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Abstract

Introduction: We reported increased spleen tyrosine kinase (SYK) expression in kidney biopsies of patients with IgA nephropathy (IgAN) and that inhibition of SYK reduces inflammatory cytokines production from IgA stimulated mesangial cells.

Methods: This study was a double-blind, randomised, placebo-controlled phase 2 trial of fostamatinib (an oral SYK inhibitor) in 76 patients with IgAN. Patients were randomised to receive placebo, fostamatinib at 100 mg or 150 mg twice daily for 24 weeks on top of maximum tolerated dose of renin-angiotensin system inhibitors (RASi). The primary end point was reduction of proteinuria. Secondary endpoints included change from baseline in eGFR and kidney histology.

Results: While we could not detect significant reduction in proteinuria with fostamatinib overall, in a pre-determined subgroup analysis, there was a trend for dose-dependent reduction in median proteinuria (from baseline to 24 weeks by 14%, 27% and 36% in the placebo, fostamatinib 100 mg and 150 mg groups respectively) in patients with baseline urinary protein to creatinine ratios (UPCR) more than 1000 mg/g. Kidney function (eGFR) remained stable in all groups. Fostamatinib was well tolerated. Side effects included diarrhea, hypertension and increased liver enzymes. Thirty-nine patients underwent repeat biopsy showing reductions in SYK staining associated with therapy at low dose (-1.5 v 1.7 SYK+ cells/glomerulus in the placebo group, $p<0.05$).

Conclusions: There was a trend towards reduction in proteinuria with fostamatinib in a predefined analysis of high risk patients with IgAN despite maximal care, as defined by baseline UPCR greater than 1000 mg/g. Further study may be warranted.

KEY WORDS: IgA nephropathy; glomerulonephritis; inflammation; signaling

INTRODUCTION

IgA nephropathy (IgAN), defined by autoimmune mediated glomerular IgA deposition, is the commonest primary glomerulonephritis worldwide. Approximately 20-40% of patients with IgAN progress to kidney failure within 20 years of diagnosis, with significant risks of recurrence of IgAN in the transplanted kidney[1]. Effective treatment of hypertension using renin-angiotensin system inhibitors (RASi), supportive therapy including statins as well as lifestyle modification is effective in reducing proteinuria and preserving kidney function in some patients[2]. However, many patients continue to have significant proteinuria despite optimal supportive care, remaining at risk for progressive kidney dysfunction[3]. Trials of immunosuppressive therapy, especially corticosteroids, have suggested possible benefits in IgAN[4,5]. Indeed, a large retrospective analysis showed a benefit with corticosteroids or combined immunosuppressive therapy, even for patients with advanced kidney dysfunction[6]. However, prospective trials have either cast doubt on the efficacy of corticosteroids and/or suggested that the risks of immunosuppressive therapy, particularly infections and infection-related death, may outweigh benefits[2,7]. Recent clinical trials with reduced dose oral corticosteroid or gut-targeted corticosteroid suggested beneficial effects in IgA nephropathy[8,9].

An alternate treatment target is spleen tyrosine kinase (SYK), downstream of immune receptors, including Fc and B cell receptors[10]. We and others have shown that stimulation of mesangial cells *in vitro* with IgA from IgAN patients resulted in SYK phosphorylation and inflammatory cytokine synthesis, including IL-6 and chemokines (MCP-1 (CCL2), RANTES (CCL5), IP-10 (CXCL10), IL-8 (CXCL8)), platelet derived growth factor (PDGF) and cell proliferation[11,12]. Furthermore, binding of patients' IgA to β1,4-galactosyltransferase 1, an IgA receptor on human mesangial cells resulted in SYK phosphorylation and IL-6 production[13]. These inflammatory responses were reduced by an inhibitor of SYK or SYK siRNA[11]. Similarly, culture media from mesangial cells stimulated by patient IgA induced a proinflammatory phenotype in kidney tubular cells via activation of NF-κB and p-42/p-44 signalling and was suppressed by SYK inhibition[14]. Increased SYK expression was detected in kidney biopsies of IgAN patients [11,12,14-16] and was particularly high in biopsies showing mesangial or endocapillary proliferation[15].

We carried out a proof-of-principle study of an oral SYK inhibitor, fostamatinib, in a double-blind, randomised, placebo-controlled phase 2 trial in patients with IgAN. Our hypothesis was that SYK is involved in the inflammation and resultant kidney injury seen with mesangial IgA deposition and a potential therapeutic target.

CONCISE METHODS

The protocol was approved by institutional review boards at each participating centre and registered with clinicaltrials.gov, (NCT02112838). Participants provided informed consent before all study procedures, per the Declaration of Helsinki.

Trial design

Inclusion and exclusion criteria

Adults, aged 18-70 with a kidney biopsy within 180 days prior to the initial study visit, diagnostic of primary IgAN, were recruited. Participation required mesangial and/or endocapillary hypercellularity, and < 50% tubular atrophy or interstitial fibrosis (T0 or T1,Oxford Classification) and < 50% glomerular crescents. Biopsies obtained > 180 days prior to screening could be accepted with prior permission from the Medical Monitor if according to the Investigator there was no significant change in kidney status.

Participant proteinuria was required at > 1 gm/day, UPCR > 100 mg/mmol (> 884 mg/g), or spot albumin creatinine ratio > 70 mg/mmol at diagnosis of IgAN or any time prior to screening. Proteinuria needed to be > 0.50 gm/day [UPCR > 50 mg/mmol (> 442 mg/g)] at screening.

The eGFR needed to be > 30 mL/min/1.73 m^2 (MDRD equation) at the time of screening, but values > 25 mL/min/1.73 m^2 were permitted if there was no recent change in renal status.

Blood pressure control was ≤130/80 mmHg with angiotensin blockade with or without other anti-hypertensive agents. Patients were required to be on maximum approved (or tolerated) ACEi or ARB before an additional anti-hypertensive agent was added. Patients with low neutrophil count < 1,500/µL, Hgb < 9 g/dL, abnormal liver enzymes (ALT or AST of > 1.5x ULN), or bilirubin > 2.0 mg/dL were

excluded. Patients with secondary IgAN, including IgA vasculitis, active infection or serology results suggestive of active hepatitis B, C or human immunodeficiency virus were excluded. Patients who used immunosuppression, including cyclophosphamide, mycophenolate, azathioprine, rituximab (or other anti-B cell therapies), or more than 15 mg/day prednisolone (or corticosteroid equivalent) within 6 months prior to the pre-study kidney biopsies were excluded. Pregnant and kidney transplant patients were excluded.

