LETTERS TO THE EDITOR

Colonic mucus and colitis

SIR,—I found the review on mucus and colitis by J M Rhodes (*Gut* 1989; **30**: 1660–6) most interesting, but I was surprised by the absence of several pertinent references. It must surely be relevant that about 8% of the general population show a constitutional lack of the O acetylated, sialidase resistant form of sialic acid.[1 2] These people do not appear to show an increased proneness to ulcerative colitis.[3] Patchy alteration in sialic acid structure (loss of O acetyl groups) is seen in hyperplastic epithelium in colitic biopsy material. Such mucosal alterations are likely to be secondary to inflammatory injury.[3] Thus mucin heterogeneity, whether genetically determined or acquired, appears to be unimportant in the aetiology of ulcerative colitis. Perhaps these observations should be added to the other negative findings catalogued by J M Rhodes. I suggest that they reduce the likelihood of a 'mucus/bacteria' hypothesis.

JEREMY R JASS
Department of Pathology, University of Auckland School of Medicine, Private Bag, Auckland, New Zealand

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Reply

SIR,—I am sorry that Professor Jass's excellent papers were not referred to in my leading article. This was, however, intended as a presentation of hypotheses to act as stimuli for further studies rather than as a comprehensive review of published work. As pointed out in the article a change in the O acetylation of mucin sialic acids is just one of many potential alterations in mucin structure that could have an effect on its interaction with bacteria. Others include alterations in sulphation, sialylation, length, and branching of oligosaccharide side chains and changes in the expression of carbohydrate receptors for bacterial adherence lectins.

J M RHODES
University Department of Medicine, University of Liverpool. PO Box 147, Liverpool L69 3BX

Scavenger effect of sulphasalazine (SASP), 5-aminosalicylic acid (5-ASA), and olsalazine (OAZ)

SIR,—We read with interest the paper by Williams and Hallett[1] on the action of SASP and 5-ASA on toxic oxygen metabolite production by neutrophils. The authors conclude by suggesting a scavenger effect is induced by both drugs, as previously reported.[2–4]

We have recently concluded a similar experiment, evaluating the influence of SASP, its metabolites (sulphapyridine and 5-ASA) and OAZ, on the generation of superoxide anions (O_2^-) by activated neutrophils and by a cell free xanthine-xanthine oxidase system. Human neutrophils were prepared from heparinised peripheral blood of healthy volunteers by using a combined dextrane/Ficoll-Hypaque separation procedure and hypotonic lysis to remove contaminating erythrocytes. The resulting cells (>95% neutrophils) were washed twice in phosphate buffer (pH 7·4) and then activated using 0·1 μg/ml of phorbol myristate acetate.

The production of O-radicals, generated by the catalysed reaction xanthine oxidase upon xanthine, was induced by incubating 0·05 IU/ml of dialysed xanthine oxidase in 100 mM k-phosphate buffer (pH 7·8) containing 0·1 mM EDTA and 0·5 mM xanthine.

O_2^- generation was measured either after spectrophotometrically reducing cytochrome c (cyt c) at 550 nm in a cuvette maintained at 37°C, or monitoring the luminol dependent light emission at 37°C on a Perkin-Elmer luminescence spectrophotometer.

For testing the scavenger effect of SASP and OAZ, we could not use the chemiluminesence method, because of its intense yellow colour in solution, which might have interfered with the light emission. We therefore used the reduction of cyt c assay for evaluating the effect of SASP, OAZ and also for sulphapyridine, but we could not use the same assay for 5-ASA. In fact 5-ASA caused a direct chemical reduction of cyt c effect, already reported by Neal *et al.*[5] Therefore, investigating the action of 5-ASA, we adopted the chemiluminescence method. In our study, 5-ASA, OAZ, and SASP produced a dose dependent inhibition of superoxide anions in both the neutrophils and cell free xanthine-xanthine oxidase system, 5-AWA being the most powerful (>50% of inhibition at 10 μM, the lowest concentration used). In contrast, sulphapyridine showed a dose dependent inhibitory effect on the cellular system, not modifying the activity of xanthine oxidase.

