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# Urinary Protein Binding Does Not Affect Response to Furosemide in Patients with Nephrotic Syndrome

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*Abstract.* Response to loop diuretics in patients with nephrotic syndrome (NS) is subnormal. Studies in animal models of NS have suggested that binding of diuretic to urinary albumin is one of the mechanisms that may be operative in this diuretic resistance. To explore this hypothesis, 12 patients with NS were studied to determine whether displacement from urinary protein binding with sulfisoxazole would restore response to 120 mg of furosemide. The study was stopped after treating seven patients because it was clear that sulfisoxazole had no effect. Sodium excretion (mean ± SD) from furosemide alone was 239 ± 90 *versus* 240 ± 115 mEq/8 h with sulfisoxazole. Sulfisoxazole had modest effects on serum pharmacokinetics of furosemide but had no effect on either the time course of furosemide urinary excretion or overall amount excreted: 49 ± 15 mg *versus* 54 ± 12 mg for furosemide alone and furosemide plus sulfisoxazole, respectively. It is concluded that urinary protein binding of loop diuretics is not a major mechanism for the diuretic resistance of NS. In turn, strategies aimed at displacing such binding are unlikely to be clinically helpful.

Loop diuretics are the mainstays of therapy for the sodium retention of nephrotic syndrome (NS). Despite their efficacy, treatment can be vexing because substantial resistance to diuretic therapy can occur. A number of studies have attempted to define the mechanisms by which subnormal responses to diuretics occur in order to more effectively use these agents (reviewed in reference 1). It is clear that the diuretic resistance of NS is multifactorial. A possible role of hypoalbuminemia causing diminished delivery of loop diuretics to their urinary site of action has been proposed (2). In addition, consistent with the hypothesis articulated by Green and Mirkin (3,4), our laboratory has shown that loop diuretics bind to urinary albumin (5–7). This process can render inactive as much as half of the dose of a diuretic. In an animal model of a nephrotic nephron, response to furosemide can be completely restored by displacing the furosemide from albumin with sulfisoxazole (7). These animal studies raise the possibility that the same therapeutic strategy could be beneficial in patients. Sulfisoxazole as a displacing agent is attractive in a clinical setting because it is administered in high doses so that in humans it would reach the urine in substantial molar excess of the loop diuretic, optimizing the likelihood of displacement. Moreover, it has a wide therapeutic index. Therefore, we conducted the present study to determine whether sulfisoxazole could enhance response to a loop diuretic in patients with NS.

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## Materials and Methods

Seven patients with nephrotic syndrome participated in a randomized, crossover comparison of response to furosemide alone *versus* furosemide plus sulfisoxazole. Our original study design entailed treating 12 patients, because we calculated that this number would provide 80% power to detect an effect of sulfisoxazole as large as 1.2 times the SD using a two-sided test at the 5% level of significance. We stopped the study when it became clear that there was no effect. Patient characteristics are listed in Table 1. The only exclusion criterion was concomitant disease that might independently affect diuretic response including renal insufficiency. As such, renal function in these patients was reasonably preserved with values of creatinine clearance that ranged from 54 to 140 ml/min.

### *Protocol*

Patients were admitted to the General Clinical Research Center (GCRC) and were begun on a metabolic diet containing 30 mEq of sodium and 3 L of fluid per day. This sodium restriction allowed discontinuation of all diuretics throughout the remainder of the study. All other medications were continued with the exception of nonsteroidal anti-inflammatory drugs, which were discontinued 2 wk before admission. Low dose aspirin (≤325 mg/d) was continued in both arms of the study in one patient. This dose of aspirin is unlikely to affect renal function or response to diuretics. Patients attained sodium balance over a period of 3 to 5 d as documented by stable body weight and 24-h urinary sodium excretion. Body weight (mean ± SD) prior to furosemide alone was 99 ± 25 kg; before furosemide plus sulfisoxazole, it was also 99 ± 25 kg.