Randomisation

Patients were stratified at randomisation by the presence or absence of endocapillary hypercellularity on biopsy. Patients were randomized to placebo, fostamatinib 100 mg or 150mg twice daily orally by a random allocation method (**Supplementary Figure S1**). Treatment was for 24 weeks.

Outcomes

Primary endpoint

The absolute reduction in proteinuria, measured by random UPCR (least square (LS) mean change) from baseline to 24 weeks was the primary endpoint, analyzed using an analysis of covariance (ANCOVA) model in the ITT population. This model included both the treatment group and presence/absence of endothelial hypercellularity at baseline as factors and adjusted for proteinuria at baseline (as a covariate). The UPCR was log-transformed prior to analysis. Missing Week 24 data was imputed using a multiple imputation procedure.

Secondary endpoint

Secondary efficacy endpoints were analyzed in the ITT and per-protocol (PP) populations. The primary efficacy endpoint was analyzed using the PP population as a sensitivity analysis. No adjustments were made for multiplicity. Histology was assessed by two independent nephropathologists, and a third pathologist adjudicated for any discrepancies. Multiple secondary endpoints were analyzed using ANCOVA models including the treatment group and baseline endothelial hypercellularity as factors,

with mean estimates of the variable adjusted for the baseline value: (1) Mean change post-treatment in mesangial hypercellularity on renal biopsies. (2) Mean change post-treatment in endocapillary hypercellularity on renal biopsies. (3) Mean change post-treatment in segmental or global glomerulosclerosis on renal biopsies. (4) Mean change post-treatment in tubulointerstitial scarring on renal biopsies. (5) Mean change from Baseline of eGFR at 12 weeks). (6) Mean change from Baseline of eGFR at 24 weeks . (7) Mean change from Baseline of proteinuria at 12 weeks . (8) Mean change of hematuria (dipstick test) from Baseline at 12 weeks . (9) Mean change of hematuria (dipstick test) from Baseline at 24 weeks .

Pre-specified subgroup analysis

Pre-specified analysis of patients with baseline UPCR >1000 mg/g was included in order to understand potential therapeutic effects in patients with different levels of proteinuria.

Determination of sample size

A sample size of 25 evaluable patients in each of the 3 treatment groups was predicted to have an 80% power to detect a 43% reduction in proteinuria from Baseline (Visit 2) to 24 weeks (Visit 9) between the pooled fostamatinib and placebo groups, using a 2-sided t-test, $\alpha = 0.05$ and log transformed data assuming that the treatment groups had the same mean and standard deviations of UPCR (1150 mg/g ± 1060 mg/g) at Baseline and that the values for the placebo group remained constant over 24 weeks. Treatment allocation ratio was 1:2 for the placebo: combined fostamatinib groups.

Data management and statistics

The ITT population included all randomised patients. All efficacy endpoints were analysed based on the ITT population, and patients were analysed according to their randomised treatment assignment. The efficacy analyses based on the ITT population were considered the primary efficacy analyses.

The PP population included all the patients in the ITT population who had no major protocol violations. Baseline measurements were the last measurement for the corresponding variable prior to the first

randomised dose. For the primary endpoint analysis, missing Week 24 (Visit 9) values of UPCR were imputed using a multiple imputation method.

RESULTS

Recruitment

Twenty-five centres from Asia, Europe and North America participated. One hundred and eleven patients were screened (**Supplementary Figure S2**). Seventy-six patients were successfully randomised (**Table 1**).

Patients received treatment with maximal approved (or tolerated) doses of angiotensin converting enzyme inhibitor (ACEi) and/or angiotensin receptor blocker (ARB) for at least 90 days prior to screening. In each group, there was one subject intolerant of any ACEi or ARB (**Table 1**). Patients remained on stable dose ACEi or ARB throughout the treatment period. No patient received sodium-glucose co-transporter-2 (SGLT2) inhibitors.

Randomisation and treatment

Fostamatinib was not previously used in patients with kidney disease; therefore, the ethics committee required the trial start with randomisation between low dose fostamatinib (100 mg bid), n=26 (intended to recruit 25 patients) and placebo (n=13) (**cohort 1, see Supplementary Figure S1**). After initial assessment of safety, the Safety Committee permitted recruitment of cohort 2, receiving higher dose fostamatinib (150 mg bid), n=25, and placebo (n=12).

Despite randomisation, baseline clinical features differed among the groups. The fostamatinib 150 mg group had lower median eGFR and more segmental glomerulosclerosis than the placebo and fostamatinib 100 mg groups. The median eGFRs were 51, 50 and 35 ml/min/1.73 m^2 in the placebo, fostamatinib 100 mg, and fostamatinib 150 mg groups, respectively (**Table 1**). The baseline urine protein/creatinine ratios (UPCR) were 1272 mg/g, 1828 mg/g, and 1878 mg/g in the placebo, fostamatinib 100 mg, and fostamatinib 150 mg groups respectively (**Table 1**). Median segmental

glomerulosclerosis scores were 13, 13.6 and 25 % in the placebo, fostamatinib 100 mg, and fostamatinib 150 mg groups respectively (**Table 2**) (NS***).***

Completion of treatment to 24 weeks

Study drug compliance was high across treatment groups (median 94.9% in the placebo group and 92.9% in the fostamatinib groups), with median exposure 169 days in the placebo group and 169 days in the fostamatinib groups.

For the placebo group, 24 of 25 patients completed treatment, 1 subject lost to follow-up. For the fostamatinib 100 mg group, 24 patients completed treatment, and 2 patients discontinued treatment with adverse events (diarrhea and pancreatitis) (**Supplementary Figure S2**). For the fostamatinib 150 mg group, 20 patients completed treatment, one subject withdrew (personal reasons), and 4 patients discontinued because of adverse events, (anaemia (1), raised liver enzymes (2) peptic ulcer perforation (1)) (**Supplementary Figure S2**).

Effect of treatment

Proteinuria

At 24 weeks, the absolute reduction in proteinuria (UPCR) averaged 177.4 mg/g (+/-342.4) in the placebo group, 577.2 mg/g (+/-335.7) in the fostamatinib 100 mg group and 157.5 mg/g (+/-345.6) in the 150 mg group in the intention-to-treat (ITT) analysis, 9%, 25%, and 8% reductions respectively. Differences between treatment groups were not statistically significant (**Figure 1**). The longitudinal changes in median UPCR are shown in **Figure 2**. Reductions of ≥30% or ≥50% in UPCR from baseline occurred in 33% and 17% respectively in patients of the placebo group, 38% and 33% respectively in the fostamatinib 100 mg group, and 50% and 24% respectively in the fostamatinib 150 mg group; not statistically significant.

