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Cimetidine Does Not Inhibit Indomethacin-Induced Small Bowel Ulceration in the Rat (40585)

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Indomethacin-induced small intestinal ulceration was first demonstrated by Kent *et al.* (1). They described the lesions as penetrating longitudinal ulcers on the mesenteric side of the distal jejunum and the ileum which progress to perforation and fatal peritonitis. Although the mechanism by which indomethacin produces these lesions of the small intestine is not known, the concomitant administration of antibiotics reduces the severity of the lesions (1) and the lesions do not occur in germ-free animals (2).

Various prostaglandin compounds have been shown to reduce significantly indomethacin-induced small intestinal damage (3). It is in this context that Robert first used the term "cytoprotection" (2).

Recently, the H_2 receptor antagonist, cimetidine, has been shown to reduce significantly gastric mucosal injury produced by intravenous (4) or intragastric (5) acetylsalicyclic acid. Since, in both of these studies, exogenous HCl was instilled into the stomach, cimetidine appeared to be exerting its protective effect by some means other than inhibition of acid secretion. Another group failed to demonstrate a protective effect of cimetidine against lesions produced by intragastric acetylsalicyclic acid plus HCl (6).

Since these earlier studies suggested that cimetidine might have a mucosal protective action additional to inhibition of acid secretion, the present study was undertaken to determine whether cimetidine inhibits indomethacin-induced small intestinal ulceration in the rat. 16-16 dimethyl prostaglandin E_2 ($DMPGE_2$), an agent of proven efficacy against this lesion, was used as a positive control.

Methods. Male Sprague–Dawley rats, 200 to 250 g body wt, were divided into four groups of 10. All drugs were administered by peroral gastric intubation during light ether anesthesia. All four groups received a single dose of indomethacin (20 mg kg^{-1}). In addition, one of the following treatments was randomly assigned to the four groups: 0.15 *M* NaCl (control), 80 mg kg^{-1} cimetidine, 400 mg kg^{-1} cimetidine, or 100 μg kg^{-1} 16-16 dimethyl PGE_2. The treatments were given twice, once 30 min before administration of indomethacin and again 8 hr after the administration of indomethacin. The rats were sacrificed by decapitation 16 hr after the administration of indomethacin. The small intestine, from pylorus to ileocecal valve, was removed and opened along the antimesenteric border. Luminal contents were washed away with 0.15 *M* NaCl. The length of each ulcer was measured to the nearest millimeter. The person scoring the lesions was given the intestines in random order and had no knowledge of which treatment an animal had received. Degree of ulceration was determined both by the number of lesions and the sum of the lengths of all the lesions per rat. Statistical evaluation was by the unpaired *t* test. Values are expressed as means plus or minus standard errors.

Solutions. Indomethacin (No. 1-7378 Sigma, St. Louis) was dissolved in 5% $NaHCO_3$, 10 mg ml^{-1}, then diluted with water 1:4 (v/v) to produce a solution which was isotonic at pH 8.0 and a concentration of 2.5 mg ml^{-1} which would allow 20 mg kg^{-1} to be given to each animal in a volume of 1.6 to 2.0 ml per rat.

Cimetidine (No. 92334, a gift of Smith, Kline and French Laboratories, Philadelphia) was dissolved in 0.5 *M* HCl, adjusted to pH 6.8 with 1 *M* NaOH and diluted with 0.15 *M* NaCl so that the final concentrations were 6.25 mg ml^{-1} (low dose) and 50 mg ml^{-1} (high dose) and the appropriate amount was administered in 1.6 to 2.0 ml per rat.

A stock solution of 1 mg ml^{-1} 16-16 di-

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methyl PGE_2 (U-37026, lot No. 13158-SAN-53F, oil; a gift of Dr. J. Pike, Upjohn Co., Kalamazoo), in absolute ethanol was added to 0.15 *M* NaCl (pH 7.0–7.4) in the amount required to allow administration of 100 $\mu g\ kg^{-1}$ to each animal in 2.0 ml.

Results. (Fig. 1). Control group: Each of 10 rats given 2.0 ml 0.15 *M* NaCl had lesions, with a range of 2 to 97 lesions per animal and 5 to 267 mm sum of length of ulcers per animal.

