THE PROTECTIVE ROLE OF DILTIZEM AND PROSTAGLANDIN E2 ON STREES ULCER IN RATS

(Received 18 April, 1995)

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ABSTRACT

Objective: This study was devised to investigate the effects of prostaglandin E2 and diltizem on stress ulcer in rats.

Methods: Twenty-one rats were used in the study. The effects of immobilization stress was examined at the stomach and jejunum, after surgical removal. The ulcer index, total acid, pH and mast cell counts were done. The first group was stressed without premedication to serve as the control. The second group received diltizem (Diltiazem 0.25 mg/Kg) and the third group received prostaglandin E2 (Dinoproston 50 microgram/Kg) before stress.

Results: The diltizem and the PGE2 pretreated groups both had significantly lower mean ulcer indexes compared to the control group ($p<0.001$). Similarly the total acid of the diltizem and the PGE2 pretreated groups were lower ($p<0.001$, $p<0.001$). The pH of the diltizem and the PGE2 pretreated groups were higher ($p<0.001$, $p<0.001$) and the rise in the PGE2 group was more pronounced than the diltizem group ($p<0.001$). Mast cells were observed in the diltizem and PGE2 groups but not in the control group.

Conclusion: PGE2 and diltizem was found to provide protection to the gastric mucosa against the erosive complications of stress and furthermore PGE2 was more effective than diltizem.

Key words: Stress ulcer, Diltizem, Prostaglandins.

INTRODUCTION

Patients with shock, sepsis, burn, respiratory insufficiency and head trauma are at particular risk for acute mucosal ulcers. These patients with bleeding ulcers have a higher incidence of fatal outcome and therefore prevention is of utmost importance. Although cimetidine and antacid have high freguency of complications, they are used for such purposes (1,2).

In this experimental study, we investigated the effects of diltizem and PGE2 in order to prevent stress ulcer formation.

MATERIALS AND METHODS

Twenty-one female (180-220 g) Wistar albino rats were used in the study at Haydarpaşa Numune Hospital laboratories. Rats were first anesthetized with ether following a 12 hour fasting. Later, they were stressed according to Brodie's protocol (3) by immobilizing them on T-rods 20 cm high by gently strapping 4 feet as not to labor breathing.

Rats were immobilized in all 3 categories for 24 hours. The first group received intraperitoneal (physiological) saline before and 12 hours following stress, the second group received diltizem (Diltiazem 0.25 mg/Kg Mustafa Nevzat) before and 12 hours following stress and the third group received 50 microgram/Kg PGE2(Dinoprostone 0.5mg-Organon) via oral-gastric lavage before and 12 hours following stress.

After stress administration, laparatomies were done. The esophagus and the second portion of the duodenum were ligated for one hour and the contents were collected for acid output and pH analysis. The acidity and pH evaluations of the gastric contents were done at the biochemistry department utilizing Merk Art 9526 Indicator papers.

Later, following gastrectomy and partial jejunectomy, all animals were killed. Their stomachs were incised along the longer-curvature. This procedure was completed in a single-blinded fashion by an independent pathologist. Petechias were counted and a group of 5 petechias were considered a 1 mm ulcer. All 3 groups were indexed according to their ulcer involvement. The mesenteric section of jejunums were incised and fixed in a 10% formaldehyde solution together with other tissues. All the gastric and jejunal specimens were sent to the pathology laboratories for mast cell and histopathologic investigations. The gastric and the jejunal mesenteric specimens were dyed with 1% toludin blue. They were examined under microscope at 400xmagnification for positive-stained cells. The total number of cells were counted at ten different sites and averages were calculated.

The results were analyzed by Wallis variant analysis (Statistical significance $p<0.001$).

RESULTS

The diltizem and the PGE2 pretreated groups both had lower mean ulcer indexes compared to the controls ($p<0.001$, Fig. 1).

The total acid of the diltizem and the PGE2 pretreated groups were lower ($p<0.001$, $p<0.001$ respectively) (Fig. 2).

The pH of the diltizem and the PGE2 pretreated groups were higher ($p<0.001$, $p<0.001$, respectively). The rise in pH for the PGE2 group was more pronounced than the diltizem group ($p<0.001$, Fig. 3).

The mucosal integrity of the control group was destroyed which was evidenced by epithelial damage and necrosis (Fig. 4). The mucosa of the diltizem group only had minimal mucosal desquamation, muscularis mucosal congestion and edema. The PGE2 pretreated group had an increase in the epithelial thickness with normal sub-epithelial glands and minimal polymorphonuclear leukocyte infiltration and edema.

