*Inflammatory Bowel Diseases*®
6(4):275–279 © 2000 Crohn's & Colitis Foundation of America, Inc.

# Glomerular and Tubular Renal Functions After Long-Term Medication of Sulphasalazine, Olsalazine, and Mesalazine in Patients with Ulcerative Colitis

Grethe Støa Birketvedt, *Knut J. Berg, †Olav Fausa, and Jon Florholmen

*Laboratory of Gastroenterology, Institute of Clinical Medicine, University of Tromsø, Tromsø; and *Laboratory for Renal Physiology and †Department of Medicinet, Rikshospitalet, Oslo, Norway*

**Summary:** To date there are only few reports evaluating the potential nephrotoxic reactions of the new 5-aminosalicylic acid (5-ASA) preparations in patients with ulcerative colitis (UC). The aim of this study was to screen the tubular and glomerular functions in patients with UC in maintenance treatment with either 5-ASA azo-compounds (sulphasalazine and olsalazine) or mesalazine. Patients with UC in clinical remission treated with either sulphasalazine, olsalazine, or mesalazine for more than 1 year were included in an open, single-blind retrospective Norwegian multicenter study. Serum and urine creatinine, serum and urine $\beta_2$-microglobulin, urine N-acetyl-β-glucoseamidase (NAG), urine alkaline phosphatase, urine microalbumin, urine alanine amino peptidase, and urine $\beta_2$-microglobulin were measured. Fifty-two females and 75 males (n = 127), ages 20–69, were evaluated. Thirty-six patients were treated with sulphasalazine (mean treatment time 10.1 ± 6.6 years [mean ± SD]), 32 patients were treated with olsalazine (2.3 ± 1.4 years), and 59 patients with mesalazine (3.2 ± 2.0 years). At inclusion, there were no significant differences in the serum or urine values between the groups. In 17 patients (1 patient [3%] in the sulphasalazine group, 4 patients [13%] in the olsalazine group, and 12 patients [20%] in the mesalazine group), at least one abnormal serum and/or urine value was detected. After 10 years of treatment, only one abnormal value was found among the 19 patients in the sulphasalazine group. The abnormal values observed in the other groups indicated minor glomerular or tubular renal damage. In conclusion, long term sulphasalazine treatment appears to be safe and free of nephrotoxic side effects, whereas minor glomerular and tubular impairment are observed in a few patients treated with olsalazine and mesalazine. **Key Words:** Nephrotoxicity—5-Aminosalicylic acid.

## INTRODUCTION

It is well documented that sulphasalazine is efficient to induce and maintain remission in ulcerative colitis (UC) (1). Sulphasalazine (Salazopyrin) consists of two components: 5-aminosalicylic acid (5-ASA), the active moiety of the drug, and sulphapyridine, most often responsible for the majority of adverse reactions observed (2).

A second generation of sulphasalazine free of sulphapyridine (mesalazine and olsalazine) and with better delivery of 5-ASA has been developed. In an enteric-coated preparation of mesalazine (delayed-release Mesasal and Asacol or sustained-release Pentasa), the 5-ASA is released into the small intestine and the proximal colon. Olsalazine (Dipentum) comprises two molecules of 5-ASA diazotized by an azo-bound identical to the azo-bound in sulphasalazine and is released to the colon. Compared with sulphasalazine and olsalazine, mesalazine is more completely absorbed in the intestine and is better delivered into the circulatory system. Moreover, 5-ASA is acetylated to acetyl-5-ASA (Ac-5-ASA) in the epithelium of the small intestine and the colon. The capacity of acetylation in the epithelium is low in the small intestine, but high in the colon (3), which may explain the differences in the concentration of 5-ASA observed in blood and urine in various 5-ASA preparations (4,5).

5-ASA is structurally similar to aspirin (acetylsalicylic acid). Free 5-ASA, but not acetylated 5-ASA, is nephrotoxic in rats (3). Nephrotoxicity is a theoretical concern with the use of mesalazine. There are several reports of nephrotoxic reactions caused by mesalazine (6–14). Among the reported cases are interstitial nephritis (6–

Received March 28, 2000; accepted July 27, 2000.

Address correspondence and reprint requests to Dr. G. Støa Birketvedt, Laboratory of Gastroenterology, Institute of Clinical Medicine, 9037 University of Tromsø, Norway.

