permuted block randomization with stratification, by initial or late resistance, was performed centrally by individuals not involved in trial implementation. The investigators were blinded to the randomization schedules and allocation was concealed in opaque sealed envelopes.

Intervention

Patients were allocated in a 1:1 ratio to receive therapy with either tacrolimus at a dose of 0.1–0.15 mg/kg/day for 12 months (Pangraf; Panacea, Mumbai, India; 0.5 and 1 mg capsules) or i.v. cyclophosphamide at 500 mg/m^2 once a month for 6 doses. Prednisolone was given at a dose of 1.5 mg/kg every other day for 2 weeks, and then tapered by 0.25 mg/kg every 2 weeks to 0.5 mg/kg on alternate days for 12 months. Dosing was based on recent body weight. All patients received treatment with enalapril (maximum 10 mg) and calcium supplements.

Dose adjustments with tacrolimus. Trough (12 h) levels were measured, using microparticle enzyme immunoassay, 2 weeks after beginning therapy or in the presence of acute nephrotoxicity. Dose adjustments, by 20–25%, were performed every 7–10 days until a tacrolimus level of 5–7 ng/ml was achieved; lower levels were accepted if patients were in remission. At each visit, a 1- to 2-month supply of tacrolimus was given and treatment adherence was assessed by pill count of the returned packs. Reversible nephrotoxicity was defined as $>30\%$ increase in the level of serum creatinine from the baseline, which improved within 2 weeks of reduction of tacrolimus dose.

Administration of cyclophosphamide. Cyclophosphamide was diluted in 200 ml normal saline and infused over 2 h. Coadministered medications included 2-mercaptoethane sulfonate sodium (same dose as cyclophosphamide; 20% i.v., 80% orally) and ondansetron (0.5 mg/kg i.v.).

Outcomes and follow-up

Parents were instructed to examine the first morning urine specimen for proteinuria by dipstick daily. Visits were scheduled once a month until 6 months, and then every 2 months until 12 months. At each visit, protocol-defined efficacy and safety parameters were reviewed. The Up/Uc ratio was measured on the first morning sample. Blood counts and levels of creatinine, albumin, cholesterol, glucose, and electrolytes were measured at each visit.

The primary efficacy end point, at 6 months, was the occurrence of complete or partial remission. Complete remission was defined as Up/Uc <0.2, confirmed once within the next 3 days; partial remission was Up/Uc 0.2–2, the absence of edema, and serum albumin >2.5 g/dl. Nonresponse was defined as Up/Uc >2, the presence of edema, or serum albumin <2.5 g/dl. Nonresponders exited the study at 6 months.

For those attaining remission, secondary end points at 12 months were as follows: (1) proportion of patients in sustained remission (Up/Uc <0.2) or with steroid-sensitive nephrotic syndrome, (2) subjects with nonnephrotic proteinuria (Up/Uc 0.2–2, no edema, albumin >2.5 g/dl) or recurrence of steroid resistance, (3) frequency and type of side effects, and (4) estimated GFR. The presence of sustained remission or steroid-sensitive course at 12 months was considered a favorable outcome.

Treatment of relapses. In patients achieving either complete or partial remission, relapse was the presence of $3–4+$ proteinuria by dipstick for 3 consecutive days. These patients were treated with prednisolone, at 2 mg/kg/day until remission (trace/negative protein for 3 days), followed by 1.5 mg/kg on alternate days for 2 weeks and then tapered.

Management of serious infections. Serious infections (for example, pneumonia, peritonitis, and cellulitis) were those that required hospitalization and were treated using standard protocols. Patients received prednisolone (0.2–0.3 mg/kg/day orally) or an equivalent dose of i.v. hydrocortisone during these infections.

Safety measures

Safety assessments included clinical and laboratory assessments, and monitoring for serious adverse events, including by the Data and Safety Monitoring Board. A 6-monthly report was submitted to the ethics committee. Patients who discontinued study medication were followed up for adverse events for 30 days after their last dose.

Treatment failure. Treatment failure was as follows: (1) nonresponse to therapy at 6 months, (2) >1 episode of serious infection requiring hospitalization, (3) persistent elevation in serum

creatinine $\geqslant$30% despite dose reduction, and (4) estimated GFR $<$50 ml/min per 1.73 m^2.

Statistical analysis

On the basis of occurrence of complete or partial remission in 50% patients receiving cyclophosphamide,[7,9] and assuming that therapy with tacrolimus would increase the remission rate by 30%, 64 patients were required per group to show significant differences at 90% power, two-tailed α of 5%, and dropout rate of 10%. Data were analyzed using Stata, version 11.2 (StataCorp 2009; Stata Statistical Software: Release 11; College Station, TX: StataCorp LP). Continuous data were expressed as the means $\pm$ s.d. Categorical data were analyzed with Pearson's χ^2 or Fisher's exact tests.

Efficacy analyses were performed on the intention-to-treat population, which included patients who returned for the first follow-up visit with last value carried forward. The effect of treatment on primary outcome was estimated by HR obtained from a Cox regression model. For patients achieving remission, the time from randomization to first achieving the 6-month response of either a complete or partial remission was taken for the time-to-event analysis. Kaplan–Meier survival estimates compared the time to achievement of remission using log-rank test. Logistic regression was used to calculate odds ratio for achieving a favorable outcome at 12 months. The number needed to be treated with tacrolimus so as to achieve one additional remission over that by i.v. cyclophosphamide was calculated.[25]

DISCLOSURE

All the authors declared no competing interests.

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