LETTERS TO THE EDITOR

Intestinal permeability

EDITOR,—We read with interest the paper by Dr Oriishi and colleagues (*Gut* 1995; 36: 891–6) investigating intestinal permeability and the immune response to enteric bacterial antigens in patients with inflammatory bowel disease. Their finding of an increased systemic concentration of antibodies to lipid A in patients with inflammatory bowel disease is in agreement with studies showing increased titres of antibodies to the endotoxin core,¹ to peptidoglycan-polysaccharide complexes,² and to a variety of enteric bacteria³ in these diseases. These studies offer indirect evidence of mucosal barrier dysfunction in patients with inflammatory bowel disease, which supports the more direct evidence provided by the measurement of systemic endotoxin concentration or by bacterial culture studies.^{1,4}

The study by Dr Oriishi and colleagues also provides confirmation that the pattern of antibody response to endotoxin antigens is different in patients with Crohn's disease from those with ulcerative colitis.¹ There is an increase in the systemic concentration of IgG to endotoxin core/lipid A in patients with Crohn's disease but not in those with ulcerative colitis. Systemic IgM concentration to endotoxin core/lipid A is not increased in either disease. With regard to IgA, we similarly found that the plasma concentration of IgA to the endotoxin core was increased (though not significantly) in patients with Crohn's disease (107.3 ± 20.5 median units) and ulcerative colitis (93.0 ± 24.2) in comparison with healthy controls (61.3 ± 15.7).⁵ We are unable, as yet, to explain these differences.

Their study is also interesting with regard to treatment for impaired gut barrier function. Lactulose has been shown to eliminate systemic endotoxaemia in a hapten induced rat model of colitis⁶ and had been suggested as treatment for patients with inflammatory bowel disease.^{6,7} The mechanism of the anti-endotoxin action of lactulose is not clear, as lactulose treatment did not have any significant effect on the faecal count of Gram negative bacteria or faecal endotoxin concentration in the experimental model of colitis.⁶ It has generally been assumed that lactulose is fermented rapidly by colonic bacteria and that colonic absorption after oral administration would be minimal.⁸ The study by Dr Oriishi and colleagues, however, suggests that there is significantly increased lactulose absorption in the presence of colonic inflammation. It is possible, therefore, that absorbed lactulose was effective as an antiendotoxin agent in experimental colitis by exerting a direct neutralising effect on systemically circulating endotoxin.^{6,9}

K R GARDINER
R J MAXWELL
B J ROWLANDS

Department of Surgery,
The Queen's University of Belfast,
Institute of Clinical Science,
Grosvenor Road,
Belfast BT12 6Bf

G R BARCLAY
The Blood Transfusion Service,
Edinburgh

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Reply

EDITOR,—We thank Mr Keith Gardiner and his colleagues for their comments.

We are aware of the anti-endotoxin action of lactulose. In our study, however, we used lactulose as a marker of intestinal permeability. Anti-lipid A antibody concentrations were not influenced by lactulose with an antiendotoxin action, because antilipid A antibody concentrations were measured just before lactulose administration.

Lactulose may be useful in both evaluation of disease activity and treatment in diseases with an increasing intestinal permeability and endotoxaemia, such as inflammatory bowel disease, alcoholism.¹ Particularly in the inactive phase of Crohn's disease, the continuous administration of lactulose may be interesting. We look forward to further study on this subject.

T ORIISHI
Second Department of Medicine,
Kurume University School,
67 Asahi-machi, Kurume,
Fukuoka 830, Japan

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Hydatid disease

EDITOR,—We read with interest the leading article by Dr D L Morris (*Gut* 1994; 35: 1517–8). We agree with the points concerning treatment of the hydatid disease, however, we are in disagreement with Dr Morris' statement 'there are two forms of echinococcus that affect the liver of humans, *E granulosus* and *E multilocularis*'.