In a pre-specified subgroup analysis of patients with baseline UPCR of more than 1000 mg/g, a dose-dependent trend for reduction in median proteinuria from baseline by 14%, 27% and 36% in the placebo, fostamatinib 100 mg group and fostamatinib 150 mg group respectively was seen at 24 weeks (p=NS) (**Figure 3**). At 24 weeks, reduction of ≥30% in UPCR from baseline occurred in 29% of patients in the placebo group, 50% of patients in the fostamatinib 100 mg group and 53% of patients in the fostamatinib

150 mg group; again not statistically significant. In the longitudinal analysis, there was a transient rise in UPCR in patients treated with fostamatinib 150 mg during weeks 1-4 of treatment. Dose dependent trends of reduction in proteinuria were seen at weeks 18 and 24 (**Figure 4**).

Post-hoc modelling of the change in UPCR from baseline to week 24 in the ITT population did not detect a significant treatment effect after including baseline clinical features of UPCR, eGFR, endothelial hypercellularity and segmental glomerulosclerosis as covariates **(Supplementary Table S3)**.

Hematuria

Hematuria results (dipstick test) showed trends in favour of fostamatinib compared with placebo. At week 12, 36% of patients in the fostamatinib 150 mg group had decreased hematuria compared with fostamatinib 100 mg (16%) or placebo groups (16%). At week 24, both fostamatinib groups had decreasing hematuria (29% fostamatinib 150 mg; 33% fostamatinib 100 mg) compared with the placebo group (21%) (NS) (**Table 3**).

Kidney function

Baseline eGFR was lower in the fostamatinib 150 mg group than in the placebo ($p<0.05$, see **Table 1**). Kidney function for all 3 groups of patients remained stable during the 24-week treatment period with no significant changes (**Figure 5**).

Renal histopathology

All patients had baseline kidney biopsies diagnostic of IgAN (**Supplementary Figure S1**) and 39 patients agreed to repeat biopsy as an optional part of the study.

Features of mesangial and/or endocapillary hypercellularity were detected in all baseline biopsies. Formal application of the Oxford Classification of IgAN (MEST-C) were not applicable in 16 biopsies because the number of glomeruli were less than 8. In 27 patients (11 placebo group, 13 fostamatinib 100 mg group, 3 fostamatinib 150 mg group), both pre- and post-treatment biopsies were sufficient for Oxford classification assessment. There were slight improvements in mean mesangial score in the fostamatinib 150 mg group (-0.3) and the fostamatinib 100 mg group (-0.2), in comparison to the

placebo group (-0.1), (p=NS). There were no differences between groups in changes of other histological features (**Table 4**).

All baseline, kidney biopsies contained SYK+ and CD68+ cells. SYK+ or CD68+ cells were enumerated when the kidney biopsy contained at least 8 glomerular cross sections. Due to the limited amount of material available for immunohistochemistry, serial comparison between baseline and post-treatment of biopsy was inconclusive. There was a trend for reduced CD68+ cells in the glomeruli in the 100 mg group (-5.3) in comparison to placebo (0). There were no matched samples for the fostamatinib 150 mg group. (**Supplementary Tables S1 and S2**).

Further analysis in all repeat biopsies, even those with fewer than 8 glomeruli, was undertaken, due to the valuable nature of this kidney tissue. Patients treated with fostamatinib 100 mg bid showed reduction (-1.5) in the number of SYK+ glomerular cells at 24 weeks from baseline (in comparison to increase of 1.7 SYK+ cells/glomerulus in placebo treated patients, $p<0.05$) (**Supplementary Table S4**). Similar trends were seen, with a significant reduction in mesangial hypercellularity in the fostamatinib 100 mg group, supportive of SYK inhibition (**Supplementary Tables S5-S7**).

Safety and adverse events

Serious adverse events (SAE) were reported in 2 patients per group (**Table 5**), with two deemed treatment-related. One subject in the fostamatinib 100 mg group developed abdominal pain and raised serum amylase, the trial medication was stopped, and findings resolved. One diabetic patient (fostamatinib 150 mg) developed abdominal pain and raised liver enzymes during the clinical trial. Fostamatinib was stopped, yet this patient deteriorated with a perforated peptic ulcer and peritonitis and subsequently died (**Table 5**). This event was reviewed and compared with literature and concluded not related to fostamatinib.

Diarrhea was equal among groups: placebo (7/25 patients), fostamatinib 100 mg (9/26 patients), fostamatinib 150 mg (7/25 patients). Hepatic enzyme elevations were seen in placebo (1/25), fostamatinib 100 mg (4/26) and fostamatinib 150 mg (4/25) groups (**Table 6**) and returned to normal with observation or stopping fostamatinib, except in the patient with perforated ulcer. Overall, results suggest no difference in clinically important liver function test abnormalities between the 2 fostamatinib

dose groups. Importantly, no subject had laboratory criteria meeting Hy's Law (i.e., ALT or AST > 3 × ULN and bilirubin > 2 × ULN and alkaline phosphatase < 2 × ULN). Some patients had transient increases in liver enzymes which improved without interruption of the trial medication.

Hypertension progressed during the trial in placebo (2/25), fostamatinib 100 mg (4/26) and fostamatinib 150 mg (4/25) patients. The dose of ACEi or ARBs were kept the same throughout the trial. Hypertension was treated by addition, or an increase in dose, of non-RASi antihypertensive medication.

Upper respiratory tract infections were more frequent in the placebo group (5/25) in comparison with the fostamatinib 100 mg group (1/26) and fostamatinib 150 mg group (2/25). Urinary tract infections were seen in the placebo group (1/25), 100 mg group (1/26) and 150 mg group (3/25) (**Table 6**).

Overall, fostamatinib was well tolerated by the majority of patients. One patient (fostamatinib 100 mg) discontinued treatment due to diarrhea (**Table 5**).