Once balance was attained, patients were randomized to receive furosemide alone or furosemide plus sulfisoxazole. When the patient was randomized to the latter, they received three separate 2-g doses of sulfisoxazole 8 h apart so that the last dose occurred the morning they were to receive furosemide. In this way, there were large amounts of sulfisoxazole in the urine throughout the time they received furosemide (vide infra). The molar ratio of sulfisoxazole to furosemide averaged 565 throughout the time of evaluation. Our previous animal studies in which displacement of furosemide from albumin occurred

*Table 1.* Patient characteristics[a]

| Patient | Gender | Age (yr) | Etiology of Nephrotic Syndrome | 24-Hour Protein Excretion (g) | Concomitant Medications | Concomitant Diagnoses |
|---|---|---|---|---|---|---|
| 1 | F | 57 | Idiopathic | 3.5 | Furosemide, fluvastatin, albuterol, nifedipine, beclomethasone, fosinopril, ipratropium, ASA, isosorbide dinitrate, and theophylline | HTN, COPD, osteoarthritis, sinusitis |
| 2 | F | 56 | Membranous glomerulonephritis | 10.7 | Bumetanide, warfarin, and isradipine | Renal and pulmonary embolism, HTN, CHF |
| 3 | M | 57 | Focal glomerulosclerosis | 7.2 | Terazosin, lovastatin, theophylline, glypizide, famotidine, and losartan | Type 2 DM, HTN, asthma |
| 4 | F | 24 | Idiopathic | 9.8 | Furosemide | HTN, eclampsia |
| 5 | M | 37 | Lupus nephritis | 7.2 | Cyclophosphamide, prednisone, lisinopril, and hydroxychloroquine | SLE, HTN |
| 6 | M | 48 | Diabetic nephropathy | 15.2 | Furosemide, spironolactone, lisinopril, insulin, and metolazone | Type 2 DM, HTN |
| 7 | M | 38 | Diabetic nephropathy | 6.8 | Lisinopril, cisapride, furosemide, insulin, and felodipine | Type 1 DM |

[a] ASA, acetylsalicylic acid; HTN, hypertension; COPD, chronic obstructive pulmonary disease; CHF, congestive heart failure; DM, diabetes mellitus; SLE, systemic lupus erythematosus.

sufficient to fully restore response used a similar molar ratio of sulfisoxazole to furosemide (6). Thus, sufficient amounts of sulfisoxazole reached the urine to adequately test our hypothesis. Moreover, these doses were as high as could be contemplated for clinical use.

On the day of the study, patients skipped breakfast but were allowed to eat lunch. A baseline blood sample was collected, and an antecedent quantitative urine collection was completed. Patients then drank 10 ml/kg distilled $H_2O$ to ensure the ability to collect frequent urine samples. Another baseline urine collection was then obtained over 1 h, after which 120 mg of furosemide was infused intravenously over 30 min. Blood and urine samples were then collected at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, and 8 h after the start of the furosemide infusion. Patients then reattained sodium balance as documented by body weight and 24-h sodium excretion, after which they underwent the alternative arm of the study.

## *Analyses*

Serum and urine samples were assayed for sodium and potassium using a flame photometer (IL-940; Instrumentation Laboratories). The inter-day precision and accuracy for the determination of sodium and potassium was less than 5 and 10%, respectively. Serum and urine creatinine concentrations were determined with a Technicon Autoanalyzer II using the Jaffe reaction. The assay was used to routinely determine serum and urine creatinine concentrations between 0.5 and 5 mg% and 1 and 15 mg%, respectively. The inter-day precision and accuracy for serum and urine was less than 10%.

