Aliment Pharmacol Ther 1997; 11: 679–684.

Retrograde colonic spread of a new mesalazine rectal enema in patients with distal ulcerative colitis

P. GIONCHETTI*, A. VENTURI*, F. RIZZELLO*, C. CORBELLI†, S. FANTI†, M. FERRETTI*, S.BOSCHI‡, M. MIGLIOLI* & M. CAMPIERI*

**Clinica Medica e Gastroenterologia, Policlinico S. Orsola, Università di Bologna, Italy; †Servizio di Medicina Nucleare, Ospedale S. Orsola, Università di Bologna, Italy; and ‡Servizio di Farmacologia Clinica, Policlinico S. Orsola, Università di Bologna, Italy*

Accepted for publication 9 February 1997

SUMMARY

Background: Rectal treatment with mesalazine enemas is the first-line therapy for distal ulcerative colitis. In order to improve the benefits of rectal therapy, a new 60 mL 5-ASA rectal gel enema preparation has been developed using a device which excludes direct contact of the inert propellant gas with the active drug. The purpose of the present study was to assess by scintigraphy the colonic distribution of this new mesalazine rectal gel enema.
Methods: Twelve patients with active ulcerative colitis were administered 4 g of the mesalazine rectal enema labelled with 100 MBq technetium sulphur colloid (^{99m}Tc-SC). Anterior scans of the abdomen were acquired at intervals for 4 h. Scans were analysed to evaluate the extent of retrograde flow and homogeneity of distribution of the radiolabelled enema in the rectum, sigmoid, descending and transverse colon. In addition, plasma levels of 5-ASA and Ac-5-ASA were measured for 6 h.
Results: All patients retained the entire rectal gel throughout the course of the study without reporting adverse events. In 11 out of 12 patients (92%) the gel had spread homogeneously beyond the sigmoid colon and had reached the upper limit of disease in all cases. The maximum spread (splenic flexure) was observed in 6 out of 12 patients (50%) within the first 2 h. The systemic absorption of mesalazine and its metabolite Ac-5-ASA was low.
Conclusions: The new mesalazine enema represents an adequate alternative and a further technological improvement in the topical treatment of distal ulcerative colitis.

INTRODUCTION

In recent years rectal administration of mesalazine has become an established treatment for distal ulcerative colitis. In fact this approach has been designed in order to ensure high local concentrations of mesalazine to the inflamed mucosa, minimizing systemic effects.[1–3]

Mesalazine solution or foam enemas and suppositories are commercially available and widely used for the treatment of patients with proctosigmoiditis or distal ulcerative colitis and proctitis, respectively.[4–9]

Available data about retrograde spread in the colon show that suppositories have a limited spread in the rectum and distal sigmoid colon[10] while enema solutions have a greater spreading capacity, even though highly variable.[11, 12]

In addition, administration of enema solutions has some disadvantages due to problems with retention, which may interfere negatively with compliance to the topical treatment.[9]

Colonic mesalazine foams provide a rapid and homogeneous distribution in the colon.[13, 14] They differ

Correspondence to: Dr. P. Gionchetti, Clinica Medicae Gastroenterologia, Pol. S. Orsola, Univ. di Bologna, Via Massarenti 9, 40138 Bologna, Italy.

substantially from the steroid rectal foams, which have a limited retrograde spread, for the reason that the mesalazine foams generate large volumes within the colon due to the addition of hydrocarbon propellants in the formulation.[11, 15]

In order to further improve tolerance and efficacy of topical therapy with mesalazine, a new formulation for rectal administration has recently been developed. The ready-to-use mesalazine enema (4 g in a total volume of 60 mL) is a suspension of high viscosity gel that ensures better adhesion, and hence better distribution of the preparation to the colonic mucosa.

The primary objective of this study was to evaluate by scintigraphy the colonic retrograde progression of the mesalazine rectal gel preparation in patients with active ulcerative colitis. The secondary objective was to determine the systemic absorption profile of the new mesalazine enema.

