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Effects of omeprazole on the number of immunoreactive gastrin- and somatostatin-cells in the rat gastric mucosa

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Summary. The effects of omeprazole - an inhibitor of gastric acid secretion - on gastrin (G)- and somatostatin (D)-cell density in the gastric antral mucosa epithelium in rats were examined, following a 5-day treatment. It was found that omeprazole increased the density of G-cells, whereas it decreased the density of D-cells. That effect was probably independent of hypergastrinaemia, since it could not be blocked by a simultaneous treatment with proglumide - a gastrin receptor blocker. It is concluded that the observed phenomenon is a direct result of a lower gastric acidity, as a consequence of omeprazole treatment.

Key words: Gastric antral mucosa, Immunohistochemistry, G- and D-cells, Omeprazole, Cell density

Introduction

It is well known that gastric acidity influences gastrin and somatostatin secretion. The antral acidification is a potent inhibitor of gastrin release (Jackson et al., 1972), whereas alkalization of antral lumen has been demonstrated to increase gastrin secretion (Becker et al., 1974). Elevated gastrin levels and an augmented number of gastrin-producing cells have been found in human subjects with achlorhydria (Polak et al., 1978; Webb et al., 1985).

Gastric acidity is also known to stimulate somatostatin secretion. It has been demonstrated that intragastric administration of acid to dogs increase somatostatin release into circulation (Schusdziarra et al., 1978). Perfusion of the mouse stomach with exogenous acid causes an elevation in somatostatin secretion in proportion to the increase in luminal acidity (Schubert et al., 1988). The basal, as well as the postprandial, somatostatin levels are lower in patients with achlorhydria, when compared to healthy subjects (Webb et al., 1985).

In the present study, we have applied omeprazole (OM); a benzimidazole derivative. This agent acts as an inhibitor of H+, K+-ATPase and, in consequence, depresses gastric acid secretion in different mammalian species, including the rat (Larsson et al., 1983). Omeprazole administration evokes a long-lasting, continuous hypergastrinaemia (Hakanson et al., 1986; Ryberg et al., 1988). Trophic effects of OM on the gastric, but not on the intestinal mucosa, have also been observed (Sundler et al., 1986; Tielemans et al., 1989).

In this investigation we have attempted to estimate the number of immunoreactive gastrin (G)- and somatostatin (D)-cells in the rat gastric mucosa, following OM treatment. In order to evaluate the role of OM-induced hypergastrinaemia, we have applied proglumide, i.e., gastrin receptor blocker. Additionally, we have investigated the effects of Sandostatin (SMS 201-995) - a long-lasting somatostatin analogue.

Materials and methods

Adult male rats of the Wistar strain, weighing 150 ± 10 g each, were used in the study. The animals were kept in a room with controlled illumination (14 h light: 10 h darkness) and temperature (22 ± 2° C) and had a constant access to granulated food (delivered by the Animal Food Manufacture in Motycz, Poland) and to tap water.

The following preparations were used in the study: omeprazole (OM; AB Hässle, Molndal, Sweden), proglumide (PRO; Milid, Rotta Research Laboratory, Milano, Italy), and SMS 201-995 (Sandostatin; Sandoz AG, Basel, Switzerland). The animals were divided

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into six groups, as follows (the doses per animal per single intraperitoneal injection are specified): Group I - controls, n = 7; Group II - OM, 40 μM/kg BW, n= 7; Group III - PRO, 80 mg/kg BW, n = 6; Group IV - SMS 201-995, 100 μg/kg BW, n = 6; Group V - OM + PRO, in doses as above, n = 7; Group VI - OM + SMS 201-995, in doses as above, n = 6. The injections were administered in 0.2 ml volumes, twice daily for 5 days, except for SMS 201-995, which was injected twice on the 5th day of the experiment only. The animals were sacrificed by decapitation. Fragments of the antral parts of stomach were collected and fixed in Bouin's fluid.

Paraffin sections (6 μm thick) were used for immunohistochemical detection of gastrin and somatostatin. For demonstration of gastrin and somatostatin - K 516 and K 512 DAKO PAP Kits were used, respectively. The microscopic preparations were counterstained with haematoxylin. The number of cells exhibiting positive reaction for gastrin (G-cells) and somatostatin (D-cells) were counted by light microscopy, in 50 fields of vision (x400 magnification). The results were expressed as a mean number of immunoreactive cells per one field of vision. In Group I (controls) and Group II (OM) blood samples were collected and serum gastrin levels were measured by the RIA method with the use of INC Biomedicals Inc. Kit (Carson, CA, USA).

Statistical evaluation of the results was performed with the use of the one-way analysis of variance (ANOVA). The significance of differences, observed between mean numbers of G- or D-cells per one field of vision in individual groups, was subsequently determined by the Newman-Keuls test (Hinkle et al., 1979). The differences in serum gastrin levels were analyzed by means of the Cochran-Cox C-test.