Urine and serum furosemide concentrations were determined using HPLC with fluorescence detection as described previously with some modification (8–10). After the addition of internal standard (500 ng of metolazone), 0.5 ml of serum was deproteinated with 1.0 ml of acetonitrile, mixed vigorously, and centrifuged at 2800 rpm for 10 min. The supernatant was transferred to a clean test-tube, evaporated to dryness, and reconstituted with 150 $\mu$l of mobile phase (35:65 acetonitrile: 50 mM sodium acetate, pH 3.6), of which a portion was injected into an HPLC. Furosemide and the internal standard were separated using a 5 $\mu$m Beckman Ultrasphere™ (25 cm $\times$ 4.6 mm inner diameter) C-18 column equipped with a 2-cm guard column. The mobile phase was delivered at a rate of 1 ml/min, and the eluate was monitored at an emission wavelength of 378 nm after excitation at a wavelength of 234 nm using a Hewlett Packard 1146A fluorescence detector. This procedure was used to assay serum furosemide concentrations between 0.01 $\mu$g/ml and 5.0 $\mu$g/ml. Inter-day precision and accuracy was less than 13 and 4% at furosemide concentrations of 0.8 and 16 $\mu$g/ml, respectively. Urine furosemide concentrations were determined as described above except that the volume of urine used was 0.25 ml; 2.0 ml of acetonitrile and 1500 ng of internal standard were added. Inter-day precision and accuracy were similar to the serum assay.

Urine sulfisoxazole concentrations were determined using HPLC with ultraviolet detection at 270 nm. To 0.25 ml of urine, 2000 ng of sulfaphenazole (internal standard) was added followed by 2.0 ml of acetonitrile. The samples were mixed vigorously and centrifuged (2800 rpm for 10 min), and the supernatant was transferred to a clean test tube and evaporated. The residue was reconstituted with mobile phase (27.5%:62.5% acetonitrile:50 mM ammonium acetate, pH 5.0), of which a portion was injected into an HPLC. Sulfisoxazole and the internal standard were separated using a 5 $\mu$m Luna™ (25 cm $\times$ 4.6 mm inner diameter; Phenomenex, Torrance, CA) C-18 column equipped with a 2-cm guard column. The mobile phase was delivered at a rate of 0.95 ml/min, and the eluate was monitored at 270 nm using an ultraviolet detector. This procedure was used to assay urinary sulfisoxazole concentrations between 0.3 and 20.0 $\mu$g/ml. Inter-day precision and accuracy were less than 10 and 2% at sulfisoxazole concentrations of 2.8 and 40 $\mu$g/ml, respectively.

## *Statistical Analyses*

Response was analyzed in several ways. Total sodium excretion was compared using a paired *t* test. Sensitivity of the nephron to

furosemide was determined, as described previously (8–10), by relating urinary furosemide to sodium excretion rate. This analysis accounts for any changes that might occur in amounts of diuretic reaching the site of action. Because sulfisoxazole could also affect the active secretion of furosemide into the urine, we also assessed the serum pharmacokinetics of furosemide. Half-life and the terminal elimination rate constant were determined from the log-linear phase of furosemide elimination. Area under the serum concentration *versus* time curve (AUC) was determined by the trapezoidal rule with extrapolation to infinity from the last measured serum concentration using the terminal elimination rate constant. Clearance was calculated as dose/AUC.

## Results

### *Furosemide Pharmacokinetics*

Figure 1 depicts the serum concentrations of furosemide *versus* time, and Table 2 lists derived pharmacokinetic parameters. Sulfisoxazole would be predicted to potentially have two effects on serum pharmacokinetics of furosemide. First, it might displace furosemide from serum albumin, which would result in decreased AUC and increases in clearance and volume of distribution as was observed in our study. Second, sulfisoxazole might compete with furosemide for active secretion via the organic acid secretory pathway of the proximal tubule, which would decrease urinary furosemide excretion. The latter did not occur (Table 2). The time course of diuretic excretion has been shown in previous studies to be an important independent determinant of response over and above the absolute amount of diuretic in the urine (11). Figure 2 shows that sulfisoxazole had no effect on furosemide excretion rate. Thus, although sulfisoxazole caused modest changes in serum furosemide pharmacokinetics, likely by displacement from binding to serum albumin, it had no effect on either total furosemide excretion (Table 2) or the time course of excretion (Figure 2).