MATERIALS AND METHODS

Patients

This was an open label, non-controlled study of a single administration of the new mesalazine rectal enema (containing 4 g of mesalazine) in 12 patients with mild to moderate active distal ulcerative colitis. Disease activity was defined by the criteria of Truelove & Witts.[16]

Activity and extent of disease were assessed by colonoscopy not more than 7 days before the start of the study. Inclusion criteria were: disease extent not less than 20 cm from the anus; and withdrawal of treatment with sulphasalazine or other mesalazine preparations at least 48 h prior to entry into the study. Patients were excluded if there was: a history of segmental colon resection; liver and kidney insufficiency; intolerance or allergy to salicylate compounds; or concomitant oral treatment with sulphasalazine or mesalazine preparations and with drugs likely to interfere with gastrointestinal motility. Women of child-bearing potential and who were breast feeding were also excluded.

Study formulation

The product used in the study was a 60 mL single-dose high-viscosity thixotropic mesalazine suspension containing 4 g of mesalazine. The preparation was contained in a canister fitted with a valve.

The spray system consisted of a two-chamber device: an inner chamber (a flexible polyethylene-coated aluminium bag) containing the mesalazine suspension, and an outer chamber (an airtight can) filled with pressurized nitrogen acting as a propellant. Upon activation of the valve, the pressurized nitrogen squeezes the inner bag containing the suspension, which is released into the colon via a disposable rectal applicator.

The novelty of this device is the fact that the propellant gas is retained inside the can and is not delivered into the colon. On the other hand the characteristics of the suspension are such that an easy and complete release of the mesalazine suspension from the canister can be achieved, as well as better adhesion to the colonic mucosa.

Scintigraphic study

A scintigraphic investigation was conducted to assess the retrograde progression of the new dosage form of mesalazine, and its distribution over the colonic mucosa. The product was labelled with 100 Mbq ^{99}technetium sulphur colloid (^{99m}Tc-SC), a gamma ray emitter. Because ^{99m}Tc-SC has a half-life of 6 h, it was necessary to label the contents of each canister not more than 2 h before administration. The radiolabelling technique consisted of incorporating 0.2 mL of the radiopharmaceutical directly into 60 mL of mesalazine suspension, which was then introduced to the canister. The total effective radiation dose per patient was 0.039×100 mSv. A previously described method was used for the preliminary assessment of tracer distribution within the gel preparation.[13] In summary, samples of the radiolabelled preparation obtained over a 4-h period were distributed on 50-cm strips. A uniform distribution of radioactivity was then verified by obtaining subsequent static images. The stability of the ^{99m}Tc-SC preparation was confirmed by thin-layer chromatography.

Abdominal images were obtained using a large-field-of-view gamma camera (Gamma-diagnost, Philips, Eindhoven NL) equipped with a low-energy, high-resolution parallel-hole collimator and linked to a dedicated nuclear medicine computer (PDP11 73 Digital Mainard, Mass., USA).

Prior to administering the radiolabelled preparation, the patients were asked to evacuate both their bowel and bladder.

All patients were administered a single dose of the radiolabelled enema while lying on their left side. The exact amount of the administered mesalazine enema

was determined by weighing the canister before and after administration.

After the enema had been administered, the patients remained in a supine position on the diagnostic table throughout the registration period, which was 4 h, after which they were free to move. The patients were not allowed to smoke or drink coffee or alcoholic beverages, and fasted from midnight prior to the study day and during the study period.

After administration of the labelled enema preparation, anterior images of the abdominal region were taken with the patient lying supine. For each scan 500 K counts were recorded, so that the data would not depend on the magnitude of the administered dose. Scintigraphic images were obtained at 5, 30, 60, 120, 180 and 240 min after administration of the radiolabelled enema.

