Experimental and Toxicologic Pathology 64 (2012) 339–343

Contents lists available at ScienceDirect

Experimental and Toxicologic Pathology

journal homepage: www.elsevier.de/etp

Comparison of the protective effects of various antiulcer agents alone or in combination on indomethacin-induced gastric ulcers in rats

Fikret Vehbi Izzettin [a], Mesut Sancar [a,*], Betul Okuyan [a], Sule Apikoglu-Rabus [a], Ugur Cevikbas [b]

[a] Clinical Pharmacy Department, Faculty of Pharmacy, Marmara University, Istanbul, Turkey
[b] Pathology Department, Istanbul Faculty of Medicine, Istanbul University, Istanbul, Turkey

ARTICLE INFO

Article history:
Received 5 January 2010
Accepted 27 September 2010

Keywords:
Indomethacin-induced gastric ulcer
Omeprazole
Misoprostol
Ranitidine
Bismuth
Rat

ABSTRACT

The aim of this study which was structured with the objective of determination of the optimum protective therapy against the long term NSAID therapy-induced ulcers was to compare the gastro-protective effects of various antiulcer drugs (ranitidine, omeprazole, bismuth and misoprostol) alone or in combination with each other in different doses on indomethacin-induced gastric ulcers in rats.

In this experimental study the protective effect of misoprostol (100 μg/kg/day and 10 μg/kg/day i.g.), omeprazole (5 mg/kg/day and 1.5 mg/kg/day i.p.), ranitidine (40 mg/kg/day and 10 mg/kg/day i.p.), bismuth (70 mg/kg/day and 15 mg/kg/day i.g.), combinations of misoprostol (10 μg/kg/day i.g.) plus omeprazole (1.5 mg/kg/day i.p.) and misoprostol (10 μg/kg/day i.g.) plus ranitidine (10 mg/kg/day i.p.) are investigated on indomethacin (50 mg/kg/day s.c.) induced gastric ulcers. Half an hour before indomethacin administration, each group received the above treatment regimens for 5 days. After 5-day treatment, the rats were sacrificed and histopathological and hematological examinations were performed. The following regimens were found to be effective in the prevention of indomethacin-induced gastric lesions: 100 μg/kg misoprostol, 10 μg/kg misoprostol, 5 mg/kg omeprazole, combination of 10 μg/kg misoprostol plus 1.5 mg/kg omeprazole and 10 μg/kg misoprostol plus 10 mg/kg ranitidine. The prevention rates achieved by these treatments were 71.4%, 50%, 47.6%, 52.4% and 50%, respectively. As a result of this study, misoprostol and omeprazol were found to be effective in protection against NSAID-induced gastric problems; while, ranitidine and bismuth were not. Also, the combinations of these agents were not found to have additive or synergistic effects.

1. Introduction

Non-steroidal anti-inflammatory drugs (NSAIDs) are widely used for their analgesic, antipiretic and antiinflammatory effects (Cooke, 1996). Gastrointestinal (GI) symptoms are the most common adverse events associated with NSAID therapy (Ehsanullah et al., 1988; Singh, 1998). Gastric ulcers, bleeding and perforation are serious side effects which are observed in long term NSAID therapy. Chronically administrated NSAIDs induce clinically significant gastric mucosal damage by two mechanisms: direct mucosal irritation and prostaglandin inhibition.

In the literature there are some experimental models of long term NSAID therapy-induced gastric ulcers in animals (Kolbasa et al., 1988; Wallace and McKnight, 1993). Animal models consisting of repeated administration of indomethacin are found both macroscopically and microscopically similar to NSAID-induced ulcers in man (Wallace and McKnight, 1993).