11), nephrotic syndrome (12), acute renal failure (13), nephrogenic diabetes insipidus (14), and unexplained pyuria, trace proteinuria, and microhematuria (15). To date there are few reports on tubular functions in patients treated with 5-ASA compounds. Some reports describe renal tubular dysfunction (16–18), whereas in other reports no tubular dysfunction could be detected (19,20). However, these studies are connected with methodological problems, particularly in measuring urinary excretion of tubular enzymes. These reports may indicate a potential clinical problem when changing the treatment of sulphasalazine to a new 5-ASA compound.

Based on sensitive renal tests, the primary objective of this study was to screen the glomerular and tubular functions in patients with UC in maintenance treatment with either 5-ASA azo-compounds (sulphasalazine and olsalazine) or mesalazine.

## PATIENTS AND METHODS

### Patients

The inclusion criteria in the study were 1) patients ages 18–70 years with diagnostic UC and UC in clinical remission (≤3 stools/day) in the last month; 2) treatment with current UC maintenance medication (Salazopyrin [sulphasalazine], Dipentum [olsalazine] [Kabi Pharmacia, Oslo, Norway], and Mesasal [mesalazine] [Smith Kline and French, Oslo, Norway] for ≥12 months); and 3) informed consent to participate on a voluntary basis. Excluded were patients with blood, protein, and/or leukocytes in urine, abnormal serum creatinine, and other clinical signs of renal disease not related to UC and/or its treatment. Also excluded were patients with cardiac or hepatic failure, diabetes mellitus, patients treated with diuretics or antihypertensive drugs, patients treated acutely or chronically with salicylates (≥300 mg/day), nonsteroidal antiinflammatory drugs, or immune modifiers (except steroids), pregnant women, and patients with acute or chronic intercurrent disease that impairs renal function. According to the inclusion and exclusion criteria above, a patient was defined as having a renal disease associated with the treatment of UC when all other causes was excluded.

### Study Design

The evaluation of urinary enzymes in patients with UC was designed as an open, controlled, single-blind retrospective multicenter study with two treatment groups: 5-ASA azo-compounds (sulphasalazine and olsalazine) and mesalazine. The intention was to recruit 100 outpatients in each group during a period of 12 months from 12 hospitals in Norway. In the group of azo-compounds, the number of patients was approximately equal for olsalazine and sulphasalazine. Clinical assessments were done at entry, recording demographic data, history of UC including drug administration, and maximum extent of disease recorded by endoscopy.

Blood was drawn and serum samples were immediately frozen at −20°C. Patients collected urine overnight (minimum 6 hours), and the urine samples were delivered to the laboratory within 2 hours after the morning urinary samplings. Volume, pH, and blood, protein, and leukocytes were measured. To follow standardized procedure prior to storage at −20°C, urine samples were pH-adjusted to 6.5–8 to measure $\beta_2$-microglobulin, and gycerol was added to measure alanine amino peptidase (AAP); no pretreatment was required for the other analyses.

Blood and urine samples were shipped to the Laboratory for Renal Physiology, Department of Medicine, Rikshospitalet, Oslo, Norway; analyses were done at 2 months ± 1 week as blind coded samples.

Serum albumin was determined according to the method of Doumas et al. (23). Serum values of creatinine and $\beta_2$-microglobulin, and overnight urine values of creatinine, N-acetyl-β-glucoseamidase (NAG), alkaline phosphatase, microalbumine, alanine amino peptidase, and $\beta_2$-microglobulin were measured according to previously described methods (22,23). The calculation of creatinine clearance was based on the concentration of creatinine in serum and urine and the volume of the timed collection of urine. Values outside the reference limits were defined as abnormal (22,23).

### Statistical Methods

Computer program package SAS (24), procedure GLM, was used to test for differences in the serum and the urine values between sulphasalazine, olsalazine, and mesalazine. Tests of seven different parameters were performed, and p values < 0.01 were considered significant. The continuous variables describing the patients' characteristics were presented by the mean ± standard deviation and/or range, and the categorical variables were presented as the number of patients in each category group.

## RESULTS

Within 12 months, 139 patients were recruited. Twelve patients did not fulfill the inclusion/exclusion criteria and were therefore excluded. The 127 remaining

patients, 75 male and 52 female, 20–69 years of age, were included in the study. The demographic data at the inclusion are shown in Table 1.