Effects of 16-16 dimethyl PGE_2 on intestinal lesions. 16-16 dimethyl PGE_2, as shown in Fig. 1, significantly ($P < 0.01$) reduced the number of lesions and significantly ($P < 0.01$) reduced the sum of lengths of ulcers. Three animals in this group ($N = 9$) had no intestinal lesions. The range of number of lesions per animal was 0 to 35 and the range of sum of lengths of ulcers was 0 to 101 mm. The results from one animal in this group which died soon after administration of indomethacin are not included.

Effect of cimetidine on intestinal lesions. Neither dose of cimetidine had a protective effect. Number of lesions per animal and sum of lengths of ulcers in cimetidine-treated animals did not differ significantly from controls. In the 80 $mg\ kg^{-1}$ cimetidine group, the range of number of lesions per animal was 11 to 98 and the range of sum of lengths of ulcers was 29 to 379 mm. In the 400 $mg\ kg^{-1}$ cimetidine group, only one animal had no intestinal lesions. The range of number of lesions per animal was 0 to 74 and the range of sum of lengths of ulcers was 0 to 328 mm.

Discussion. These data indicate that cimetidine had no effect on ulceration of the small intestine produced by oral indomethacin. We included the 16-16 dimethyl PGE_2 group to verify under the conditions of our study that these lesions could be inhibited by prostaglandins as had been shown in earlier studies (3).

Robert has shown (3) that administering 16-16 dimethyl PGE_2 30 min prior to or up to 1 hr following indomethacin affords protection in this model. Although it has been shown that only a single dose of 16-16 dimethyl PGE_2 is necessary to prevent these lesions, we chose to administer both agents 30 min prior to and 8 hr after the indomethacin so that high levels of the drugs would be present during the entire period of observation.

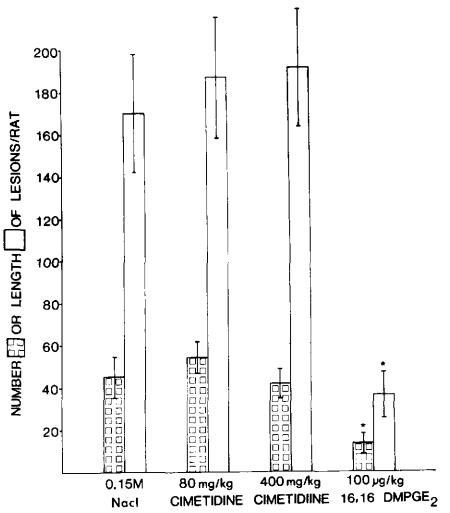

FIG. 1. Effect of cimetidine, 80 and 400 $mg\ kg^{-1}$, and 16-16 dimethyl PGE_2, 100 $\mu g\ kg^{-1}$, on mean ± SE number of small intestinal lesions and sum of lengths of ulcers compared to the control group. There were 10 animals in each group except in the 16-16 $DMPGE_2$ group in which there were 9. Statistical analysis by unpaired *t* test. (*$P < 0.01$).

The administration of a single dose of indomethacin to rats produces both gastric mucosal erosions which appear within 3 hr (7) as well as lesions in the small intestine which are more delayed in development. Since indomethacin and acetylsalicylic acid inhibit prostaglandin biosynthesis (9) and since exogenous prostaglandins protect against both gastric and intestinal lesions, this suggests that these two lesions might share, at least in part, a common mechanism of pathogenesis. Conversely, the observation that cimetidine protects against gastric but not intestinal lesions indicates that there are also differences in their mechanisms of formation.

Summary. In earlier studies, cimetidine was found to protect against acetylsalicylic acid-induced gastric mucosal lesions by a mechanism other than inhibition of gastric acid secretion. The present study examined the possibility that cimetidine might protect against the intestinal mucosal injury produced by indomethacin.

Small intestinal ulceration produced by oral indomethacin (20 $mg\ kg^{-1}$) was studied in rats treated with 0.15 *M* NaCl (control); 80

mg kg^{-1} cimetidine; 400 mg kg^{-1} cimetidine; and 100 $\mu g\ kg^{-1}$ 16-16 dimethyl prostaglandin E_2. The treatments were given 30 min before and 8 hr after indomethacin. Confirming previous studies, 16-16 $DMPGE_2$ significantly ($P < 0.01$) reduced the number of lesions and the sum of the lengths of ulcers per rat when compared to the control group. Neither dose of cimetidine had any effect on the intestinal lesions.

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