### *Response to Furosemide*

Response to furosemide was assessed in three ways. Table 3 presents the total amounts of sodium and potassium excreted over 8 h indicating no effect of sulfisoxazole. Figure 3 depicts the time course of sodium excretion wherein sulfisoxazole again had no effect. Finally, we and others have shown that the most precise way to assess the pharmacodynamics of a loop diuretic is to relate urinary excretion rate of the diuretic, which reflects amounts reaching the site of action, to response (8–10). Figure 4 shows that this relationship is not changed by sulfisoxazole. Overall, then, sulfisoxazole had no effect on either delivery of furosemide to its site of action (Table 2 and Figure 2) or on sensitivity of the nephron to furosemide (Table 3 and Figures 3 and 4).

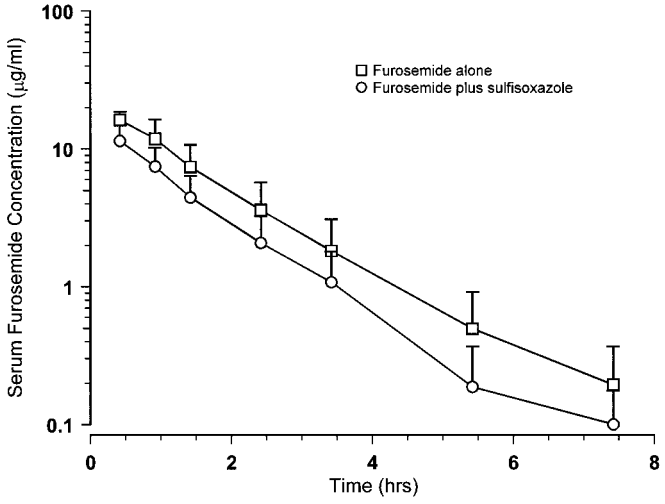


*Figure 1.* Serum concentration of furosemide over time with and without sulfisoxazole.

## Discussion

Patients with NS are often difficult to manage in terms of their volume status because even large doses of potent diuretics and combinations of diuretics have diminished effects. Several potential mechanisms for this diuretic resistance have been identified and include decreased delivery of diuretic to the urinary site of action due to renal insufficiency (12–14) or hypoalbuminemia (2), binding of diuretic to urinary albumin (1,2,5–7), or decreased sensitivity of the nephron to diuretic (15–20). The quantitative contribution of each potential mechanism has not been delineated. Thus, clinicians have been frustrated by the lack of sufficient information to develop therapeutic strategies targeted toward the pathophysiology of diuretic resistance. In the current study, we specifically addressed the importance of binding of loop diuretic to urinary albumin. Previous studies from our laboratory in an animal model of NS indicated that such binding accounted for the majority, if not all, of the subnormal diuretic response in NS (5–7). As such, we expected a salutary effect of sulfisoxazole in the current study. We found a difference in urinary sodium excretion with and without sulfisoxazole of 0.43 mEq with an SD of 55.1 mEq. From these data, we would need to recruit $1.29 \times 10^5$ patients to show a difference with and without sulfisoxazole on natriuretic response using a two-tailed test with 80% power and at the 5% significance level. Such a study is obviously not feasible or biologically relevant. This analysis also supports that we did not have an underpowered study. These data thereby indicate that in patients with NS, other mechanisms for diuretic resistance are operative and that clinical strategies to displace loop diuretics from urinary protein binding are not indicated.

Numerous studies have shown that loop diuretics must reach the urine to exert their effects, since they inhibit the $Na^+$-$K^+$-$2Cl^-$ transporter from the lumen side of the nephron (1). In patients with NS and concomitant decreases in renal function, less diuretic reaches the urine; however, this problem can be overcome by administering sufficiently large doses of diuretic to attain effective amounts in the urine (1).