The mast cell numbers were decreased in damaged tissues whereas they were found to be more abundant in undamaged mucosal tissues. Mast cells were observered in the diltizem and the PGE2 groups

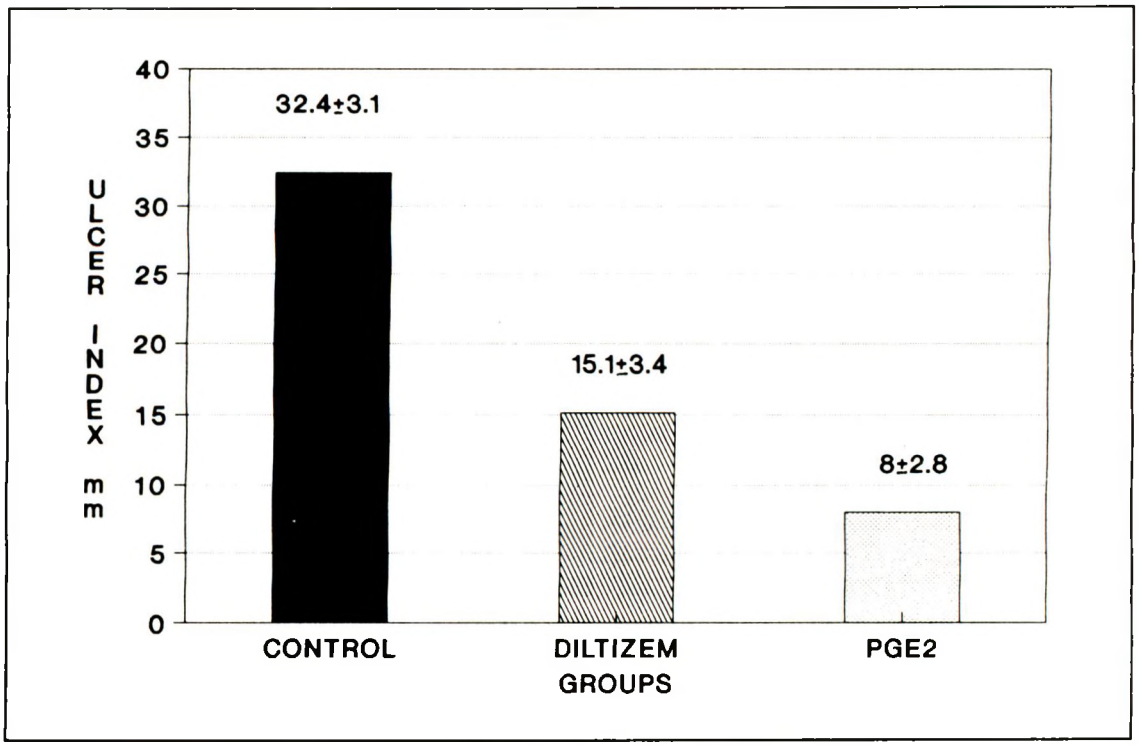

Fig. 1: *The values of mean ulcer index of control, diltizem and PGE2 groups.*

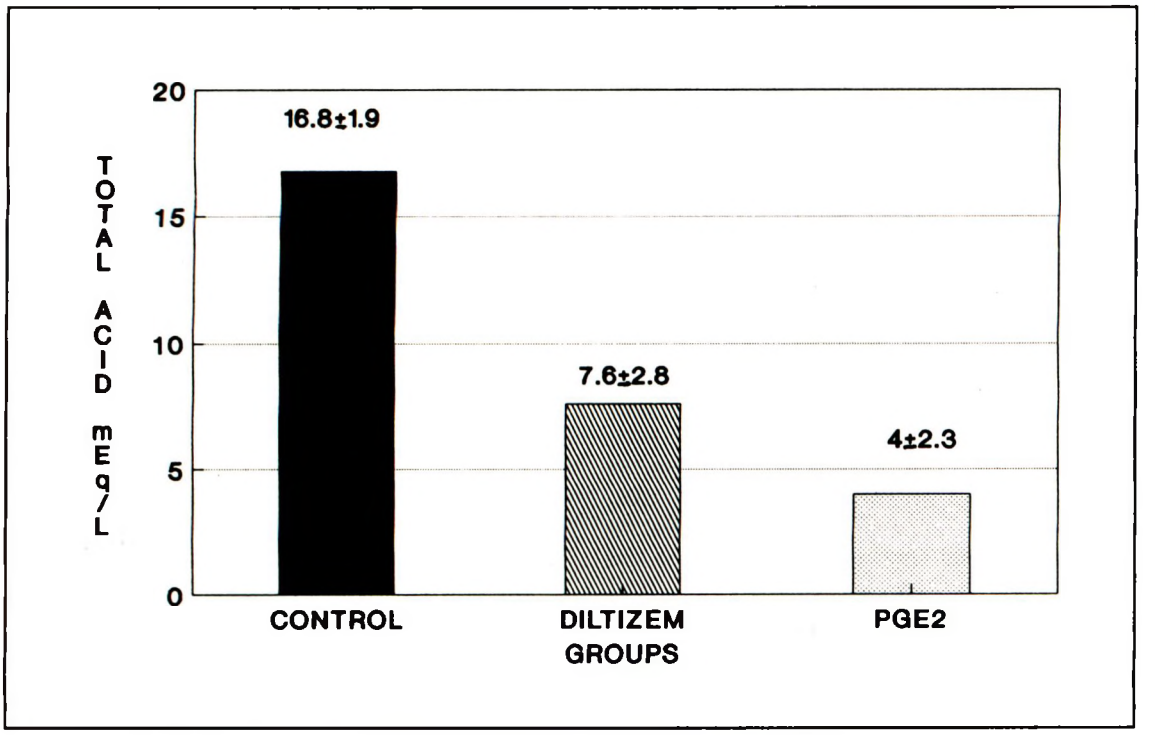

Fig. 2: *The values of total acid output of control, diltizem and PGE2 groups.*

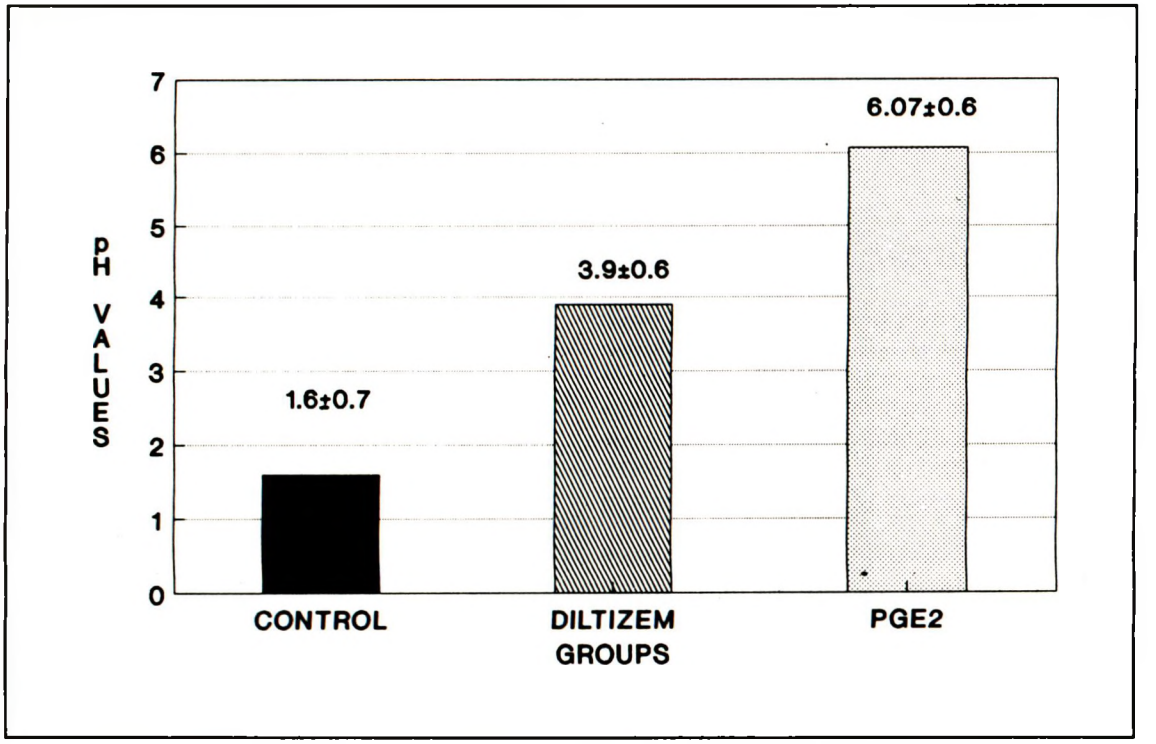

Fig. 3: *The values of pH of control, diltizem and PGE2 groups.*

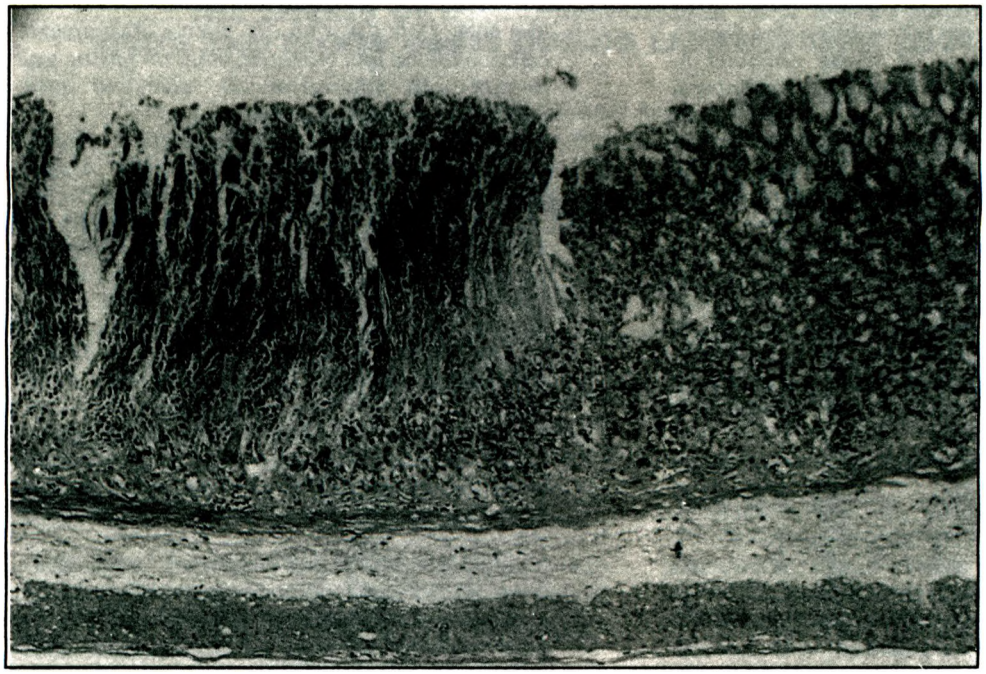

Fig. 4:
Acute Gastric Ulcer (HEx40) At the left side of photograph, bleeding and necrosis are easily seen in the whole mucosa layer.

but not in the control group. The average numbers of mast cells in the diltizem and the PGE2 groups were similar ($p>0.05$).

DISCUSSION

Acute mucosal erosions usually occur by drugs, acid hypersecretion and/or the loss of mucosal integrity (4-6). Antacids and cimetidine were found to be effective in controlling the mucosal erosion but with minimal protection against mucosal lesions (1,2,7). The exact mechanism of