Rausch and Bernstein in 1972 described a new species of echinococcus named *E vogeli*.¹ Furthermore *E vogeli* was found to be the aetiological agent of the hydatid disease in several patients from Colombia, Venezuela, Ecuador, and Panama, most of them showing hepatic involvement by the disease.² More recently, we had the opportunity to study nine patients with hydatid disease, seven of them

from the Brazilian Amazon region; eight of nine showed extensive involvement of the liver.³ Another study of six additional patients from the state of Acre (Amazon region) showed severe involvement of the liver (Meneghelli et al, unpublished data). All of these patients showed pathological findings that allowed us to establish the diagnosis of echinococcosis caused by *E vogeli*. Subsequently new cases of this neotropical hydatidosis have been detected in Brazil by Ferreira et al⁴ and by D'Alessandro et al (unpublished data). It seems that the disease has an extensive distribution in South America, mainly in the Amazon region. All patients we studied had extensive involvement of the liver making a surgical approach impossible. We found that albendazole is effective for the treatment of the disease.⁵ Thus we consider that we have enough evidence to say that there are at least three species of echinococcus that affect the liver of humans: *E granulosus*, *E multilocularis*, and *E vogeli*. Moreover there is a possibility, although rare, that another species, *E oligarthrus*, may also cause hepatic disease in humans.

U G MENEGHELLI
A L C MARTINELLI
M G VILLANOVA
Department of Internal Medicine,
Faculty of Medicine of Ribeirão Preto,
University of São Paulo, CEP 14049-900,
Ribeirão Preto, São Paulo, Brazil

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Gastric mucus viscosity and Helicobacter pylori

EDITOR,—The article in *Gut* by Markesich et al reports that *H pylori* infection does not cause reduction in the viscosity of human gastric mucus gel (*Gut* 1995; 36: 327–9). The findings contradict the results of earlier studies,^{1–3} and misquote the source of materials with which the work was carried out.^{1,2} Contrary to the authors' assumption, we never used commercially obtained mucin and the work included human gastric mucus, as well as its purified mucin.^{4,5} Furthermore, our assays of *H pylori* enzymatic activities were carried out with enzyme enriched protein fraction under well controlled conditions. These studies showed that *H pylori* through its mucolytic enzyme actions is capable of exerting a detrimental effect on gastric mucus gel viscosity.⁶

The viscosity data presented by Markesich et al were obtained directly on gastric juice samples from patients with and without *H pylori* infection, and do not provide any information on the mucolytic enzyme activities of the bacterium, nor for that matter on

the effect of other degradative enzymes present in the samples. Hence, the results obtained by Markesich *et al* are not comparable to that of ours.^{1,2,4-6} Studies with gastric juice performed by others,³ however, showed lower viscosity values for *H. pylori* positive patients than that of *H. pylori* negative patients. Also hydrophobicity measurement data⁷ totally argue against the conclusions reached by Markesich *et al*.

There may be several reasons for the different results obtained by Markesich *et al*. The most important are the lack of control for the peptic activity during the processing and the mode of mucus gel sample preparation for the measurements. As such, the 'mucus gel' samples obtained after the centrifugation in both cases represent only the undegraded mucus, as the degraded one would remain in the supernatant. Hence, the viscosity measurements on the materials prepared from patients with and without *H. pylori* infection were carried out only on the undegraded 'insoluble mucus gel'. No reference of any sort is made as to the sample concentration (for example, mg protein/ml). The conditions used for 'mucus gel preparation' (for example, centrifugation 10000 rpm for 15 min) would result in the insoluble fraction consisting of some undegraded mucus, cell debris, and other particulate matter. Consequently, this kind of material would give very high and erroneous viscosity values, as is the case in this study.

We would also like to point out that the study by Markesich *et al*, in contrast with their contention, represents typical in vitro experiments, and unless a method is devised to take the viscosity measurements directly in the stomach, the claim is not valid.