A semiquantitative analysis based on scintigraphic data from recorded images was used for assessing the uniformity of tracer distribution. For this purpose the image showing the maximum retrograde progression of tracer was chosen for each patient; regions of interest (created by a threshold mechanism to reduce the possible incidence of observer's bias in the identification of such regions) were then marked out, representing the various intestinal segments under examination: namely the rectum colon, sigmoid colon, distal descending colon, proximal descending colon, and transverse colon. For each segment the value of mean counts per pixel (count/pixel) was calculated, the values being expressed as percentage of total recorded radioactivity. In this way we obtained a meaningful description of tracer distribution in the various segments of the colon, independent of segment size—this being marked by considerable individual variability.

The scintigrams of each patient were analysed by three examiners who evaluated individually the anatomic site and timing of maximum progression and homogeneity of distribution.

Plasma levels of 5-ASA and Ac-5-ASA metabolite

Venous blood samples (6 mL each) were drawn from each participating subject before drug administration (time 0) and at 2, 4 and 6 h after administration. Each blood sample was collected in a heparinized test tube and centrifuged for 10 min at 3000 r.p.m. Next, the supernatant was subdivided into two equal portions in standard test tubes and stored at −20 °C until analysis.

Plasma levels of 5-ASA and Ac-5-ASA metabolite were determined by HPLC according to confirmed methods.[17]

The equipment used for chromatographic separation featured a Gilson HPLC pump, model 305; the detector was a Perkin Elmer 3000 spectrofluorimeter.

Assessing tolerability

The tolerability of the new pharmaceutical dosage form of 5-ASA was assessed by assembling all adverse reactions voiced spontaneously by patients or detected by the physicians; enema retention times were also noted.

Informed consent

The study was approved by the ethical committee and was conducted according to the Declaration of Helsinki. All patients provided informed consent before participating in the study.

RESULTS

Twelve out-patients (9 males, 3 females) aged between 30 and 69 years (mean age ± s.d.: 46.92 ± 12.34) with mild to moderate ulcerative colitis entered the study. Ulcerative colitis was defined by the criteria of Truelove & Witts.[16] Disease activity and extent were assessed by colonoscopy. Of the 12 patients, five had mild and seven had moderate inflammatory activity. In six patients the diseased area extended up to the sigmoid, in five it extended to the descending colon and in one to the transverse colon.

All patients were able to complete the scintiscanning procedure and venous blood sample collections without any inconvenience.

Table 1 shows for each patient the intestinal segment where maximum retrograde progression of tracer was detected, and the timing of such peak values (patients were grouped according to disease extent). The radioactive enema progressed beyond the sigmoid colon in 11 out of 12 patients (92%) and to the transverse colon in 6 out of 12 patients (50%). Thus, relative to the extent of disease and hence to the possibility of actual topical action by the product, we can say that the tracer enema reached the sites of ulcerative colitis lesions in all patients (12/12).

The retrograde progression of tracer enema was completed within 4 h of administration, with peak

Table 1. Maximum spread of ^{99}Tc-labelled mesalazine gel enema in 12 patients with left-sided ulcerative colitis

Patient	Disease Extent	Activity	Segment* and time (min) of maximum spread		Time(min) to progression up to splenic flexure
1	rectum – sigmoid	moderate	d.D	(240)	
3	rectum – sigmoid	mild	S	(240)	
5	rectum – sigmoid	moderate	p.D	(240)	
6	rectum – sigmoid	mild	T	(240)	30
9	rectum – sigmoid	mild	d.D	(180)	
10	rectum – sigmoid	moderate	T	(180)	30
2	distal descending colon	moderate	T	(180)	120
4	distal descending colon	mild	T	(240)	30
7	distal descending colon	moderate	d.D	(180)	
8	distal descending colon	moderate	d.D	(180)	
11	distal descending colon	mild	T	(180)	120
12	transverse colon	moderate	T	(240)	60

*S, sigmoid; d.D, distal descending; p.D, proximal descending; T, transverse.

progression being detected at 180 min in six patients and at 240 min in the other six.

The time needed for retrograde progression to reach the splenic flexure in the six patients in which the tracer reached the transverse colon was 30 min in three patients, 60 min in one patient, and 120 min in two patients.