Results

Serum gastrin levels

Serum gastrin levels in rats treated with OM were significantly higher than those in controls (Group II: 372.2 ± 36.2 pg/ml vs. Group I: 212.7 ± 6.0 pg/ml, $p < 0.005$).

Immunoreactive gastrin cells (G-cells)

The administration of OM caused a significant increase in the mean number of G-cells per one field of vision in the gastric antral mucosa epithelium (Fig. 3A). This increase was not blocked by the administration of proglumide, but was significantly suppressed by the simultaneous treatment with SMS 201-995. Both proglumide and SMS 201-995, when administered alone, did not exert any significant effect. However, a slight tendency towards a lower density of G-cells could be noticed, following the administration of SMS 201-995 (Fig. 1).

Immunoreactive somatostatin cells (D-cells)

Omeprazole significantly decreased the mean number of D-cells per one field of vision in the rat gastric antral mucosa epithelium (Fig. 3B). This effect was neither influenced by SMS 201-995, nor affected by proglumide, although both SMS 201-995 and

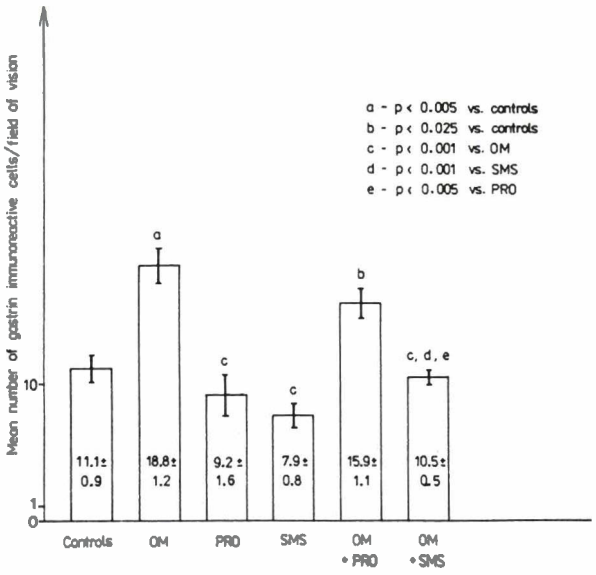

Fig. 1. The mean number of G-cells per one field of vision in the examined groups of rats. × 400. p - level of significance; OM - omeprazole; PRO - proglumide; SMS 201-995 - Sandostatin. Bars represent mean ± SEM.

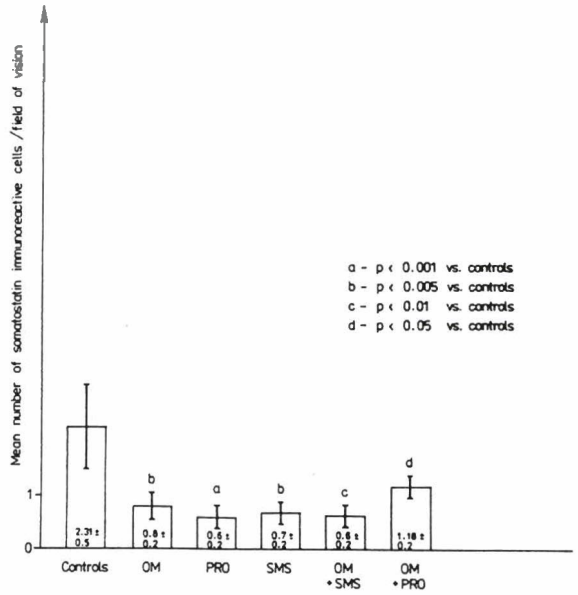

Fig. 2. The mean number of D-cells per one field of vision in the examined groups of rats. × 400. p - level of significance; OM - omeprazole; PRO - proglumide; SMS 201-995 - Sandostatin. Bars represent mean ± SEM.