DISCUSSION

This is the first report of a SYK inhibitor investigated in a clinical trial of kidney disease. Prior research findings strongly support a role for SYK in IgAN [11,15]. At baseline, all patients had persistent, significant proteinuria despite being on stable, maximal tolerable doses of ACEI or ARB for at least 12 weeks. Baseline kidney biopsies showed diagnostic features of IgAN together with mesangial and/or endocapillary hypercellularity. Importantly, CD68+ monocytes/macrophages and SYK+ cells were detected in the glomeruli in all baseline kidney biopsies. Previous work has shown an increased number of SYK+ cells in kidney biopsies of patients with IgAN in comparison to less inflammatory kidney diseases, including thin basement membrane disease and minimal change disease[11-13,15,16]. In the limited number of serial kidney biopsies, treatment with fostamatinib 100 mg bid trended to decrease glomerular macrophages from baseline to week 24 treatment, (supplementary Table S2). Kashem and colleagues found that increase glomerular FcαRI(CD89) mRNA was associated with expression of mRNA for inflammatory cytokines in the kidney biopsies from patients with IgA nephropathy[17]. Kanamaru and colleagues showed that serum from patients activates cell lines co-expressing FcαRI and FcɣR, with expression of multiple phosphoproteins, including pp72 and production of TNFα[18]. Furthermore, FcαRI/FcɣR mediated inflammatory cytokine production, (TNFα and chemokine CCL2),

and glomerular macrophage infiltration in a transgenic model of IgA nephropathy. Previously, we found that binding of IgA from patients with IgA nephropathy led to expression of phosphorylated SYK (72 kD) and inflammation cytokine production. including chemokine CCL2. Therefore, our limited biopsy results suggest that treatment of SYK inhibitor may abrogate some effects of IgA complex activation of receptor-mediated chemokine production and monocyte/macrophage infiltration of the glomeruli.

Treatment with fostamatinib or placebo was safely added to standard of care therapy. There were differences at baseline among the treatment groups. In particular, the 150 mg group had lower eGFR and a higher proportion of glomerular segmental sclerosis. These differences are likely to be related to stratification issues due to sequential design of the clinical trial, required by the ethics committee. In the ITT analysis, there were no significant differences in proteinuria following 24 weeks treatment with fostamatinib in comparison to placebo. To evaluate potential therapeutic effect of fostamatinib in patients at high progression risk, we included a pre-specified analysis of patients with baseline UPCR >1000 mg/g. Here, we detected a dose dependent trend in reduction of proteinuria, although there were only 14 to 16 patients in the subgroups. eGFR remained stable in placebo and fostamatinib treatment groups. To further explore the potential contribution of variation in baseline UPCR, eGFR, endocapillary hypercellularity and segmental glomerulosclerosis, analyses of covariance were carried out (**Supplementary Table S3**), but it did not detect significant effects, possibly limited by sample size.

An important effort of this study was to utilize the potential of evaluating for pathologic changes in clinical trials by repeat biopsy. Demonstrating changes in the actual kidney disease activity and injury is an important goal for clinical studies. Thirty-nine patients underwent post-treatment kidney biopsy. Unfortunately, several of these kidney biopsies did not contain a minimum of 8 glomeruli making them inadequate for the Oxford Classification. This resulted in a marked reduction in the number of paired kidney biopsies available for analysis. Indeed, only 3 patients had adequate paired kidney samples in the fostamatinib 150 mg group. Still, a trend in reduction of the mesangial hypercellularity score in comparison to the placebo group was demonstrated (**Table 4**). The sample numbers were, however, too few for meaningful statistical assessment. Utilizing all available tissue allowed evaluation of data with results supportive of the potential for SYK inhibition to reduce IgA induced mesangial cell proliferation [11] (**Supplementary Table S7**).

The choice of fostamatinib doses in this trial was based on experiences in treating immune thrombocytopenia (ITP).[19] We only tested a limited dose range for a short duration in this first clinical trial of fostamatinib in IgAN. Performing a trial of longer duration and/or higher dose could be considered in the future. Fostamatinib was generally well tolerated by patients, with no differences in infection rates between fostamatinib and placebo treated patients. Known side-effects of fostamatinib including hypertension, diarrhea, and raised liver enzymes were less frequent than reported in previous experiences and not severe[20]. Likely unrelated and not reported previously, one patient on fostamatinib suffered a fatal perforated peptic ulcer.

Limitations of this study include greater variation in proteinuria due to analysis of untimed UPCR, rather than from a timed collection, which is generally regarded as more accurate.[21] Furthermore, the 24-week treatment may have been too short to impact maximally on disease activity, particularly as IgAN is a slowly progressive chronic disease. We noticed that in some patients, there were transient increases in proteinuria after initiation of fostamatinib, particularly in the 150 mg fostamatinib group (**Figures 2 and 4**). We were unable to identify a specific cause for this and by 12 weeks of treatment, the UPCR had fallen below baseline. Our assessment of efficacy is also limited by the disproportionate impact of fluctuations in protein excretion in patients with low levels of proteinuria.

In conclusion, this was the first clinical trial of a SYK inhibitor in kidney disease. There was a trend for a dose dependent reduction in proteinuria with fostamatinib in IgAN patients with baseline UPCR >1000 mg/g. Future clinical trials to assess the safety and efficacy of fostamatinib or other SYK inhibitors in IgAN should focus on patients with proteinuria >1 gram/day and evaluate fostamatinib over a longer duration. Follow up biopsies are achievable and may be of value in assessing impact of therapies on IgA nephropathy.

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AI technologies have not been used in the preparation of this manuscript.

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Data Sharing statement:

Rigel Pharmaceuticals, Inc. is committed to data transparency and will consider data sharing requests on a case-by-case basis. Additionally, Rigel Pharmaceuticals, Inc. will provide the study protocol as well as post results on ClinicalTrials.gov as required.

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Author Contributions:

FT, JB, IR, CR HC, EM, SM, MK conceived and designed the clinical trial. All the authors contributed to acquiring the data and/or played an important role in interpreting the results. FT, JT, JB, BR, IR, CR, HC, LT, CS, ST, LT, ST and RL drafted the initial manuscript. All the authors contributed to revision and approval of the manuscript.

Supplementary Materials

Supplementary Table S1. IHC Total SYK – Summary of Average Number of Cells per Glomeruli (Population of Patients with ≥8 Glomeruli)

Supplementary Table S2. IHC CD68 – Summary of Average Number of Cells per Glomeruli (Population of Patients With ≥8 Glomeruli)

Supplementary Figures:

Consort statement

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Figure legends

Figure 1. Primary Efficacy Endpoint: Mean Change From Baseline in UPCR at Week 24 (All Intent-to-Treat Patients). UPCR = Urinary Protein to Creatinine Ratio; N = Number of patients in the intent-to-treat population 25, 26, 25 and 51 in the placebo, fostamatinib 150 mg bid, fostamatinib 150 mg bid and fostamatinib any doses (100 mg or 150 mg bid) groups respectively. Reported means and standard errors were adjusted for baseline UPCR (mg/g) and baseline endocapillary hypercellularity (absent/present) using an analysis of covariance model. *p=NS for comparison with placebo.