There were no significant differences in the serum or urine test results between these three groups (Table 2). Seventeen patients had a total of 20 abnormal (outside the reference limits) serum and/or urine values: 1 patient (3%) in the sulphasalazine group, 4 patients (13%) in the olsalazine group, and 12 patients (20%) in the mesalazine group. In one patient (mesalazine), the serum and urine concentrations of $\beta_2$-microglobulin were both abnormal, in another patient (mesalazine), the urine concentrations of NAG and $\beta_2$-microglobulin were both abnormal. In a third patient (olsalazine), the creatinine clearance was just below the lower reference limit, and the urine concentration of $\beta_2$-microglobulin was abnormal. In the remaining patients, one abnormal value was observed. In five patients (two on olsalazine and three on mesalazine), the creatinine clearance limit (range 45–59 ml/min/1.73 $m^2$) was just below the lower reference limit. In the 17 patients with abnormal values, the duration of disease and treatment, as well as the dose of drug used, did not seem to differ from the rest of the patients (data not shown). All abnormal values found indicated only a minor glomerular or tubular renal impairment/damage, and no patients were judged as having clinically significant renal disease caused by 5-ASA treatment.

Among the 19 patients in the sulphasalazine group with a total treatment time of more than 10 years, only 1 patient had one abnormal value (NAG).

## DISCUSSION

In this study we have characterized the glomerular and tubular functions in patients with UC in maintenance treatment with either 5-ASA azo-compounds (sulphasalazine and olsalazine) or mesalazine based on sensitive tubular and glomerular tests. Only minor signs of renal disease were observed. None of these patients were judged as having clinically significant renal disease caused by 5-ASA treatment.

In patients treated with sulphasalazine, including those subjects with a relatively long duration of treatment (>10 years), only 3% showed traces of renal dysfunction based on these sensitive renal tests. During the 50 years that sulphasalazine has been available as a treatment in patients with UC, only a few cases have been reported on drug-induced renal failure (25,26). Both glomerular (25) and tubular nephrotoxicity (26) were described in these reports in patients treated for longer periods. The apparent low incidence of sulphasalazine-induced nephrotoxicity is most likely explained by drug pharmacokinetics. The nephrotoxic component is the 5-ASA molecule that is released in the colon. The colonic epithelium has a great capacity to acetylate 5-ASA to Ac-5ASA (3). We also know from pharmacokinetic studies that the systemic load of 5-ASA is low in patients with UC treated with sulphasalazine (4,5).

In the olsalazine and mesalazine groups, indications of tubular and/or glomerular impairment were observed in 13% and 20%, respectively. The treatment time of these two new 5-ASA preparations was 2–3 years, compared with 10 years in patients treated with sulphasalazine. This indicated that abnormal tests were observed very early on in patients treated with the new 5-ASA compounds, compared with patients treated with sulphasalazine. The abnormal tests indicated that the tubular function was impaired more frequently than the glomerular function, especially when expressed as abnormal urine concentrations of $\beta_2$-microglobulin in 6 of the 17 patients with abnormal renal tests. In patients with UC treated with the new 5-ASA preparations, impaired tubular function (16–18) had been observed in about 20% of the patients, whereas no tubular impairment had been

**TABLE 1.** *Demographic data*

| Treatment groups | Sulphasalazine | Olsalazine | Mesalazine |
|---|---|---|---|
| Female/male (number) | 8/28 | 15/17 | 29/30 |
| Age (year) | 43.1 ± 11.3 | 43.5 ± 12.9 | 41.4 ± 12.4 |
| Duration (year) of disease | 12.5 ± 7.9 | 8.5 ± 7.1 | 9.1 ± 6.9 |
| Duration (year) of present treatment | 10.1 ± 6.6 | 2.3 ± 1.4 | 3.2 ± 2.0 |
| Dose of present treatment (g/day) | 2.1 ± 0.5 | 1.2 ± 0.5 | 1.4 ± 0.4 |
| Duration (year) of previous treatment | | | |
| Sulphasalazine | 1.5 ± 3.3 | 4.0 ± 5.3 | 1.7 ± 3.0 |
| Olsalazine | 0 | 0.03 ± 0.18 | 0.05 ± 0.22 |
| Mesalazine | 0.03 ± 0.17 | 0.09 ± 0.30 | 0.20 ± 0.87 |
| Number of episodes of active ulcerative colitis last 12 months | 0.5 ± 0.7 | 0.5 ± 1.0 | 0.7 ± 0.9 |
| Maximum extent of disease ever recorded by endoscopy | | | |
| Rectum | 4 | 1 | 6 |
| Left colon | 13 | 18 | 23 |
| Total colon | 19 | 13 | 30 |