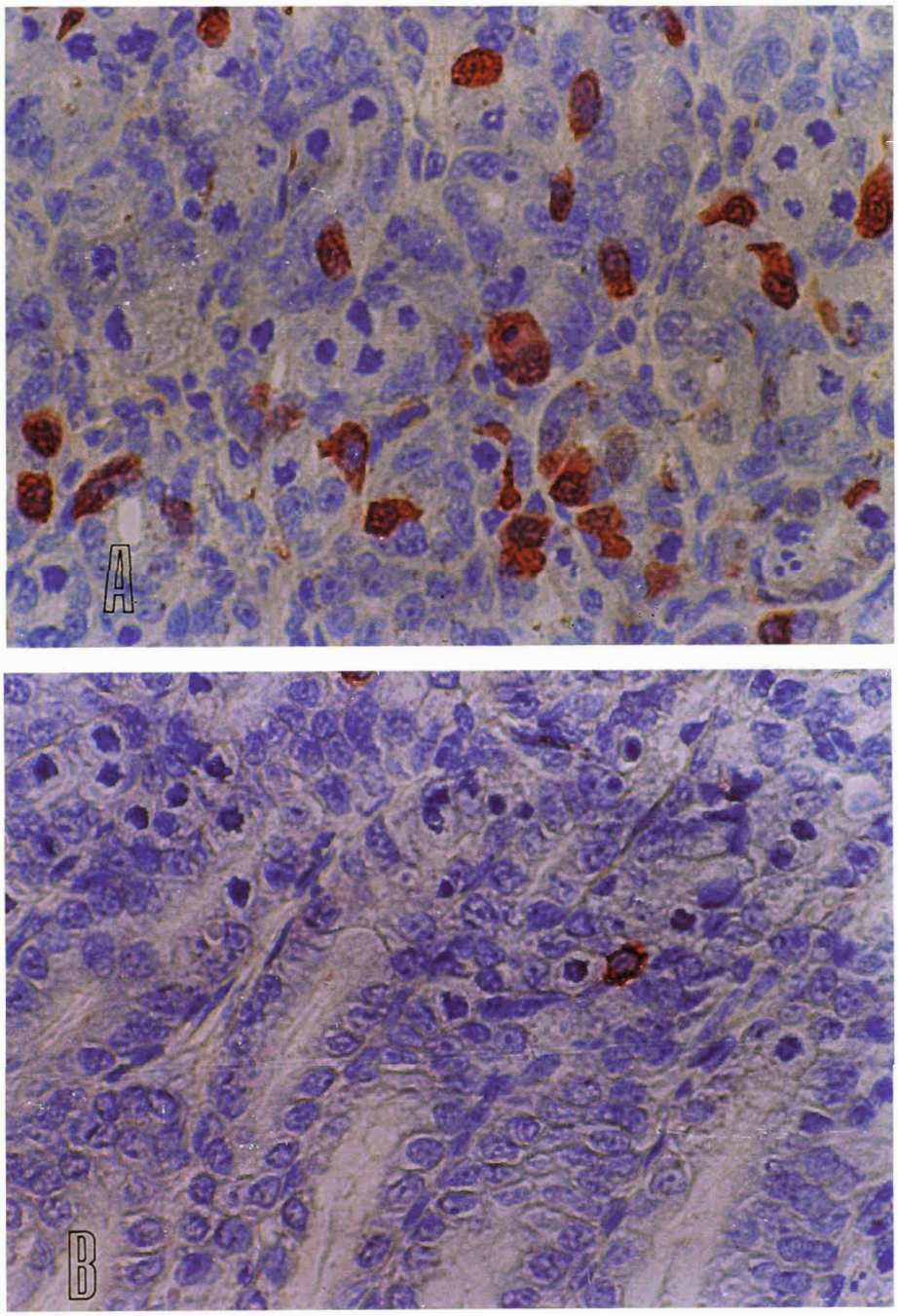

Fig. 3. Immunoreactive gastrin (A) and somatostain (B) cells (red-stained cells) in the rat gastric antral mucosa epithelium. × 400

proglumide, when applied alone, significantly decreased the mean number of D-cells in the gastric antral mucosa epithelium (Fig. 2).

Discussion

As expected, treatment with OM resulted in an increase of serum gastrin concentration. This observation is in agreement with the earlier data of other authors (Schusdziarra et al., 1978; Hakanson et al., 1986; Pawlikowski, 1986). Our data demonstrate that an administration of OM increases the density of G-cells, while decreasing the density of D-cells, in the rat gastric antral mucosa. This effect, most probably, does not depend on the hypergastrinaemia, since it could not be blocked by a simultaneous treatment of rats with proglumide. It appears that the observed phenomenon is a direct result of a lower gastric acidity, as a consequence of OM treatment. A similar mechanism may be responsible for a decrease in the mean number of D-cells, following the treatment with proglumide or SMS 201-995. Both drugs cause an

inhibition of gastric acid secretion, although the mechanism of such an inhibition is completely different. It is noteworthy that Tielemans et al. (1989) have demonstrated an increased proliferation of the, so-called, enterochromaffin-like cells (ECL cells) in OM-treated rats. The cited authors found a positive correlation between the plasma gastrin concentrations and the ECL cell ^{3}(H)-thymidine labelling index after OM-treatment and concluded a causal relationship between these two phenomena.

The results of our present experiment indicate that the anti-acid treatment not only leads to temporary alterations of gastrin and somatostatin secretion, but also creates possibilities for more prolonged changes in G- and D-cell populations. Such changes may result in a chronic gastrin-somatostatin imbalance. Since gastrin is known to be a trophic factor for gastrointestinal mucosa, while somatostatin exerts an opposite, antiproliferative action, such an imbalance may increase the risk of gastric neoplasia in patients suffering from hypo- or achlorhydria (Pawlikowski, 1986).

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