Figure 2. Median Percent Change From Baseline in UPCR Over Time (All Intent-to-Treat Patients). UPCR = Urinary Protein to Creatinine Ratio.

Figure 3. Median Percent Change From Baseline in UPCR at Week 24 (All Intent-to-Treat Patients With Baseline UPCR > 1000 mg/g). UPCR=Urinary Protein to Creatinine ratio. *p=NS for comparison with placebo.

Figure 4. Median Percent Change From Baseline in UPCR Over Time (All Intent-to-Treat Patients With Baseline UPCR > 1000 mg/g). UPCR = Urinary Protein to Creatinine Ratio.

Figure 5. Median Percent Change From Baseline in eGFR Over Time (All Intent-to-Treat Patients With Baseline UPCR >1000 mg/g). UPCR = Urinary Protein to Creatinine Ratio.

Table 1. Baseline Demographics and Characteristics (Safety Population)

	Placebo (N=25)	**Fostamatinib 100 mg bid (N=26)**	**Fostamatinib 150 mg bid (N=25)**
Age at Baseline (years) [median (range)]	40 (20, 59)	42 (19, 67)	41 (20, 68)
≥65 years [n (%)]	0	1 (4%)	3 (12%)
Female [n (%)]	12 (48%)	12 (46%)	12 (48%)
Race			
White [n (%)]	19 (76%)	19 (73%)	13 (52%)
Asian [n (%)]	6 (24%)	6 (23%)	11 (44%)
Body Mass Index (kg/m^2) [median (range)]	27.7 (21.3, 39.5)	27.2 (19.0, 47.6)	25.1 (18.7, 45.3)
Duration of IgAN (years) [median (range)]	3.4 (0.2, 25)	2.5 (0.2, 14)	3.2 (0.1, 18)
History of Type 2 Diabetes [n (%)]	1 (4%)	3 (12%)	2 (8%)
ACEi/ARB Use (months) [median (range)]	9.1 (3, 192)	8.9 (3, 128)	8.5 (3, 67)
>6 months [n (%)]	15 (60%)	15 (58%)	15 (60%)
None [n (%)]	1 (4%)	1 (4%)	1 (4%)
UPCR (mg/g) at Baseline [median (range)]	1272 (525, 9938)	1828 (387, 16259)	1878 (664, 4076)
>1000 mg/g [n (%)]	15 (60%)	17 (65%)	18 (72%)
>2000 mg/g [n (%)]	7 (28%)	11 (42%)	11 (44%)
>3500 mg/g [n (%)]	3 (12%)	3 (12%)	1 (4%)
Serum creatinine (µmol/L) [median (range)]	106 (64, 239)	124 (60, 290)	157 (55, 309)*
eGFR (mL/min/1.73 m²) [median (range)]	51 (25, 104)	50 (20, 109)	35 (18, 103)*

eGFR > 60 mL/min/1.73 m² [n (%)]	12 (48%)	10 (38%)	6 (24%)
SBP [median (range)]	118 (100, 125)	117 (102, 135)	119 (97, 149)
DBP [median (range)]	78 (46, 90)	74.5 (61, 85)	76 (57, 98)

Serum creatinine concentration was higher in the Fostamatinib 150 mg bid group in comparison to the Placebo group (*<0.05). Baseline eGFR was lower in the Fostamatinib 150 mg bid group in comparison to the Placebo Group (*p<0.05).

Table 2. Renal Biopsy - Baseline Characteristics (Safety Population)

	Placebo (N=25)	**Fostamatinib 100 mg bid (N=26)**	**Fostamatinib 150 mg bid (N=25)**
Mesangial Hypercellularity [median (range)]	0.6 (0, 1.18)	0.6 (0, 1.86)	0.5 (0, 1.25)
Oxford Classification M_1 [n (%)]	12 (48%)	14 (54%)	9 (36%)
Endocapillary Hypercellularity [median (range)]	0 (0, 30)	0 (0, 37)	0 (0, 15.79)
Oxford Classification E_1 [n (%)]	9 (36%)	10 (38%)	7 (28%)
Segmental Glomerulosclerosis [median (range)]	13.0 (0, 50)	13.6 (0, 53.85)	25.0 (0, 50)
Oxford Classification S_1 [n (%)]	20 (80%)	17 (65%)	17 (68%)
Tubular Atrophy/Interstitial Fibrosis [median (range)]	24.8 (10, 50)	30 (0, 60)	30 (0, 40)
Oxford Classification T_2 [n (%)]	0	1 (4%)	0

N=Number of patients in the intent-to-treat population; UPCR = Spot Creatinine to Protein Ratio; SBP = systolic blood pressure; DBP = diastolic blood pressure; eGFR = Estimated Glomerular Filtration Rate; M_1 = > 50% of glomeruli show mesangial hypercellularity; E_1 = Any glomeruli show endocapillary hypercellularity; S_1 = present in any glomeruli; T_2 = > 50% of cortical area shows tubular atrophy or interstitial fibrosis

Table 3. Fostamatinib Treatment Reduces Hematuria at Week 12

n (%)	**Placebo (N=25)**	**Fostamatinib 100 mg bid (N=26)**	**Fostamatinib 150 mg bid (N=25)**
Decrease in Hematuria From Baseline			
Week 12	4 (16%)	4 (16%)	8 (36%)
Week 24	5 (21%)	8 (33%)	6 (29%)

Table 4. Outcomes in the Biopsy Population

	Placebo (N=11)	Fostamatinib 100 mg bid (N=13)	Fostamatinib 150 mg bid (N=3)
Mean Mesangial Hypercellularity Scores			
Pre-Treatment [mean (SD)]	0.4 (0.30)	0.8 (0.56)	0.5 (0.30)
Post-Treatment [mean (SD)]	0.4 (0.30)	0.5 (0.48)	0.3 (0.23)
Difference [LS mean (SE)]*	-0.1 (0.08)	-0.2 (0.08)	-0.3 (0.16)
Difference (95%CI)]*	-	-0.12 (-0.4, 0.1)	-0.20 (-0.6, 0.2)
Mean Endocapillary Hypercellularity Scores			
Pre-Treatment [mean (SD)]	6.9 (9.4)	8.6 (11.4)	5.3 (9.1)
Post-Treatment [mean (SD)]	1.9 (3.9)	3.6 (7.3)	3.7 (6.4)
Difference [LS mean (SE)]**	-5.4 (1.6)	-4.0 (1.5)	-3.1 (3.2)
Difference (95%CI)]**	-	(-3.2, 6.1)	(-5.1, 6.9)

N=Number of patients who had both pre-treatment and post-treatment biopsies with at least 8 evaluable glomeruli

*Reported LS means and standard errors were adjusted for baseline UPCR (mg/g) and baseline Endocapillary Hypercellularity (absent/present) using an analysis of covariance model

**Reported statistics are using analysis of covariance model with change from baseline in Endocapillary Hypercellularity % (%) as a dependent variable, treatment (Presence/Absence) as factors, and baseline Endocapillary Hypercellularity % (%) as a covariate.