As can be seen, our data are only partly in agreement with the findings of Williams and Hallett. In our opinion, this study is not without certain methodological limitations. The authors, in their experiment, did not report the interference of SASP on light emission, because of its intense yellow colour in solution, therefore the inhibition on chemiluminescent response determined by SASP in their experiment could be partly attributed to the quenching effect on light emission by this drug. Finally, the authors did not highlight the direct chemical reduction provoked by 5-ASA on cyt c, as already mentioned. This, again, might determine a limitation of their results.

P GIONCHETTI, C GUARNIERI,* M CAMPIERI, A BELLUZZI, C BRIGNOLA, P IANNONE, M MIGLIOLI, L BARBARA
*Istituto di Clinica Medica e Gastroenterologia, *Dipartimento di Biochimica, Universita di Bologna, Italy*

1 Williams JG, Hallett MB. Effect of sulphasalazine and its active metabolite, 5-aminosalicylic acid, on toxic oxygen metabolite production by neutrophils. *Gut* 1989; **30**: 1581–7.
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Address for correspondence: Dr Paolo Gionchetti, Istituto di Clinica Medica e Gastroenterologia, Policlinico S Orsola, Via Massarenti 9, Bologna 40138, Italy.

Reply

SIR,—Gionchetti *et al*, although providing evidence which supports our contention that sulphasalazine and particularly 5-aminosalicylate are scavengers of a neutrophil derived oxygen metabolite, have questioned two methodological points in our paper.[1]

The first point raised was that the inhibition of luminol dependent chemiluminescence (LDCL) by sulphasalazine was because of its 'intense yellow colour', presumably by absorbing emitted photons, rather than by scavenging a luminol reactive molecule. We do not think the absorbance of sulphasalazine can account for our results for two reasons: (i) at the emission wavelength for luminol (425 nm) in our luminometer (maximum photon pathlength 0·8 cm) a concentration of sulphasalazine which inhibited the peptide-induced neutrophil LDCL by 50% (16·5 μM) would reduce detection of photons by a maximum of only 8%; (ii) absorbance of photons alone could not account for the differential effect of sulphasalazine on the LDCL response induced by the peptide f-met-leu-phe and phorbol myristate acetate (PMA) (Fig 1b). At high concentrations of sulphasalazine, however, its absorbance would be expected to cause interference. This probably accounts for part of the small inhibition of the PMA induced response we observed with 50 μM sulphasalazine (expected reduction in photon detection about 20%). The important point, however, is that absorption of photons cannot account for the inhibition by 5-aminosalicylate, as this compound would produce no significant reduction of transmission at concentrations which totally inhibit peptide induced LDCL.

The second point raised concerned the use of cytochrome c reduction as an assay of superoxide production. As cytochrome c readily accepts electrons, only the reduction of cytochrome c which can be inhibited by superoxide dismutase can be defined as being due to superoxide.[2] At the concentrations of 5-aminosalicylate we used (0·5 μM-50 μM) interference was not a problem. Neal *et al*[3] were unable to use 5-aminosalicylate in this assay but were using considerably higher concentrations of 5-aminosalicylate (up to 1000 μM). The widely recognised problems with cytochrome c reduction[4] led us to measure oxygen consumption also and so determine whether regeneration of oxygen as a result of superoxide scavenging had occurred. As we detected no inhibition of oxygen consumption (nor did Neal *et al*[3] in concentrations up to 1000 μM) this confirmed our conclusion that 5-aminosalicylate did not scavenge superoxide. We therefore suggested that the inhibition of LDCL by 5-aminosalicylate resulted from reaction with another oxygen metabolite which triggers luminol chemiluminescence, namely hypochlorite.[5] We have also produced more direct evidence for this. In chemically generating hypochlorite systems (xanthine/xanthine oxidase plus peroxidase and peroxidase plus hydrogen peroxide) or hypochlorite alone, the accompanying LDCL was inhibited by 5-aminosalicylate.[6] Furthermore, the fluores-