**TABLE 2.** *Serum and urine glomerular and tubular function parameters in patients with ulcerative colitis on long-term medication (>1 year) of 5-aminosalicylic acid*

| Serum/urine parameter | | Sulphasalazine | | Olsalazine | | Mesalazine | | p value** | Upper reference limit |
|---|---|---|---|---|---|---|---|---|---|
| Serum $\beta_2$-microglobulin μg/l | N | 35 | 0* | 32 | 0* | 59 | 3* | 0.02 | 2170 |
| | Mean | 1,434 | | 1,260 | | 1,466 | | | |
| | SD | 302 | | 338 | | 347 | | | |
| | Min | 1,029 | | 434 | | 860 | | | |
| | Max | 2,150 | | 2,100 | | 2,390 | | | |
| Urine N-acetyl-β-glucose-amidase U/mmol creatinine | N | 35 | 1 | 32 | 1 | 59 | 1 | 0.86 | 0.284 |
| | Mean | 0.21 | | 0.123 | | 0.114 | | | |
| | SD | 0.103 | | 0.065 | | 0.073 | | | |
| | Min | 0.010 | | 0.020 | | 0.003 | | | |
| | Max | 0.560 | | 0.320 | | 0.380 | | | |
| Urine alkaline phosphatase U/mmol creatinine | N | 34 | 0 | 32 | 0 | 59 | 0 | 0.19 | 0.723 |
| | Mean | 0.144 | | 0.153 | | 0.181 | | | |
| | SD | 0.105 | | 0.095 | | 0.108 | | | |
| | Min | 0.010 | | 0.020 | | 0.020 | | | |
| | Max | 0.390 | | 0.360 | | 0.590 | | | |
| Urine albumin μg/μmol creatinine | N | 32 | 0 | 30 | 0 | 59 | 1 | 0.67 | 4.462 |
| | Mean | 0.544 | | 0.502 | | 0.727 | | | |
| | SD | 0.690 | | 0.376 | | 1.706 | | | |
| | Min | 0.030 | | 0.100 | | 0.030 | | | |
| | Max | 3.270 | | 1.690 | | 13.080 | | | |
| Urine alanine amino peptidase U/mmol creatinine | N | 32 | 0 | 29 | 0 | 58 | 2 | 0.53 | 1.388 |
| | Mean | 0.468 | | 0.556 | | 0.551 | | | |
| | SD | 0.293 | | 0.383 | | 0.385 | | | |
| | Min | 0.010 | | 0.020 | | 0.010 | | | |
| | Max | 1.130 | | 1.310 | | 1.730 | | | |
| Urine $\beta_2$-microglobulin μg/mmol creatinine | N | 35 | 0 | 32 | 2 | 59 | 4 | 0.22 | 20.334 |
| | Mean | 7.72 | | 8.97 | | 12.75 | | | |
| | SD | 3.76 | | 7.04 | | 20.30 | | | |
| | Min | 1.57 | | 1.94 | | 1.16 | | | |
| | Max | 18.02 | | 41.78 | | 158.4 | | | |
| Creatinine clearance ml/min/1.73 $m^2$ | N | 35 | 0 | 32 | 2 | 59 | 3 | 0.75 | 60*** |
| | Mean | 101 | | 101 | | 97 | | | |
| | SD | 24 | | 27 | | 27 | | | |
| | Min | 64 | | 45 | | 45 | | | |
| | Max | 154 | | 178 | | 211 | | | |

* Number of patients out of normal range.
** Test of equal means in the analysis of variance.
*** Lower reference limit.
SD, standard deviation.

found in two other studies (19,20). The discrepancies between these studies are difficult to explain.