stress-related gastric lesions in rats is still unknown but it is postulated that the increase in the gastric contractility leading to a decrease (8,9) in hemo-perfusion is an etiologic mechanism (10,11). Yeğen et al (12) found that, calcium channel blockers such as nicardipine and verapamil prevented stress ulcer formation. Verapamil, an organic calcium channel blocker, was postulated to decrease the mast cell degranulation (9,13), gastric acid secretion and motility (14,15). Al-Mashhadani et at (16) found a considerable decrease in mast cell numbers in mucosa, submucosa and musculature of rats under stress and gastric ulcer has occurred in ninety percent of cases, In the same study the mucosal mast cell numbers were higher than in the nifedipine pre-treated rats which had undamaged gastric mucosa. We also observed an increase in mast cell numbers in gastric tissues of rats which were treated by the calcium channel blocker, diltizem. Vagal fibers are postulated to be responsible for mast cell degranulation and stress ulcer formation (17).

Although the role of gastric acid in the formation of ulcers were claimed to be minimal in rats by some investigators (18) Ogle et al (9) found the anti-acid therapy to have a protective role in prevention of ulcers. Our results are in accordance with Ogle's studies to disclose a significant correlation between the amount of acid and the ulcer formation.

Prostaglandins are the endogenous substances which are synthesized in almost all tissues (19). It has been shown that PGE2 and PDI, produced by the gastric mucosa, are important prostaglandins which may prevent the gastric mucosal injury (20). It is reported that prostaglandins increase the gastric mucosal blood flow, secretion of mucus and bicarbonate and increase c-AMP level and also stabilize the sodium pump (21). We found that the group which have taken PGE2, had significantly lower values of ulcer index and total acid output than the values of control group. Mast cells were found in high amount in PGE2 group. We noticed that PG protected gastric mucosa as in other studies (19, 21-23).

As a conclusion, PGE2 and diltizem protect gastric mucosa from induced injury. PGE2 has a stronger effect than diltizem.

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