Four different drugs are recently used for the prevention of GI side effects of NSAIDs: histamine H2 receptor blockers, proton pump inhibitors (PPI), sucralfat and misoprostol (Agrawal, 1995). In the literature there are different reports concerning the protective effect of drugs in NSAID-induced ulcers. Although various studies showed that misoprostol has better protective effects when compared with other drugs (Agrawal, 1995; Brooks and Day, 1991; Cooke, 1996; Gabriel et al., 1993), PPIs were found to be more effective in other studies (Brown and Yeomans, 1999; Steen et al., 1999; Yeomans, 1999). The clinical use of misoprostol is restricted due to its high cost and intolerable side effects (Cooke, 1996). Therefore, we investigated the effects of the combination of low dose misoprostol (10 μg/kg i.g.) with low dose ranitidine (10 mg/kg/day i.p) or omeprazole (1.5 mg/kg/day i.p.) in the present study.

This study was structured with the objective of determination of the optimum protective therapy against the long term NSAID therapy-induced ulcers. With this objective, in this study, we aimed to compare the gastro-protective effects of various antiulcer drugs (ranitidine, omeprazole, bismuth and misoprostol) alone or

* Corresponding author at: Clinical Pharmacy Department, Faculty of Pharmacy, Marmara University, Tibbiye Cad. No. 49, 34668 Istanbul, Turkey.
Tel.: +90 216 3464060; fax: +90 216 3452952.
E-mail address: sancarmesut@yahoo.com (M. Sancar).

0940-2993/$ – see front matter © 2010 Elsevier GmbH. All rights reserved.
doi:10.1016/j.etp.2010.09.007

Table 1
Prophylactic treatment protocols.

Groups	Prophylactic treatment protocol
I-C	Indomethacin 50 mg/kg s.c.
M-T1	Indomethacin 50 mg/kg s.c. + Misoprostol 100 μg/kg i.g.
M-T2	Indomethacin 50 mg/kg s.c. + Misoprostol 10 μg/kg i.g.
O-T1	Indomethacin 50 mg/kg s.c. + Omeprazole 5 mg/kg i.p.
O-T2	Indomethacin 50 mg/kg s.c. + Omeprazole 1.5 mg/kg i.p.
R-T1	Indomethacin 50 mg/kg s.c. + Ranitidine 40 mg/kg i.p.
R-T2	Indomethacin 50 mg/kg s.c. + Ranitidine 10 mg/kg i.p.
B-T1	Indomethacin 50 mg/kg s.c. + Bismuth 70 mg/kg i.g.
B-T2	Indomethacin 50 mg/kg s.c. + Bismuth 15 mg/kg i.g.
MO-T	Indomethacin 50 mg/kg s.c. + Misoprostol 10 μg/kg i.g. + Omeprazole 1.5 mg/kg i.p.
MR-T	Indomethacin 50 mg/kg s.c. + Misoprostol 10 μg/kg i.g. + Ranitidine 10 mg/kg i.p.
C	Normal saline 0.5 ml s.c.

n = 10 for each group; s.c.: subcutaneous; i.g.: intragastric; i.p.: intraperitoneal; I: indomethacin; M: misoprostol; O: omeprazole; R: ranitidine; B: bismuth; C: control.

in combination with each other in different doses on indomethacin-induced gastric ulcers in rats.

2. Materials and methods

2.1. Animals

Wistar albino rats of both sexes, weighing 200–250 g and approximately of the same age were obtained from the animal house of Marmara University, Faculty of Medicine. The experimental animals were maintained under standard laboratory conditions with alternating light and dark cycles of 12 h each. During the study they were allowed to take standard laboratory food pellets and water ad libitum and were housed in suitable and adequate conditions that fulfill the animal house instructions. The experimental protocol was approved by Marmara University, Faculty of Medicine Experimental Animals Research Ethics Committee (Reg. No. 32.2000.mar).

2.2. Drugs

Indomethacin (Deva Drug Company, Turkey), misoprostol (Ali Raif Drug Company, Turkey) and omeprazole (Losec®, Astra, Turkey), ranitidine (Ulcuran®, Abfar, Turkey), colloidal bismuth subcitrate (Denol®, Eczacibasi, Turkey) were used.

2.3. Vehicle

Indomethacin was homogenized in 20 ml saline containing one drop of Tween 80. Omeprazole and ranitidine was dissolved in their specific solvent solutions ready for injection. Misoprostol and bismuth subcitrate were dissolved in a sufficient amount of distilled water.