The mechanisms behind the 5-ASA-induced nephrotoxicity are poorly understood. Hypersensitivity reactions to 5-ASA have been proposed to cause the acute interstitial nephritis observed in some patients (9,26). High serum concentrations of 5-ASA, which have been shown to exert toxic renal effect in animal models (3), may be another potential pathophysiological mechanism. It is interesting to note that in our study the abnormal renal tests were most frequently observed in the mesalazine group. It is well documented that mesalazine causes nephrotoxic side effects (6–15). As far as we know, no reports exist on nephrotoxicity caused by olsalazine. Moreover, in two previous reports, serum concentrations of 5-ASA were higher in the mesalazine group, compared with the olsalazine and the sulphasalazine groups (4,5). In the two latter groups, unexpectedly high levels of both urinary and serum 5-ASA concentrations were observed, especially in patients treated with mesalazine (5). This may indicate that among the new 5-ASA compounds, mesalazine has the greatest potential to cause renal damage.

The clinical implications of our findings are unresolved. Administration of sulphasalazine for more than 10 years most likely does not cause renal damage. In contrast, minor tubular and glomerular renal damage apparently exists in patients treated with the new 5-ASA

preparations within 2–3 years of treatment. To date there are no reports indicating the clinical significance of minor tubular and glomerular damage based on the very sensitive tests used in this study. We do not know if these changes represent progressive renal disease. However, our observations agree with the increasing number of case reports of serious nephrotoxicity associated with some of the new 5-ASA preparations. Prospective studies are required to fully evaluate the apparent potential nephrotoxic effect of long-term administration of the new 5-ASA preparations. Therefore our study, along with other studies (18,27), warn clinicians to be aware of the potential renal toxicity in long-term treatment with the new 5-ASA compounds. As proposed by Mehta (27), clinicians should avoid the new 5-ASA preparations in patients with renal disease; it is advisable to monitor renal function in patients on chronic 5-ASA therapy for inflammatory bowel disease. Moreover, the same caution is recommended in patients at risk for developing renal disease, such as patients with diabetes mellitus and hypertension (21).

In conclusion, long-term sulphasalazine treatment appears to be safe from nephrotoxic side effects, whereas indications of minor glomerular and tubular impairment have been observed in a few patients treated with olsalazine and mesalazine.

**Acknowledgment:** The authors are grateful to the other investigators of the multicenter study: Erling Aadland, Aker Sykehus, and Idar Lygren, Ullevål Sykehus, Oslo; Ståle Barstad , Sentralsykehuset i Rogaland, Stavanger; Svein Ødegaard, Haukeland Sykehus, Bergen; Mikal Tønder, Sentralsykehuset i Vestfold, Tønsberg; Asbjørn Stallemo, Sentralsykehuset i Vest Agder, Kristiansand; Stein Kildebo, Sentralsykehuset i Hedemark, Elverum; Ivar Blix, Fykessykehuset i Kristiansund, Kristiansund; Ulf Fjøsne, Innherred Sykehus, Levanger; and Anna-Maria Wessel-Berg, Sentralsykehuset i Nordland, Bødø, Norway. Special thanks to Gary Jansson and Helena Eriksson of the Medical Department, Pharmacia & Upjohn, Uppsala, Sweden, and Gøril Knutsen, Pharmacia & Upjohn, Oslo, Norway, for statistical analysis and study coordination; and to Kirsten Klingenberg Lund at the Laboratory for Renal Physiology, Rikshospitalet, Oslo, Norway, for superb technical assistance. This study was financially supported by Pharmacia & Upjohn, Uppsala, Sweden.