cence of 5-aminosalicylate decreased, suggesting a chemical reaction between 5-aminosalicylate and hypochlorite. Activated neutrophils also produced a peroxidase-generated oxygen metabolite which reacted with 5-aminosalicylate to produce a non-fluorescent product.[6] We therefore propose that 5-aminosalicylate acts as a hypochlorite scavenger and also as a hypochlorite indicator. Identification and detection of neutrophil derived metabolites of 5-aminosalicylate in the faecal stream of patients receiving 5-aminosalicylate would thus provide evidence for the production of hypochlorite in the inflamed bowel and may also be useful for monitoring the progress of the disease.

M B HALLETT
J G WILLIAMS
L E HUGHES
University Department of Surgery, University of Wales College of Medicine, Heath Park, Cardiff, CF4 4XN

1 Williams JG, Hallett MB. Effect of sulphasalazine and its active metabolite, 5-aminosalicylic acid, on toxic oxygen metabolite production by neutrophils. *Gut* 1989; **30**: 1581–7.
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British Digestive Foundation

SIR,—In an effort to increase public awareness of gastrointestinal diseases as well as the profile of the British Digestive Foundation and the research which it supports, we have begun a service to the media which provides information on new developments in research and British gastroenterology. The information published in *Inside Out – the British Digestive Foundation News Service* will be accompanied by comment from one of the foundation's medical advisors with the aim of putting the findings in context. Already some medical correspondents have welcomed the idea.

The editors of *Gut* and *Alimentary Pharmacology and Therapeutics* are allowing us to see proofs of articles before publication to enable us to select work of particular interest. However, it is impossible to make such arrangements with all of the medical journals in which we publish our work and I would therefore encourage you to approach me directly when you are about to publish a piece of work of special interest. This is just one of the ways in which the British Digestive Foundation is working to increase its impact and fund raising activities in the United Kingdom, but one in which we can all participate and hopefully ultimately benefit. Please contact me in writing at:

Department of Gastroenterology,
St Bartholomew's Hospital,
West Smithfield,
London EC1A 7BA.
Tel: 071 601 8508/9
Fax: 071 601 8510.

MICHAEL J G FARTHING
Scientific Publications Press Officer, British Digestive Foundation

BOOK REVIEWS

Splanchnic ischaemia and multiple organ failure. By A Marston, G F Bulkley, R G Fiddian-Green, and U H Hagland. (Pp 405; illustrated; £65.) London: Edward Arnold, 1989.

On the fringe or in the centre? This is a book that inevitably crosses conventional disciplines and functional systems. The role of the splanchnic circulation in the aetiology of multiple system organ failure and the effect of shock on gut blood flow are important and topical issues. This is a subject that unites the resources and endeavours of a variety of clinicians and scientists. The subject is both bold, because it crosses so many boundaries, and timely. The contents of this book will be of importance to anaesthetists and intensive care specialists, microbiologists, transplant surgeons, and renal physicians, immunologists, intestinal and vascular surgeons, clinical physiologists and those involved in the management of major trauma and cardiac surgery. Central to the theme of this text is the arachidonic acid pathway, protease inhibitors, free oxygen radicals, xanthene oxidase and superoxide dismutase, tumour necrosis factor, gut microflora and endotoxins. Other factors include endothelial and intestinal permeability as well as platelet, macrophage and neutrophil function. The subject is bold, painted in some detail and on a large canvass, it will have appeal to a wide audience and most I suspect will conclude that the result has been a success, particularly at the price.