2.4. Experimental procedure

Rats were randomly divided into 12 groups and each group had 10 rats. Experiments were initiated following a 24 h fasting with water *ad libitum*. Gastric ulcerations were induced according to the method described by Kolbasa et al. (1988) by subcutaneous (s.c.) indomethacin (50 mg/kg) administration for 5 consecutive days. Prophylactic treatment protocols for each group were as shown in Table 1. All antiulcer drug regimens were administered half an hour before indomethacin administration. Indomethacin was administered by subcutaneous injection, while, omeprazole and ranitidine were injected intraperitoneally and, misoprostol and bismuth were administered via orogastric tube.

2.5. Histopathological examination

To assess the presence and extent of indomethacin-induced gastric lesions the animals were sacrificed at the end of the treatment period (at 6th day); laporatomy was performed to excise the stomach out including part of the duodenum (Eastwood and Quimby, 1982).

The stomach of each animal was opened along the greater curvature, emptied of its contents, washed with stream water and stored in 10% formalin solution until the day of the examination (Bauer et al., 1986; Kuwayama et al., 1991; Segami et al., 1996). For light microscopic evaluation skin tissue samples were fixed in 10% formaldehyde and processed routinely for embedding in paraffin. Paraffin sections were stained with Hematoxylin and Eosin to indicate histological degeneration (Wang et al., 1989; Kuwayama et al., 1991).

The size of lesions was measured using a light microscope and were graded according to the scoring system which was structured using various methods mentioned in the literature (Jiranek et al., 1989; Lee et al., 1996; Schmassmann et al., 1998). The scoring system used was as follows: (0) without lesion, (1) mild edema and rare erosions, (2) common erosion and necrosis, (3) ulcer with <2 mm diameter, (4) ulcer with 2–4 mm diameter, and (5) ulcer with >4 mm diameter.

2.6. Hematological examination

Blood samples were taken from the animals' tail veins to measure pre- and post-treatment hematocrit and hemoglobin levels (Zeller et al., 1998).

2.7. Statistical analysis

All results were expressed as mean ± standard deviation (SD). Differences between ulcer scores among groups were analyzed using one-way ANOVA followed by Dunnett's multiple comparison tests as the post hoc test. The significance of the difference between the pre- and post-treatment hematocrit and hemoglobin levels was compared using independent *t*-test. A p-value <0.05 was considered as statistically significant.

3. Results

Histopatologic properties of normal gastric mucosa with of the rats administered normal saline (0.5 ml s.c.) was as shown in Fig. 1a. The I-C group, expressing the indomethacin-induced gastric ulcer model, had a mean (SD) ulcer score of 4.2 (1.0) (Fig. 1b). Significantly differences in the mean (SD) ulcer scores were found between the following groups and the I-C group:

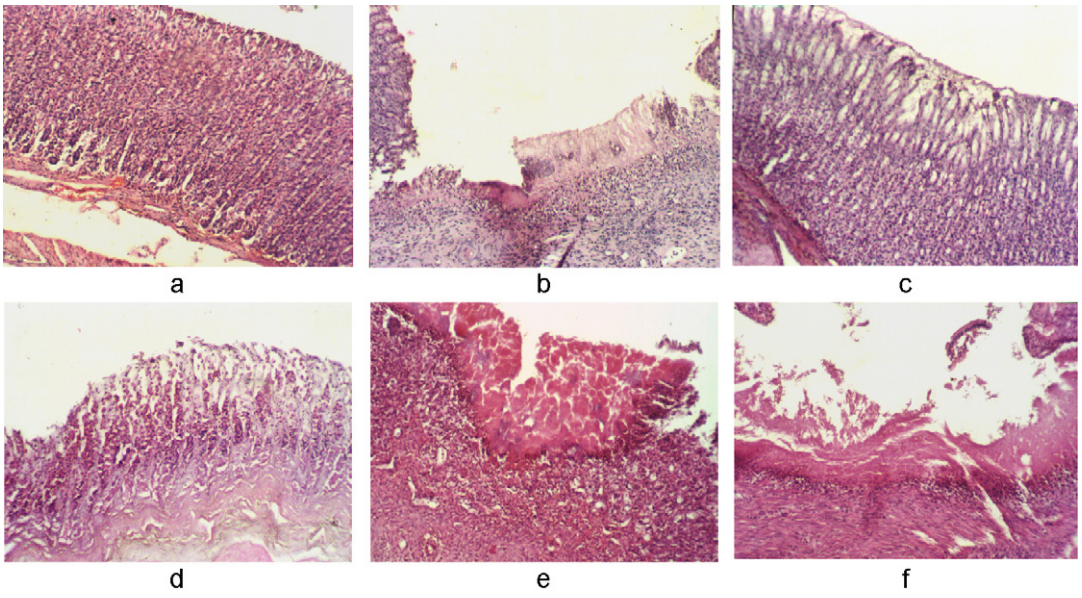

Fig. 1. (a) Normal gastric mucosa of normal saline (0.5 ml, s.c.) administered rats. (b) Deep and severe gastric ulcer formation observed in indomethacin (50 mg/kg; s.c.) administered rats. (c) Recovery of gastric mucosa of the misoprostol (100 μg/kg; i.g.) treated rats. (d) Mild gastric ulcerations and erosion in a half area of mucosa observed in misoprostol (10 μg/kg; i.g.) alone and combination with ranitidine (10 mg/kg i.p.) and omeprazole (1.5 mg/kg; i.p.) and omeprazole (5 mg/kg; i.p.) alone treated rats. (e) Moderate gastric ulcerations in bismuth (70 mg/kg/day i.p.) alone treated rats. (f) Severe gastric ulcer view representative of the omeprazole (1.5 mg/kg/day i.p.) alone, ranitidine (40 mg/kg/day and 10 mg/kg/day i.p.) alone and bismuth (15 mg/kg/day i.g.) alone treated rats. Hematoxylin–Eosin staining; original magnifications: 125×.

M-T1 (100 μg/kg misoprostol; 1.2 ± 1.6), M-T2 (10 μg/kg misoprostol; 2.1 ± 1.8), O-T1 (5 mg/kg omeprazole; 2.2 ± 1.5), MO-T (10 μg/kg misoprostol plus 1.5 mg/kg omeprazole; 2.0 ± 1.6) and MR-T (10 μg/kg misoprostol plus 10 mg/kg ranitidine; 2.1 ± 1.7) ($p < 0.05$) (Fig. 1c and d). No significant difference was observed between other groups (O-T2, R-T1, R-T2, B-T1, B-T2) and the I-C group ($p > 0.05$; Fig. 1e and f). The ulcer scores of the groups which were histopathologically evaluated were as shown in Fig. 2.

Hematocrit and hemoglobin levels were evaluated in groups I-C, M-T1, O-T1, R-T1, B-T1 and C. The pre-treatment hematocrit and hemoglobin levels were similar for all groups ($p > 0.05$). When the pre-treatment hematocrit and hemoglobin levels were compared with the post-treatment levels, statistically significant reductions were observed for the groups I-C, M-T1, O-T1, R-T1 and B-T1 ($p < 0.05$); while, the pre- and post-treatment levels were similar for the control group (C; 0.5 ml normal saline; s.c.; $p > 0.05$). Pre- and post-treatment hematocrit and hemoglobin levels were as shown in Tables 2 and 3.