Table 5. Summary of Adverse Events (AE). All Patients in Safety Population

n (%)	**Placebo (N=25)**	**Fostamatinib 100 mg bid (N=26)**	**Fostamatinib 150 mg bid (N=25)**
Treatment Emergent AEs	21 (84%)	22 (85%)	24 (96%)
Mild	15 (60%)	14 (54%)	16 (64%)
Moderate	5 (20%)	8 (31%)	5 (20%)
Severe	1 (4%)	0	3 (12%)
Serious AEs[1]	2 (8%)	2 (8%)	2 (8%)
Treatment Related AEs[2, 3]	0	1 (4%)	1 (4%)
Death[3]	0	0	1 (4%)
Treatment Interrupted or Withdrawn due to AEs[4]	1 (4%)	4 (15%)	7 (28%)
Treatment Related[2]	1 (4%)	3 (12%)	7 (28%)

[1]Serious AEs were post procedural hematuria and procedural pain in the placebo group; concussion and pancreatitis in the 100 mg group; and hepatic enzyme abnormal, peptic ulcer perforation /septic shock, and a mild post-biopsy hematoma in the 150 mg group.

[2]Treatment-related events are events deemed by investigator as probably or possibly related to study treatment.

[3]Peptic ulcer perforation was not treatment-related

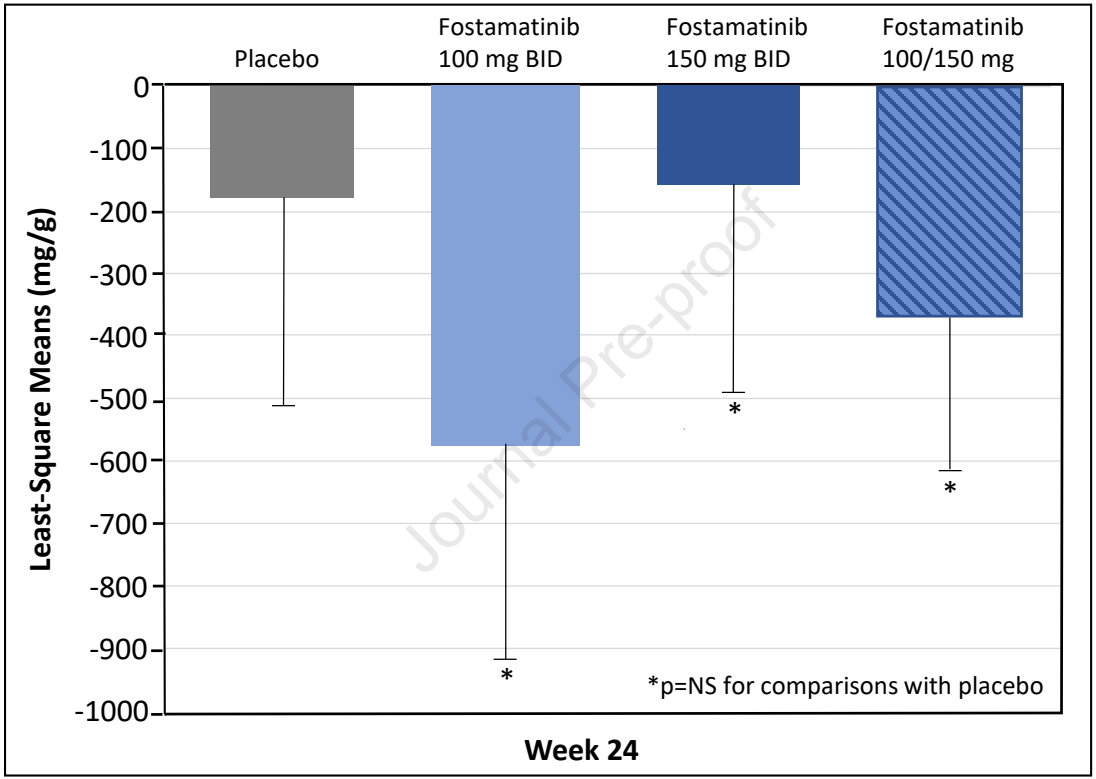
[4]Treatment was withdrawn due to diarrhea and pancreatitis in the 100 mg group and due to anemia, peptic ulcer perforation, ALT increased, and hepatic enzyme (both ALT and AST) increased in the 150 mg group.

Table 6. The Most Common Treatment-Emergent Adverse Events (AE) (>10% of Patients in Any Treatment Group) and Adverse Events Grouped by Standardised MedDRA Query (SMQ) (All Patients in the Safety Population)

n (%) of patients	Placebo (N=25)	Fostamatinib 100 mg bid (N=26)	Fostamatinib 150 mg bid (n=25)
Diarrhea	7	9	7
Nasopharyngitis	7	1	4
Nausea	3	6	5
Headache	1	3	5
Upper Respiratory Tract Infection	5	1	2
Cough	4	2	2
Muscle Spasms	0	1	4
Vomiting	1	3	4
Hypertension	1	4	3
Oropharyngeal Pain	2	2	3
Urinary Tract Infection	1	1	3
Abdominal Pain	1	3	0
Alanine Aminotransferase Increased	1	3	2
Dizziness	1	3	1
Rash	0	3	1
Non-infectious diarrhea SMQ*	7	9	7
Hepatic Disorders SMQ*	1	4	4
Hypertension SMQ*	2	4	4
Neutropenia SMQ*	0	0	0