Another putative mechanism for decreased delivery of diuretic into the urine in patients with NS occurs through the effects of hypoalbuminemia (2). The high serum protein binding (>95%) of loop diuretics causes these diuretics to have a small volume of distribution and remain in the plasma compartment (as opposed to distributing widely into tissues), where they are delivered to proximal tubular secretory sites allowing

*Table 2.* Effect of sulfisoxazole on the pharmacokinetics of furosemide in seven patients with nephrotic syndrome[a]

| Group | Subject | | | | | | | Mean ± SD |
|---|---|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | |
| Furosemide alone | | | | | | | | |
| $t_{½}$ (h) | 1.4 | 1.0 | 1.5 | 1.2 | 0.6 | 0.9 | 1.1 | 1.1 ± 0.3 |
| $V_d$ (L) | 7.0 | 10.9 | 11.0 | 5.1 | 6.1 | 9.4 | 6.3 | 8.0 ± 2.4 |
| CL (L/h) | 3.6 | 7.4 | 5.1 | 2.9 | 6.8 | 7.0 | 4.0 | 5.2 ± 1.8 |
| AUC (μg/L per h) | 33.6 | 16.2 | 23.6 | 41.8 | 17.8 | 17.3 | 30.4 | 25.8 ± 9.8 |
| furosemide excretion (mg) | 33.5 | 54.0 | 30.9 | 59.9 | 57.2 | 73.5 | 32.3 | 48.8 ± 15.2 |
| Furosemide + sulfisoxazole | | | | | | | | |
| $t_{½}$ (h) | 0.9 | 0.8 | 1.0 | 1.1 | 0.7 | 0.9 | 1.1 | 0.9 ± 0.1 |
| $V_d$ (L) | 8.4 | 11.8 | 17.0 | 7.6 | 11.6 | 13.0 | 7.9 | 11.0 ± 3.4[b] |
| CL (L/h) | 6.4 | 10.9 | 11.4 | 4.7 | 10.9 | 9.8 | 5.2 | 8.5 ± 2.9[b] |
| AUC (μg/L per h) | 18.8 | 11.0 | 10.6 | 25.4 | 11.0 | 12.2 | 23.2 | 16.0 ± 6.4[b] |
| furosemide excretion (mg) | 46.1 | 47.2 | 40.0 | 57.7 | 54.2 | 76.0 | 35.9 | 54.0 ± 12.3 |

[a] $t_{½}$, half-life; $V_d$, volume of distribution; CL, clearance; AUC, area under the serum concentration *versus* time curve.
[b] $P < 0.05$, significantly different from furosemide alone.

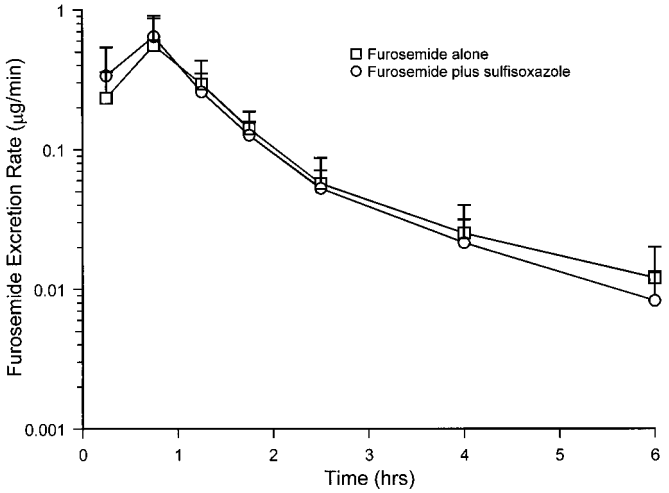


*Figure 2.* Urinary excretion rate of furosemide with and without sulfisoxazole.

their access to the urine. In hypoalbuminemia, it has been postulated that insufficient amounts of diuretic are "trapped" in the plasma with subsequently diminished delivery of diuretic to the urinary site of action (2). In turn, this pathophysiology can be reversed by administering albumin and loop diuretics intravenously. Although our study did not address this potential mechanism of diuretic resistance directly, it is clear from the pharmacokinetic data that ample furosemide reached the urine in these patients with NS (Table 2 and Figure 2). Moreover, although sulfisoxazole increased the volume of distribution of furosemide substantially, this effect did not decrease delivery of furosemide into the urine (Table 2). Thus, data from our study extend those of other recent studies that refute the importance of hypoalbuminemia as a cause of decreased delivery of loop diuretics to their site of action (21–23).