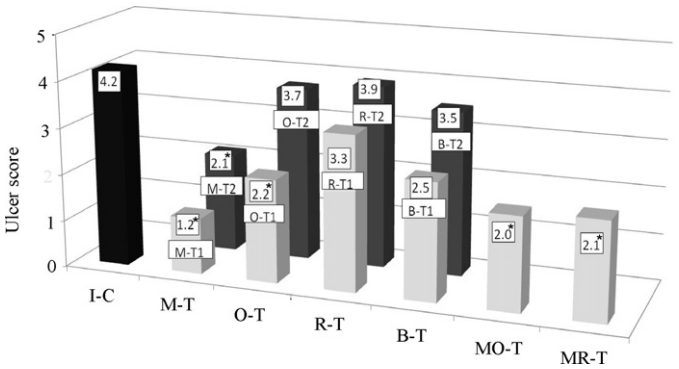

Fig. 2. Effect of different treatment regimens on ulcer scores in rats. M-T1: 100 μg/kg i.g. misoprostol; M-T2: 10 μg/kg i.g. misoprostol; O-T1: 5 mg/kg i.p. omeprazole; O-T2: 1,5 mg/kg i.p. omeprazole; R-T1: 40 mg/kg i.p. ranitidine; R-T2: 10 mg/kg i.p. ranitidine; B-T1: 70 mg/kg i.g. bismuth; B-T2: 15 mg/kg i.g. bismuth; MO-T: 10 μg/kg i.g. misoprostol + 1,5 mg/kg i.p. omeprazole; MR-T: 10 μg/kg i.g. misoprostol + 10 mg/kg i.p. ranitidine; I-C: 50 mg/kg s.c.; values are expressed as mean (standard deviation); * $p < 0.05$ when compared with controls.

Table 2
The pre- and post-treatment hemoglobin levels of different groups.

Groups	Pre-treatment	Post-treatment
I-C	13.66 ± 0.467	6.43 ± 0.631*
M-T1	13.88 ± 0.711	7.38 ± 0.895*
O-T1	14.20 ± 0.981	7.23 ± 0.759*
R-T1	14.17 ± 0.826	6.70 ± 0.680*
B-T1	13.87 ± 0.760	7.18 ± 0.865*
C	14.12 ± 0.571	14.30 ± 0.602

I-C: 50 mg/kg indomethacin; M-T1: 100 μg/kg misoprostol; O-T1: 5 mg/kg omeprazole; R-T1: 40 mg/kg ranitidine; B-T1: 70 mg/kg bismuth; C: control, 0.5 ml normal saline.
* $p < 0.05$ when compared with the pre-treatment levels.

Table 3
The pre- and post-treatment hematocrit values of different groups.

Groups	Pre-treatment	Post-treatment
I-C	43.60 ± 1.17	23.70 ± 2.58*
M-T1	44.00 ± 1.15	26.10 ± 3.75*
O-T1	44.30 ± 1.95	25.50 ± 2.84*
R-T1	44.20 ± 1.81	24.80 ± 3.85*
B-T1	43.50 ± 1.43	24.60 ± 4.09*
C	44.80 ± 1.48	45.30 ± 1.77

I-C: 50 mg/kg indomethacin; M-T1: 100 (g/kg misoprostol; O-T1: 5 mg/kg omeprazole; R-T1: 40 mg/kg ranitidine; B-T1: 70 mg/kg bismuth; C: Control 0.5 ml normal saline.
* $p < 0.05$ as compared with pre-treatment value.

4. Discussion

In the literature there are many studies on the treatment of NSAID-induced gastric ulcers; however, only a few studies examine the prevention of these ulcers. Therefore, we planned this study with the aim of comparing the effects of different antiulcer drugs with different doses and as combinations in the prevention of NSAID-induced gastric ulcers.

In our study the protective effects of misoprostol was found to be dose-related. This finding was in accordance with other studies in the literature that were conducted on experimental animals (Bauer et al., 1986) and healthy volunteers (Jiranek et al., 1989).

In the literature, there are various studies assessing the efficacy of omeprazole in prevention of NSAID-induced GI problems. In one of these studies, Ekstrom et al. (1996) evaluated the protective effect of daily 20 mg omeprazole administration in comparison with placebo in patients taking NSAIDs with a history of peptic ulcer and dyspepsia. They found that omeprazole provided effective protection by relieving symptoms of NSAID-induced peptic ulcer and dyspepsia. In an other study conducted on 20 volunteers receiving 650 mg aspirin four times daily for 2 weeks, Cooke (1996) reported that 40 mg omeprazole provided more effective protection than placebo in the healing of both gastric or duodenal mucosa damage. In their study which was similar to ours in terms of treatment duration and indomethacin doses, Lee et al. (1996) assessed the protective effects of omeprazole comparing the treatment group which received 40 mg/kg s.c indomethacin + 40 mg/kg omeprazole for 4 days with the control group which received only 40 mg/kg s.c indomethacin for 4 days. They assessed the difference between groups in terms of mucosal damage and prostaglandin production and found that omeprazole decreased the production of gastric ulcers by more than 80%.