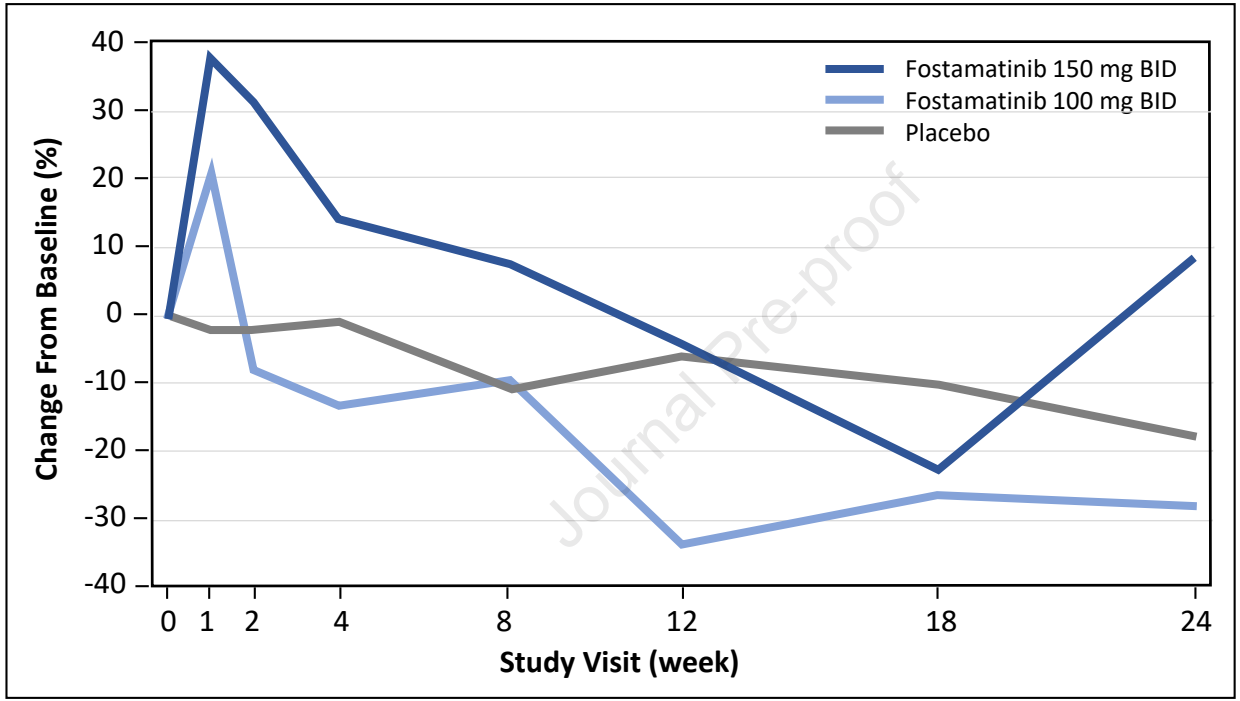
*Derived using standard MedDRA queries (SMQ) version 19.1

Figure 1. Primary Efficacy Endpoint: Mean Change From Baseline in UPCR at Week 24 (All Intent-to-Treat Patients)



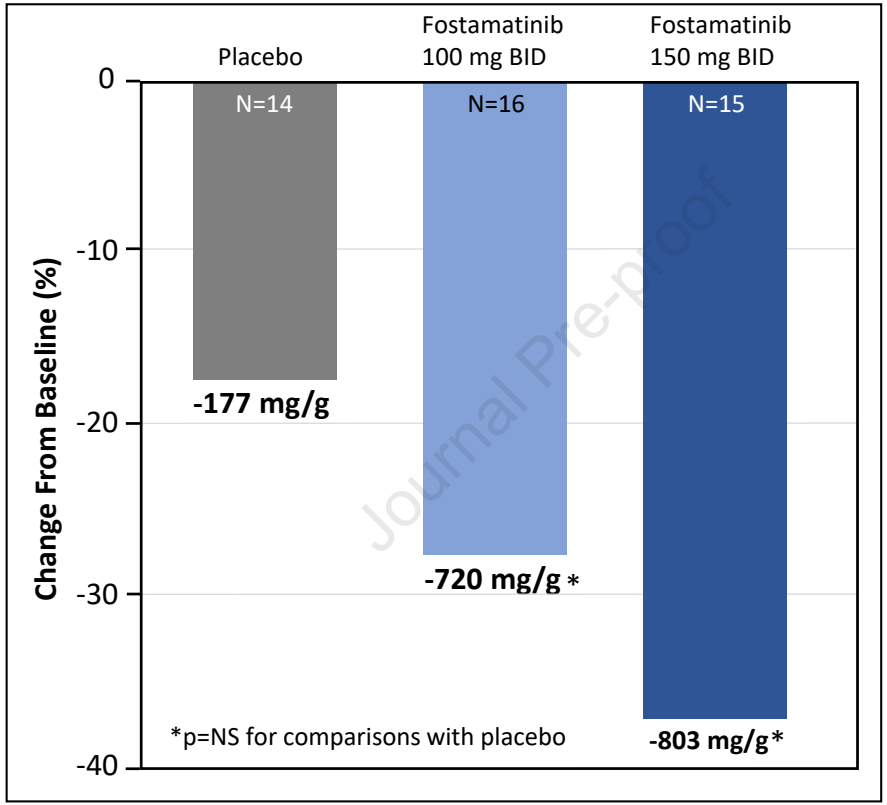
UPCR = Urinary Protein to Creatinine Ratio. Reported means and standard errors were adjusted for baseline UPCR (mg/g) and baseline endocapillary hypercellularity (absent/present) using an analysis of covariance model

Figure 2. Median Percent Change From Baseline in UPCR Over Time (All Intent-to-Treat Patients)