## REFERENCES

1. Sutherland LR, Roth DE, Beck PL. Alternatives to sulphasalazine: a metaanalysis of 5-ASA in the treatment of ulcerative colitis. *Inflammatory Bowel Diseases* 1997;3:65–78.
2. Aazad Khan AK, Piris J, Truelove SC. An experiment to determine the active therapeutic moiety of sulphasalazine. *Lancet* 1977;2: 892–5.
3. Calder IC, Funder CC, Green CR, et al. Comparative toxicity of aspirin and phenacetin derivatives. *BMJ* 1971;4:518–21.
4. Stærk Laursen L, Stokholm M, Bukhave K, et al. Disposition of 5-aminosalicylic acid by olsalazine and mesalazine preparations in patients with ulcerative colitis: comparison of intraluminal colonic concentrations, serum values, and urinary excretion. *Gut* 1990;31: 1271–6.
5. Støa Birketevdt G, Florholmen J. The systemic load and efficient delivery of active 5-aminosalicylic acid in patients with ulcerative colitis on treatment with olsalazine or mesalazine. *Aliment Pharmacol Ther* 1999;13:357–61.
6. Ruf-Ballauf W, Hofstädter F, Krentz K. Akute interstitielle nephritis durch 5- aminosalicylsäure? *Internist* 1989;30:262–4.
7. Mehta RP. Acute interstitial nephritis due to 5-aminosalicylic acid. *Can Med Assoc J* 1990;143:1031–2.
8. Henning HV, Meinhold J, Eisenhauer T, et al. Chronische interstitielle nephritis nach behandlung mit 5-aminosalicylsäure. *Dtsch Med Wochenschr* 1989;114:1091.
9. Thuluvath PJ, Ninkovic M, Calsam J, et al. Mesalazine induced interstitial nephritis. *Gut* 1994;35:1493–6.
10. SmildeTJ, van Liebergen FJ, Koolen MI, et al. Tubulointerstitial nephritis caused by mesalazine (5-ASA) agents. *Ned Tijdschr Geneeskd* 1994;138:2557–61.
11. Brouillard M, Gheerbrant JD, Gheysens Y, et al. Chronic interstitial nephritis and mesalazine: 3 new cases? *Gastroenterol Clin Biol* 1998;22:724–6.
12. Novis BH, Korzets Z, Chen P, et al. Nephrotic syndrome after treatment with 5-aminosalicylic acid. *Gut* 1988;296:1442.
13. Garcia-Diaz M, Nevado L, Berenguer A, et al. Acute renal failure associated to 5-aminosalicylic acid in the intestinal inflammatory disease. *Gastroenterol Hepatol* 1995;18:18–21.
14. Masson EA, Rhodes JM. Mesalazine associated nephrogenic diabetes insipidus presenting as weight loss. *Gut* 1992;33:563–4.
15. Tremaine WJ, Schroeder KW. Urinary sediment abnormalities in patients on long-term oral 5-aminosalicylic acid (5-ASA) for chronic ulcerative colitis (CUC). *Gastroenterology* 1988;94:465.
16. Tsamis D, Macpherson A, Forgac I, et al. Urine enzyme profiles as indicators of renal tubular dysfunction in inflammatory bowel disease (ID). *Gut* 1992;33(suppl 1):W34.
17. Riley SA, Lloyd D, Mani V. Test of renal function in patients with quiescent colitis: effects of drug treatment. *Gut* 1992;33:1348–52.
18. Schreiber S, Hämling J, Zehnter E, et al. Renal tubular dysfunction in patients with inflammatory bowel disease treated with aminosalicylate. *Gut* 1997;40:761–6.
19. Diener U, Tuczek HV, Fischer C, et al. Renal function was not impaired by treatment with 5-aminosalicylic acid in rats and man. *Arch Pharmacol* 1984;326:278–82.
20. Biddle W, Miner PB. Evaluation of potential nephrotoxicity of oral 5-ASA treatment of patients with ulcerative colitis. *Gastroenterol* 1989;96:A44.
21. Doumas BT, Peters T. Serum and urine albumin: a progress report on their measurement and clinical significance. *Clin Chim Acta* 1997;258:3–20.
22. Berg KJ, Kolmannskog F, Lillevold PE, et al. Iopentol in patients with chronic renal failure: its effect on renal function and its use as glomerular filtration rate parameter. *Scand J Clin Lab Invest* 1992; 52:27–33.
23. Berg KJ, Kristoffersen DT, Djøseland O, et al. Reference range of some enzymes and proteins in untimed overnight urine and their stability after freezing. *Clin Chim Acta* 1998;272:225–30.
24. SAS Institute, Inc. *SAS STAT User's Guide*, vol. 2, 4th ed., version 6. Cary, NC: SAS Institute Inc, 1989:846.
25. Barbour VM, Williams PF. Nephrotic syndrome associated with sulphasalazine. *BMJ* 1990;301:818.
26. Dwarakanath AD, Michael J, Allan RN. Sulphasalazine induced renal failure. *Gut* 1992;33:1006–7.
27. Mehta RP. Nephropathy from 5-aminosalicylate preparations. *Inflammopharmacol* 1993;2:297–300.