Green and Mirkin originally suggested from whole animal studies that binding of furosemide to urinary albumin and thereby rendering it inactive might account for diminished diuretic response in NS (3,4). Studies in our laboratory of rats using *in vivo* microperfusion showed that intratubular albumin in concentrations reflective of those occurring in clinical NS substantially diminished response to furosemide, that this affect was specific for albumin, and that response could be completely restored by displacement from binding (5–7). One of the drugs used to displace furosemide in these studies was sulfisoxazole. It was purposefully examined because of its potential for clinical use, in which one would need a displacing drug that reached the urine in substantial molar excess to furosemide so that displacement would be assured, coupled with the fact that sulfisoxazole has a wide margin of safety.

Because of the clear effects of sulfisoxazole in our animal model, we expected a beneficial effect in our patient study. The lack of effect we observed has several possible explanations. First, sulfisoxazole could potentially compete for proximal tubular secretion of furosemide so that less diuretic reached the site of action. Data in Table 2 and Figure 2 confirm that this did not occur. Second, the dose of sulfisoxazole could have been too low so that its concentration in urine was not sufficient to displace furosemide. Our measurements of urinary sulfisoxazole relative to furosemide indicate that the doses resulted in a large molar excess of the former (500-fold). Importantly, this ratio is comparable to that which we showed from previous studies to be sufficient to displace furosemide from binding to albumin (5–7). Third, if the dose of furosemide chosen were so large that it caused a maximal response without sulfisoxazole, then displacement would not have had an effect. Data shown in Figure 4 confirm that this was not the case.

We conclude therefore that our results are explained by the fact that mechanisms of diuretic resistance other than that which we examined are quantitatively more important. As such, previous studies in both animal models (24) and in humans (25–28) have shown that there is tubular resistance to

*Table 3.* Effect of sulfisoxazole on the response to furosemide in 7 patients with nephrotic syndrome

| Group | Subject | | | | | | | Mean ± SD |
|---|---|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | |
| Furosemide alone | | | | | | | | |
| sodium excretion (mEq) | 185 | 268 | 146 | 197 | 339 | 377 | 164 | 239 ± 90 |
| potassium excretion (mEq) | 27.4 | 34.1 | 16.6 | 32.4 | 40.6 | 30.2 | 26.2 | 29.6 ± 6.9 |
| Furosemide + sulfisoxazole | | | | | | | | |
| sodium excretion (mEq) | 186 | 183 | 146 | 246 | 311 | 463 | 144 | 240 ± 115 |
| potassium excretion (mEq) | 28.2 | 28.4 | 25.6 | 39.2 | 35.9 | 44.9 | 25.8 | 32.6 ± 6.9 |

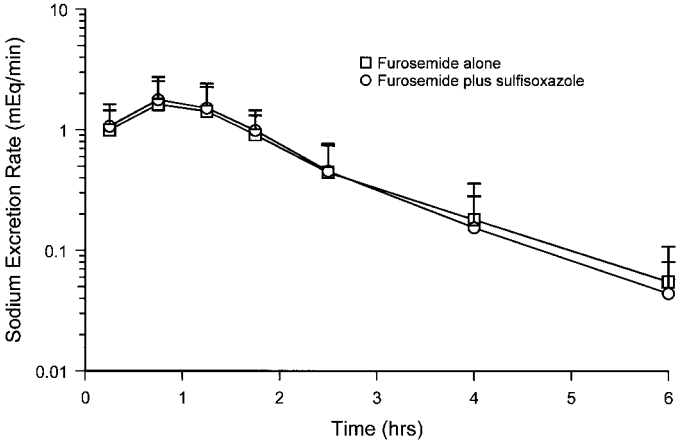


*Figure 3.* Urinary sodium excretion rate caused by furosemide with and without sulfisoxazole.