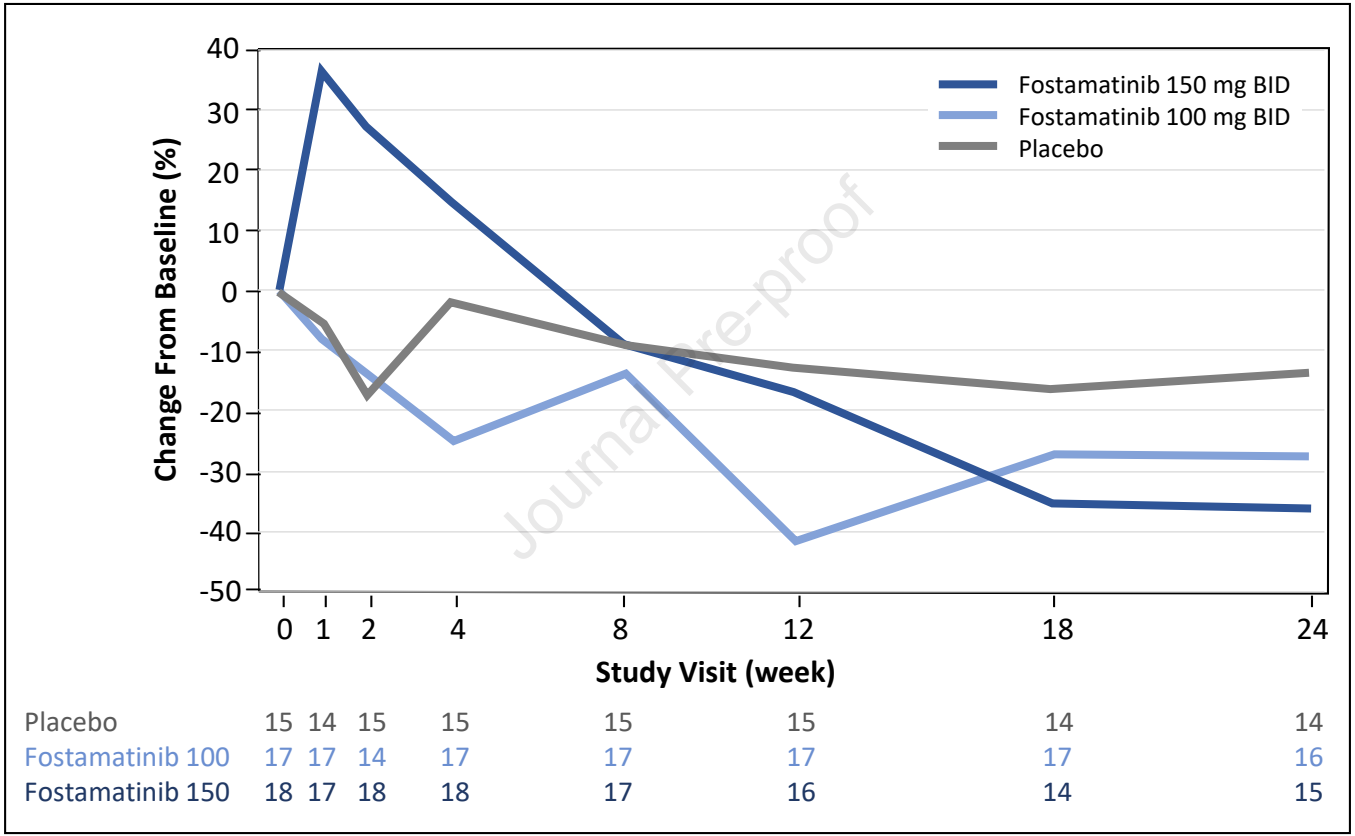
UPCR = Urinary Protein to Creatinine Ratio

Figure 3. Median Percent Change From Baseline in UPCR at Week 24 (All Intent-to-Treat Patients With Baseline UPCR > 1000 mg/g)



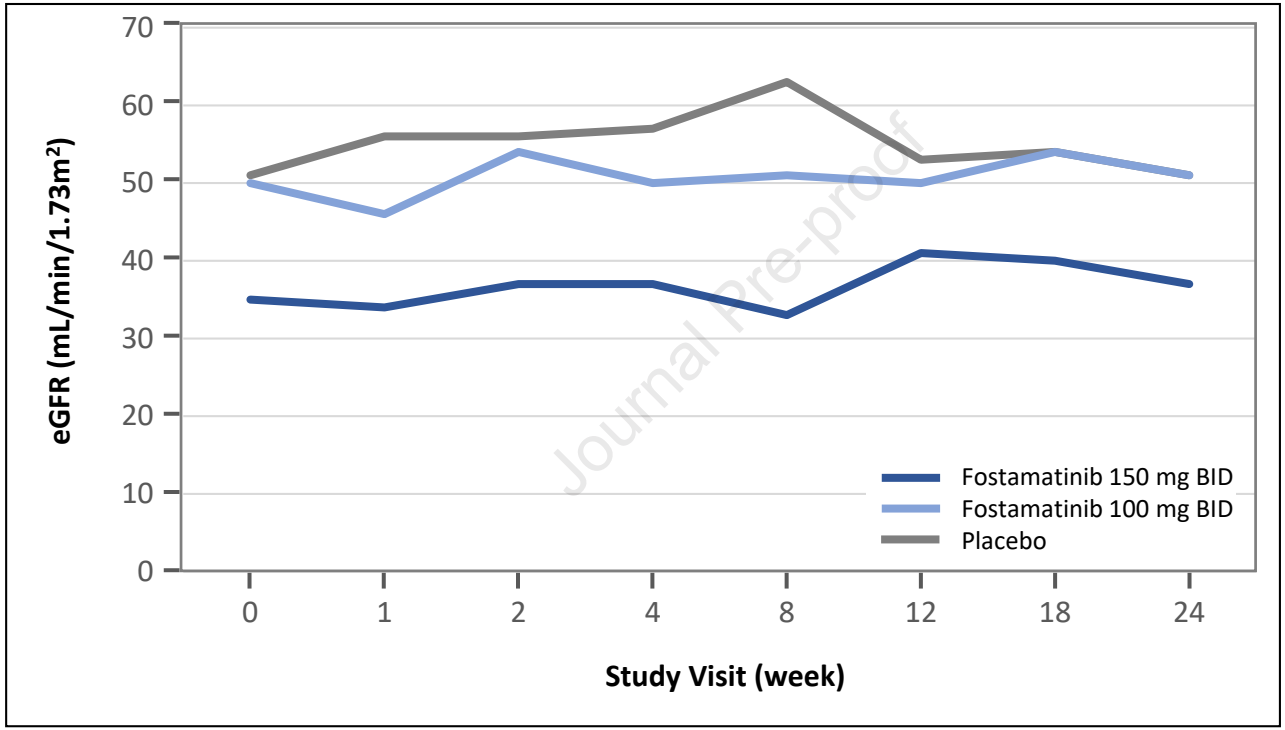
UPCR = Urinary Protein to Creatinine Ratio

Figure 4. Median Percent Change From Baseline in UPCR Over Time (All Intent-to-Treat Patients With Baseline UPCR > 1000 mg/g)



UPCR = Urinary Protein to Creatinine Ratio

Figure 5. Median Estimated Glomerular Filtration Rate (eGFR) Over Time (All Intent-to-Treat Patients)



UPCR = Urinary Protein to Creatinine Ratio