With respect to the distribution of tracer enema in the several segments of colon considered (rectum, sigmoid, distal descending colon, proximal descending colon and transverse colon), semiquantitative analysis demonstrated a fairly uniform distribution of radioactivity, with a significant percentage of enema also present in patients showing retrograde progression to the proximal descending and transverse colon (patient nos. 2, 4, 5, 6, 10, 11 and 12) (Figures 1 and 2).

In none of the patients was there any backflow of enema fluid after maximum retrograde progression.

Table 2 shows the mean values for 5-ASA and Ac-5-ASA plasma levels at 0, 2, 4 and 6 h after dosing.

Analysis of HPLC returns for tracer and its metabolite reveals low systemic absorption from the gut, with mean 5-ASA values of 0.9, 1.2 and 1.1 μg/mL, respectively, at 2, 4 and 6 h after dosing, and Ac-5-ASA values of 2.0, 2.6 and 2.8 μg/mL, respectively, at the same time points.

As regards tolerability, none of the participating patients complained of adverse reactions attributable

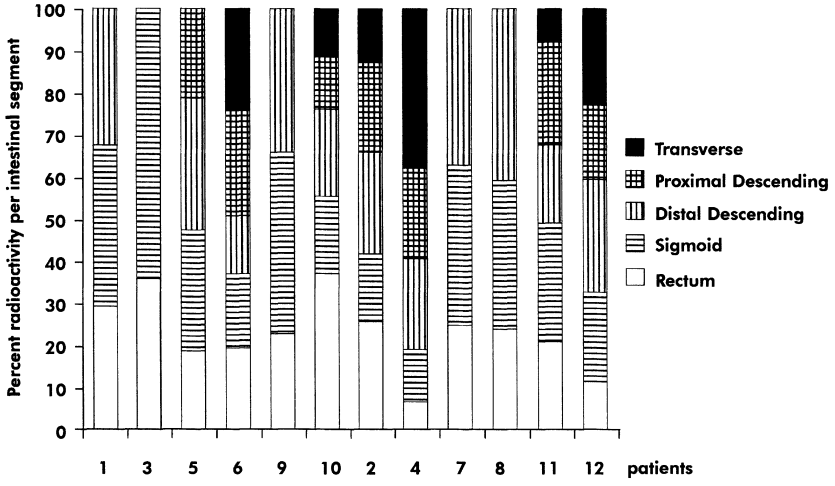

Figure 1. Intestinal segment of maximum spread and regional distribution of radioactivity (% dose) of the radiolabelled mesalazine enema at 4 h after dosing. For patients 1, 3, 5, 6, 9, 10, the disease extended to the rectum/sigmoid; for patients 2, 4, 7, 8, 11 to the distal descending colon and for patient 12 to the transverse colon.

