LETTERS TO THE EDITOR

Absence of antibodies stimulating H_2-receptor mediated cyclic adenosine monophosphate (cAMP) production in peptic ulcer disease

SIR,—We read with great interest the paper by Burman *et al* (*Gut* 1991; **32**: 620–3). Using porcine gastric mucosal cells as an in vitro test system, the authors could not find any stimulatory effect on cyclic adenosine monophosphate (cAMP) production by sera or immunoglobulin (Ig) fractions of 57 patients with relapsing ulcer disease. As the authors reported, their results are in contrast with data obtained by De Lazzari *et al*.[1] This group found in duodenal ulcer patients antibodies stimulating cAMP production in enriched guinea pig parietal cells. Thus, De Lazzari *et al* have suggested that duodenal ulcer disease may be caused by gastric cell stimulating antibodies to H_2-receptors, which therefore are proposed as 'a new addition to the growing list of receptor antibodies in human diseases'.

By using another in vitro test system, we have also investigated the possible role of cAMP stimulatory antibodies in peptic ulcer disease. We tested the effects of Ig preparations on cAMP production in cultured human gastric tumour cells (HGT-1[2]; kindly provided by Dr C Gespach, INSERM U 55, Paris). These cells have H_2-receptors and are considered to be a useful tool for studies of cAMP mediated gastric acid secretion.[3,4] Igs were derived from sera of 36 peptic ulcer patients. The patients were classified as adequate (AR; n=16) and inadequate responders to ranitidine (IR; n=20) by intragastric pH monitoring.[5] Sera were not tested directly because of several undefined components which decreased cell viability from 91 to 52% after four hour incubation of the cells with 20% serum. IgG was isolated by column chromatography on protein G-sepharose and concentrated by microultrafiltration. Other proteins were removed and non-IgG was precipitated by ammonium sulfate (1·6 mol/l). The Igs were tested at concentrations of 4 (IgG) and 1 (non-IgG) mg protein/ml medium. HGT-1 cells were grown as monolayers and incubated for 10, 30, 60, 180, and 360 minutes with Igs in the presence of 1 mmol/l phosphodiesterase inhibitor (IBMX). Standard IgG (Behringwerke) was used as control. The total amount of cAMP was measured by radioimmunoassay (Amersham Buchler).

The basal value of cAMP production in HGT-1 cells was mean (SD) 10·7 (1·7) pmol/mg protein and was stimulated after 10 minutes exposure to histamine (10 µmol/l) to 80·4 (15·0) pmol/mg protein in all experiments (n=36). The stimulation could be blocked by the H_2-receptor antagonists cimetidine and ranitidine with IC50 values of 0·400 and 0·034 µmol/l, respectively, confirming the presence and specificity of H_2-receptors on this cell type.[6,7] No statistically significant stimulation of cAMP production could be obtained after incubation of HGT-1 cells with any Ig preparation tested (see Table).

Our results agree with those of Burman *et al*. In neither in vitro test systems was there evidence for cAMP stimulating antibodies in peptic ulcer patients. In addition, we conclude that antibodies to H_2-receptors do not cause an inadequate response to H_2-antagonists as has been assumed.[1] However, despite these results, it can not be totally ruled out that autoimmunological processes have a role in specific subpopulations of patients with peptic ulcer disease. Moreover, the differing results of Burman *et al* and our group on one hand and De Lazarri *et al* on the other may be caused by the different in vitro test systems used. In particular, De Lazzari *et al* failed to detect any cAMP stimulatory effects on antibodies, unless the parietal cell content in cell suspensions was at least 50%.

Cyclic adenosine monophosphate (cAMP) production (pmol/mg protein) in human gastric tumour cells HGT-1 after incubation with Ig preparations derived from sera of ulcer patients with adequate and inadequate responses to ranitidine. (Values, mean (SD))

	Time (min)				
	0	10	30	180	360
Control	11·8 (0·5)	12·2 (1·0)	12·6 (1·3)	12·8 (1·7)	11·5 (1·0)
Adequate responders (n=16):					
IgG	10·4 (1·7)	10·7 (1·7)	10·4 (2·2)	12·0 (2·1)	11·4 (2·2)
Non-IgG	10·6 (1·2)	13·0 (1·6)	13·1 (1·3)	11·9 (2·0)	11·8 (2·5)
Inadequate responders (n=20):					
IgG	10·1 (1·7)	10·5 (1·1)	10·1 (1·8)	11·8 (2·5)	11·0 (1·9)
Non-IgG	10·7 (0·9)	12·7 (1·0)	12·3 (1·6)	11·7 (2·4)	11·7 (2·0)

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Reply

SIR,—The letter of Drs A Sarem-Aslani, Ch Bergmann, S Walker, D Ratge, U Klotz, and H Wisser supports our paper recently published in *Gut*.[1] In contrast to a previous report by DeLazarri *et al*,[2] we could not find any evidence of gastric acid stimulatory antibodies in patients with severe ulcer disease. Our assay system of porcine gastric mucosal cells exhibited an approximately 20 fold increase in cAMP upon addition of 10^{-4} mol/l histamine. This system seems superior to that of DeLazzari *et al*, who used guinea pig gastric mucosal cells, which had to be enriched for parietal cells (at least 50%) in order to produce a measurable response, but then produced only a fivefold cAMP response to 10^{-3} mol/l histamine. As the results of the two studies were divergent, an argument of species specificity could be made. In this context, the data of Sarem-Aslani *et al*, using a human histamine responsive cell line is of great interest. The sensitivity of their system is comparable to that of ours and the conclusions are in total agreement. Thus, we can emphasise that although it is not possible to exclude histamine receptor stimulatory antibodies in exceptional cases, these are not a cause of severe ulcer disease in humans.

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Non-colonic symptomatology and the irritable bowel: is it really of diagnostic value?

SIR,—We read the paper by Maxton and colleagues on the diagnostic value of 'non-colonic' symptoms in the irritable bowel syndrome with interest.[1] We have some major concerns about their study, however, which we hope they can clarify.

Our first concern relates to the study design. No healthy controls were included for comparison; this makes it difficult to determine the clinical significance of 'non-colonic' symptoms, which are known to be common in otherwise healthy people.[2] The measurement of symptoms is also of concern because the reliability and validity of the symptom assessment was not documented. If the authors did not define and measure each of the symptoms carefully, these might have been interpreted in many different ways by the interviewer and the patients, and thus the discriminatory ability of