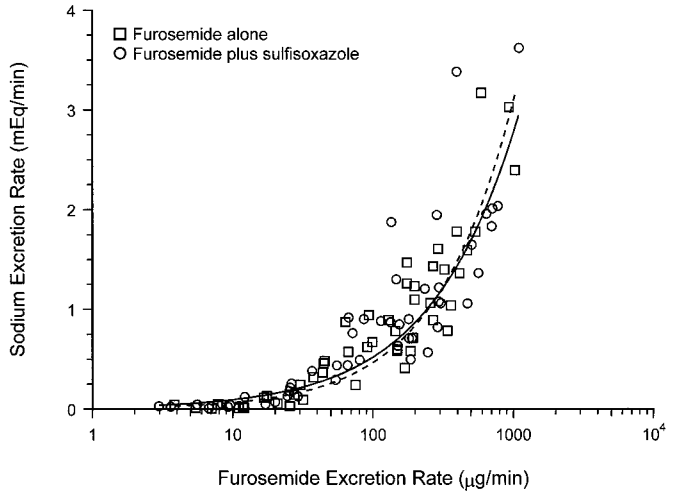


*Figure 4.* Relationship between urinary excretion rate of furosemide and sodium excretion rate with and without sulfisoxazole.

the effects of loop diuretics in patients with NS. The human studies could be explained by increased proximal tubular reabsorption of sodium, increased distal tubular reabsorption of sodium, and/or an effect of NS to somehow alter the dynamics of the inhibition of the $Na^+$-$K^+$-$2Cl^-$ transporter by a loop diuretic. At the very least, the animal study indicates that the last of these possibilities occurs but does not exclude increased solute reabsorption proximally or distally.

The results of this study indicate that therapeutic strategies aimed at displacing furosemide from urinary protein binding are not indicated. Moreover, we propose that our results coupled with other studies of possible mechanisms of diuretic resistance in NS argue that the dominant mechanism is nephron resistance to the effect of loop diuretics. Future studies should attempt to further dissect the mechanisms of this tubular resistance so that rational therapeutic strategies addressing this pathophysiology are possible.

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## References

1. Brater DC: Diuretic therapy. *N Engl J Med* 339: 387–395, 1998
2. Inoue M, Okajima K, Itoh K, Ando Y, Watanabe N, Yasaka T, Nagase S, Morino Y: Mechanism of furosemide resistance in analbuminemic rats and hypoalbuminemic patients. *Kidney Int* 32: 198–203, 1987
3. Green TP, Mirkin BL: Resistance of proteinuric rats to furosemide: Urinary drug protein binding as a determinant of drug effect. *Life Sci* 26: 623–630, 1980
4. Green TP, Mirkin BL: Furosemide disposition in normal and proteinuric rats: Urinary drug-protein binding as a determinant of drug excretion. *J Pharmacol Exp Ther* 218: 122–127, 1981
5. Voelker JR, Jameson DM, Brater DC: In vitro evidence that urine composition affects the fraction of active furosemide in the nephrotic syndrome. *J Pharmacol Exp Ther* 250: 772–778, 1989
6. Kirchner KA, Voelker JR, Brater DC: Intratubular albumin blunts the response to furosemide—A mechanism for diuretic resistance in the nephrotic syndrome. *J Pharmacol Exp Ther* 252: 1097–1101, 1990
7. Kirchner KA, Voelker JR, Brater DC: Binding inhibitors restore furosemide potency in tubule fluid containing albumin. *Kidney Int* 40: 418–424, 1991
8. Chennavasin P, Seiwell R, Brater DC: Pharmacodynamic analysis of the furosemide-probenecid interaction in man. *Kidney Int* 16: 187–195, 1979
9. Chennavasin P, Seiwell R, Brater DC: Pharmacokinetic-dynamic analysis of the indomethacin-furosemide interaction in man. *J Pharmacol Exp Ther* 215: 77–81, 1980