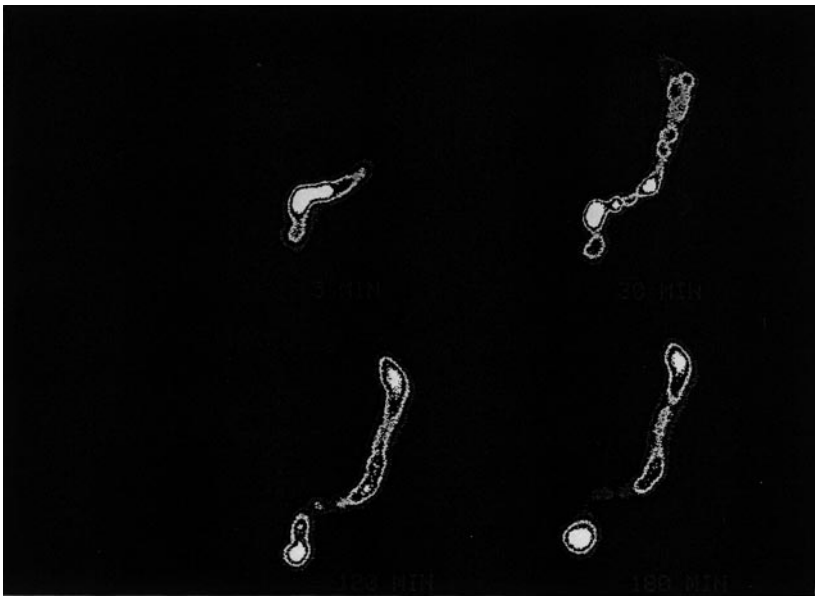

Figure 2. Scintigraphic images in patient number 6 after 5, 30, 120 and 180 min.

Table 2. Mean values (s.e.) of plasma levels of 5-ASA and Ac-5-ASA (μg/mL) in 11 patients, at 2, 4 and 6 h after administration of a single enema containing 4 g of mesalazine

Time (h)	5-ASA	Ac-5-ASA
0	0	0
2	0.9 (0.1)	2.0 (0.3)
4	1.2 (0.2)	2.6 (0.4)
6	1.1 (0.1)	2.8 (0.4)

to the study product; all patients were able to retain the enema completely throughout the study period.

DISCUSSION

Gamma scintigraphy is a reliable, non-invasive method for the evaluation of location and distribution of rectal enema. The extent of the spread, the homogeneity of distribution, and the length of persistence of the rectal preparation are all factors of great importance when treating ulcerative colitis patients topically.

In this study only patients with mild to moderate left-sided colitis were included, because these patients were more likely to be treated with topical mesalazine alone.

An overall analysis of scintiscan and systemic absorption data obtained with the formulation of mesalazine enema in gel form, supports the claim that this new pharmaceutical preparation form finds appropriate indication in the topical treatment of distal ulcerative colitis.

With respect to the degree of retrograde progression of tracer enema, we must note that the rectal gel enema reached beyond the sigmoid colon in 92% of test subjects, and reached the transverse colon in 50% of cases, thus displaying a good capacity for retrograde progression.

In particular, this new mesalazine preparation was seen to reach the sites of ulcerative colitis lesions in all patients, and in 8/12 patients it reached a segment beyond the upper limit of inflammation.

Top retrograde progression of tracer enema was detected within 3 or 4 h, thereby proving that the preparation was gradually and constantly present at lesion sites.

Concerning the data on homogeneous distribution, semiquantitative analysis revealed satisfactory homogeneity of tracer distribution in the various intestinal segments, including the more proximal segments.

In previous studies, activity and extent of disease have been shown to influence the spreading of the rectal enema into the colon: the more extensive and more active the disease, the greater the possibility of reaching the proximal portions of the colon.[13, 14, 18] The principal reasons for this are the decreased colonic tone and the absence of stools in the inflamed colon, both factors aiding the proximal spread of the enema.

In this regard our results, i.e. that the extent of the spread does not correlate either with disease extent or with disease activity, are in agreement with the findings

of Van Bodegraven and colleagues, who recently showed that disease activity is not an important determinant of retrograde colonic spread of mesalazine enemas.[19]

The observed plasma levels of 5-ASA and Ac-5-ASA obtained with the new preparation are consistent with those reported in the current literature;[2] the systemic absorption of the mesalazine enema was low. However, these results need to be confirmed with additional bioavailability studies.

An important finding to emerge from our study is that the new formulation is well tolerated: none of the patients developed any adverse reactions and all were able to retain the enema without difficulty throughout the study period. This is related to the fact that no gas is introduced into the colon and that the new rectal gel preparation, due to its physical properties, spreads more slowly and gradually.

In conclusion, this new gel mesalazine preparation may represent a further valid alternative for the treatment of active distal ulcerative colitis.

ACKNOWLEDGEMENTS

This study was supported by a grant from Crinos S.p.A., Como, Italy, who also supplied the new mesalazine rectal formulation.