In a decade that has seen the establishment of cardiac and hepatic transplantation, it is inevitable that clinicians engaged in intensive care management have recognised the central role of the gut as a determinant of outcome in multiple organ failure. A comprehensive book with contributors from a panel of international experts on this subject is timely. The editors must be congratulated on their choice of contributors who represent expertise from North America and Europe as well as their avoidance of unnecessary duplication. Most chapters are clear, succinct and well illustrated and are comprehensively referenced.

The book is divided into four sections. The first is concerned with anatomy and physiology of the splanchnic circulation. For most readers the second section of pathophysiology will be the most important and unique component of the book. This deals with the effect of splanchnic vascular occlusion and shock on intestinal perfusion followed by a detailed account of local and systemic mediators of intestinal ischaemia. The contributions on free radicals, mucosal cytoprotection, proteases, enteric bacterial toxins, bacterial translocation, endotoxin and eicosanoids are impressive for their clarity, quality of information and interest even to the non-specialised reader. The role of the Kupfer cell in protecting against endotoxaemia will no doubt be addressed in the future by studies in the anhepatic recipient during liver transplantation. The third section is concerned with an account of clinical syndromes resulting from intestinal ischaemia: stress ulceration, ischaemic hepatitis, pancreatitis, and cholecystitis as well as the consequences of large and small vessel occlusion in the small and large bowel. These chapters are comprehensive but unlike the earlier section may be found elsewhere in the surgical literature. Finally, there is an account of multiple organ failure syndromes which describe the role of monitoring and therapeutic options available for clinical management.

Although gut ischaemia is the name of this pitch, on the boundary there are aspects dealing with the pathogenesis of inflammatory bowel disease, hepatic failure, and mucosal protection which will be of immense interest to gastroenterologists as well as surgeons.

Unlike many books I have reviewed, this one will not gather dust; there are exciting concepts here which many clinicians will want to pursue. This subject is not on the fringe, the gut has proved to be the central pivot in the shocked patient; we need to learn more about it.

M R B KEIGHLEY

Therapy of inflammatory bowel disease. New medical and surgical approaches. By Mark A Peppercorn. (Pp 312; illustrated; $150.) New York: Marcel Dekker, 1990.

Mark Peppercorn, who has contributed many important controlled studies to the published work on inflammatory bowel disease, edits this new volume predominantly concerned with drug and nutritional treatment of inflammatory bowel disease. A pot pourri of six additional chapters has been included (alternatives to ileostomy, extraintestinal and liver complications, pregnancy and nursing, paediatric inflammatory bowel disease, psychosocial issues, and case studies). They distract the reader from the prime thrust of this book and could be deleted with advantage from the second edition.

The book's subtitle 'New medical therapy' is misleading since most contributions are excellent reviews but of well established treatment. Much of the information is already available in standard textbooks – for example, the third edition of Kirsner and Shorter *Inflammatory Bowel Diseases* – while new developments are well covered in such volumes as *Recent Advances in Gastroneterology*.

Does this book therefore provide new insight, new ideas, or novel views of treatment? Each chapter considers a specific drug and includes pharmocokinetics and mechanisms of action. The opening chapter deals with oral and parenteral corticosteroids and is a good summary of their potential value and many side effects. The reference list is excellent.

Topical corticosteroids are dealt with only briefly. Information concerning newer drugs such as tixocortol pivalate, which induces local anti-inflammatory activity but has no systemic glucocorticoid activity, should have been expanded. The editor himself reviews the role of sulphasalazine, drawing together the pharmacokinetics, clinical action, drug interaction, and adverse reactions with a good summary of published work. The newer oral and topical aminosalicylates are extensively covered by Stephen Hanauer of Chicago – an excellent summary of the present situation. Burton Korelitz's advocacy of immunosuppressive treatment is well known and his contribution is a vigorous defence of his enthusiasm which perhaps extends beyond the evidence available from controlled clinical trials. For the European reader this chapter has a salutary ending – a seven page summary of potential benefits and hazards of the treatment which the potential patient has to read and sign indicating informed consent has been given before treat-