10. Voelker JR, Brown-Cartwright D, Anderson S, Leinfelder J, Sica DA, Kokko JP, Brater DC: Comparison of loop diuretics in patients with chronic renal insufficiency: Mechanism of difference in response. *Kidney Int* 32: 572–578, 1987
11. Kaojarern S, Day B, Brater DC: The time course of delivery of furosemide into urine is an independent determinant of overall response. *Kidney Int* 22: 69–74, 1982
12. Beerman B, Dalén E, Lindström B: Elimination of furosemide in healthy subjects and in those with renal failure. *Clin Pharmacol Ther* 22: 70–78, 1977
13. Rose HJ, O'Malley K, Pruitt AW: Depression of renal clearance of furosemide in man by azotemia. *Clin Pharmacol Ther* 21: 141–146, 1976
14. Traeger A, Stein G, Sperschneider H, Keil E: Pharmacokinetic and pharmacodynamic effects of furosemide in patients with impaired renal function. *Int J Clin Pharmacol Toxicol* 22: 481–486, 1984
15. Brater DC, Chennavasin P, Seiwell: Furosemide in patients with heart failure: Shift of the dose-response relationship. *Clin Pharmacol Ther* 28: 182–186, 1980
16. Vargo D, Kramer WG, Black PK, Smith WB, Serpas T, Brater DC: Bioavailability, pharmacokinetics, and pharmacodynamics of torsemide and furosemide in patients with congestive heart failure. *Clin Pharmacol Ther* 57: 601–609, 1995
17. Fredrick MJ, Pound DC, Hall SD, Brater DC: Furosemide absorption in patients with cirrhosis. *Clin Pharmacol Ther* 49: 241–247, 1991
18. Fuller R, Hoppel C, Ingalls ST: Furosemide kinetics in patients with hepatic cirrhosis with ascites. *Clin Pharmacol Ther* 30: 461–467, 1981
19. Villeneuve JP, Verbeeck RK, Wilkinson GR, Branch RA: Furosemide kinetics and dynamics in patients with cirrhosis. *Clin Pharmacol Ther* 40: 14–20, 1986
20. Verbeeck RK, Patwardhan RV, Villeneuve JP, Wilkinson GR, Branch RA: Furosemide disposition in cirrhosis. *Clin Pharmacol Ther* 31: 719–725, 1982
21. Mees EJD: Does it make sense to administer albumin to the patient with nephrotic oedema? *Nephrol Dial Transplant* 11: 1224–1226, 1996
22. Akcicek F, Yalniz T, Basci A, Ok E, Mees EJD: Diuretic effect of furosemide in patients with nephrotic syndrome: Is it potentiated by intravenous albumin? *Br Med J* 310: 162–163, 1995
23. Fliser D, Zurbrüggen, Mutschuler E, Bischoff I, Nussberger J, Franek E, Ritz E: Coadministration of albumin and furosemide in patients with the nephrotic syndrome. *Kidney Int* 55: 629–634, 1999
24. Kirchner KA, Voelker JR, Brater DC: Tubular resistance to furosemide contributes to the attenuated diuretic response in nephrotic rats. *J Am Soc Nephrol* 2: 1201–1207, 1992
25. Keller E, Hoppe-Seyler G, Schollmeyer P: Disposition and diuretic effect of furosemide in the nephrotic syndrome. *Clin Pharmacol Ther* 32: 442–449, 1982
26. Prandota J: Urinary elimination kinetics and diuretic effect of intravenous furosemide in nephrotic children. *Dev Pharmacol Ther* 5: 98–108, 1982
27. Smith DE, Hyneck ML, Berardi RR, Port FK: Urinary protein binding, kinetics, and dynamics of furosemide in nephrotic patients. *J Pharm Sci* 74: 603–607, 1985
28. Sjöström PA, Odlind BG, Beermann BA, Karlberg BE: Pharmacokinetics and effects of furosemide in patients with the nephrotic syndrome. *Eur J Clin Pharmacol* 37: 173–180, 1989