REFERENCES

1 Campieri M, Bazzocchi G, Franzin G, *et al.* Treatment of ulcerative colitis with high dose 5-aminosalicylic acid enemas. Lancet 1981; ii: 270–1.
2 Campieri M, Lanfranchi GA, Boschi S, *et al.* Topical administration of 5-aminosalicylic acid enemas in patients with ulcerative colitis. Studies on rectal absorption and excretion. Gut 1985; 26: 400–5.
3 Thomson ABR. Review article: new developments in the use of 5-aminosalicylic acid in patients with inflammatory bowel disease. Aliment Pharmacol Ther 1991; 5: 449–70.
4 Sutherland LR, Martin F, Greer S, *et al.* 5-Aminosalicylic acid enema in the treatment of distal ulcerative colitis, proctosigmoiditis and proctitis. Gastroenterology 1987; 92: 1894–8.
5 Campieri M, Gionchetti P, Belluzzi A, *et al.* Optimum dosage of 5-aminosalicylic acid as rectal enemas in patients with active ulcerative colitis. Gut 1991; 32: 929–31.
6 Biddle WL, Miner PB. Long-term use of mesalamine enemas to induce remission of ulcerative colitis. Gastroenterology 1990; 99: 113–8.
7 Marshall JK, Irvine ES. Rectal aminosalicylate therapy for distal ulcerative colitis: a meta-analysis. Aliment Pharmacol Ther 1995; 9: 293–300.
8 Campieri M, De Franchis R, Bianchi Porro G, *et al.* Mesalazine (5-aminosalicylic) suppositories in the treatment of ulcerative proctitis or distal proctosigmoiditis. A randomized controlled trial. Scand J Gastroenterol 1990; 25: 663–8.
9 Campieri M, Paoluzi P, D'Albasio G, Brunetti G, Pero A, Barbara L. Better quality of therapy with 5-ASA colonic foam in active ulcerative colitis. A multicenter comparative trial with 5-ASA enema. Dig Dis Sci 1993; 38(10): 1843–50.
10 Williams CN, Haber G, Aquino GA. Double blind, placebo controlled evaluation of 5-ASA suppositories in active distal colitis and measurement of extent spread using ^{99m}Tc labelled 5-ASA suppositories. Dig Dis Sci 1990, 32(Suppl.): S71–5.
11 Wood E, Wilson CG, Hardy JG. The spreading of foam and solution enemas. Int J Pharm 1985; 25: 191–7.
12 Campieri M, Lanfranchi GA, Brignola C, *et al.* Retrograde spread of 5-aminosalicylic acid enemas in patients with ulcerative colitis. Dis Colon Rectum 1986; 29: 108–10.
13 Campieri M, Corbelli C, Gionchetti P, *et al.* Spread and distribution of 5-ASA colonic foam and 5-ASA enema in patients with ulcerative colitis Dig Dis Sci 1992; 37(12): 1890–7.
14 Wilding IR, Kenyon CY, Chauhan S, *et al.* Colonic spreading of a non-cholorofluorocarbon mesalazine rectal foam enema in patients with quiescent ulcerative colitis. Aliment Pharmacol Ther 1995; 9: 161–6.
15 Hay DJ, Sharma H, Irving HH. Spread of steroid containing foam after intrarectal administration. Br Med J 1979; 1: 1751–3.
16 Truelove SC, Witts LJ. Cortisone in ulcerative colitis. Br Med J 1955; 29: 1041–8.
17 Allgayer H, Ahnfelt NO, Kruis W, *et al.* Colonic N-acetylation of 5-aminosalicylic acid in inflammatory bowel disease. Gastroenterology 1989; 97: 38–41.
18 Hardy JG, Wood E, Clark AG, Reynolds JR. Colonic motility and enema spreading. Eur J Nucl Med 1986; 12: 176–8.
19 Van Bodegraven AA, Boer RO, Lourens J, *et al.* Distribution of mesalazine enemas in active and quiescent ulcerative colitis. Aliment Pharmacol Ther 1996; 10: 327–32.