B L SLOMIANY
A SLOMIANY
V L N MURTY
J PIOTROWSKI
Research Center,
UMDNJ-NJDS, Newark,
NJ 07103, USA

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Reply

EDITOR,—Slomiany and coworkers not surprisingly are unhappy with the results of our evaluation of the viscosity of gastric mucus gel. We believe that we studied the real stuff. I am surprised that the objectors did not comment on, or attempt to deal with, the fact that data were internally consistent and changed with treatment. We apologise because upon

re-reading our manuscript it could be read that we implied that previous workers always purchased mucus in a bottle (*Gut* 1995; **36**: 327-9). We do contend that mucus obtained by instilling buffer into freshly dissected pig stomachs followed by filtering through a 0.45 µm filter, raising the pH to 9 for 30 minutes, dialysing it against distilled water, and then lyophilising it may not be comparable to the fresh mucus gel obtained from human beings.¹

Our goal was to try to test reality. Our detractors believed that we failed in that objective. Their approach has been to identify reality through careful reconstructive experiments using sophisticated biochemical techniques. Slomiany and coworkers are internationally recognised authorities in mucus biochemistry; our study was of mucus biology. Since our paper was published we have even found an earlier reference with similar findings that also give references to even earlier work.² Thus, as is often the case, we were not the first but instead extend previous findings in populations whose *H. pylori* status is defined. Time will tell whether God has been unkind to them or to Slomiany and coworker.

Finally, we would like to draw attention to the fact that the gastric mucus hydrophobicity story may be somewhat more complicated than it first seemed.^{3,4}

D Y GRAHAM
B S ANAND
Department of Medicine and
Division of Molecular Virology,
Veterans Affairs Medical Center and
Baylor College of Medicine,
Houston, Texas, USA

Correspondence to: Dr D Y Graham, VA Medical Center (111D), 2002 Holcombe Blvd, Houston, TX 77030, USA.

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Copper associated childhood cirrhosis

EDITOR,—The report of copper associated childhood cirrhosis by Horslen *et al* (*Gut* 1994; **35**: 1497-500) is of considerable interest. But I take issue with one small point.

The authors cite a case reported by Bartók *et al* in 1971¹ as an instance of idiopathic hepatic copper toxicosis. A glance at the

Figure 1: Edge of nodule. Granules resistant to distase digestion take the periodic acid-Schiff stain.

Figure 2: The granules mark for α_1 antitrypsin by immunostain.

Figure 3: At a similar site to Fig 2 rhodanine stainable copper is shown.

original publication arouses doubts whether another diagnosis might be correct. Early last year, through the courtesy of Professor J Ormos of the University of Szeged, I was sent a block of liver tissue from that case (see Figures). Stainable copper in large amounts was present, but other findings – using both immunohistochemical and standard techniques – confirmed my suspicion that the underlying disease was α_1 antitrypsin deficiency. This diagnosis had been recognised and published in the Hungarian literature²; I was not aware of this when I asked for tissue.

In any event, it is time that this patient's case no longer be invoked as an instance of non-Indian copper associated childhood cirrhosis or idiopathic hepatic copper toxicosis. Given parental consanguinity,^{1,2} she of course may have had both α_1 antitrypsin deficiency and another defect involving copper handling, but this is speculation.

A S KNISELY
Department of Pathology,
Children's Hospital of Pittsburgh,
Pittsburgh, PA 15213 USA

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Reply

EDITOR,—We are extremely grateful to Dr Knisely for this further information on the case reported by Bartók *et al*,¹ and we agree that this case should not now be considered as a case of copper associated childhood cirrhosis (CACC). The postmortem liver copper content in this child, now known to have α_1 antitrypsin deficiency, was between 2947 and 3564 µg/g dry weight (normal <50 µg/g dry weight) thus emphasising the need to distinguish between secondary and primary causes of copper accumulation.

Evidence is now available to suggest that the remaining cases categorised as CACC type II do not all represent a single condition. The child described by Adamson *et al*² has