Another study which comparatively assessed the effects of misoprostol and omeprazol on indomethacin-induced gastric ulcers in rats found that misoprostol completely reversed the gastric lesions; while, omeprazole failed to fully reverse the effect of indomethacin on granulation tissue maturation (Arisawa et al., 2006).

In this study we found that misoprostol at both doses produced effective protection; while, omeprazole had significant protective effects only at high doses. This result complies with various others in the literature.

Arroyo et al. (2000) found that ranitidine had no protective effect on indomethacin induced corpus and antrum gastric lesions, however in the same study ebrotidine and omeprazole significantly prevented indomethacin induced gastric lesions.

In our previous study which aimed to compare effect of *Glycyrrhiza glabra* (liquorice) root decoction vs. omeprazole and misoprostol for the treatment of aspirin-induced gastric ulcers in rats, misoprostol was found to be significantly protective according to histopathological observations (Sancar et al., 2009).

When considering the lesions formed in the stomach, ranitidine and colloidal bismuth subcitrate, at either dose were not found to provide adequate protection.

In one of the few studies assessing the efficacy of bismuth in NSAID-induced ulcers, Konturek et al. (1987), found that colloidal bismuth subcitrate was rather effective in protection against ulcer formation.

In our study, the following regimens were found to be effective in preventing indomethacin-induced gastric lesions: 100 μg/kg misoprostol, 10 μg/kg misoprostol, 5 mg/kg omeprazole, combination of 10 μg/kg misoprostol plus 1.5 mg/kg omeprazole and 10 μg/kg misoprostol plus 10 mg/kg ranitidine. The prevention rates achieved by these treatments were 71.4%, 50%, 47.6%, 52.4% and 50%, respectively. The ulcer scores in groups treated with ranitidine, bismuth and low dose omeprazole were not found to be different from those of the indomethacin group; therefore, the efficacy of these regimens was not enough to prevent indomethacin induced ulcers.

In the literature there are studies reporting the beneficiary effects of omeprazole in NSAID-induced ulcers. Hawkey et al. (1998) compared the ulcer healing and protective effects of 20 mg omeprazole, 40 mg omeprazole and 800 μg misoprostol in 935 patients who had to receive NSAIDs chronically. They reported no difference among the groups by means of ulcer healing; while, at the maintenance dose of 20 mg, omeprazole provided a more effective protection than misoprostol. Also, they observed that omeprazole was well-tolerated by patients.

In their study conducted on 541 patients who had to receive NSAIDs continuously, Yeomans et al. (1998), reported that ulcer healing and protective effects of both 20 mg and 40 mg doses of omeprazole were better than that was observed at twice daily ranitidine administration.

Misoprostol is generally considered as not very favorable due to its high cost and side effects. Although, side effects would be decreased in lower doses, gastroprotective effects would also reduce in a dose related manner. Therefore, in our study we tested omeprazole and ratinidine both separately and in combinations with misoprostol. The results obtained showed that the protective effect of 10 μg/kg misoprostol alone was similar to those achieved by combinations.

Post-treatment hemoglobin and hematocrit levels were lower than those measured before the treatment; this could be due to the NSAID-induced blood loss in the animals. Recorded decrease rates in both parameters were not significantly different among groups. However, the lowest decrease was obtained in the misoprostol 100 μg/kg group. Thus, we can say that, although not statistically significant, the effects of misoprostol in prevention of GI hemorrhage was greater than other drugs

The results of this study suggest that while misoprostol and omeprazole may be effective in protection against NSAID-induced gastric problems, ranitidine and bismuth may not. Also, combining the lower doses of these drugs would not result in any additive or synergistic effect.

References

Agrawal NM. Epidemiology and prevention of non-steroidal anti-inflammatory drug effects in the gastrointestinal tract. Br J Rheumatol 1995;34(Suppl. 1):5–10.

Arisawa T, Harata M, Kamiya Y, Shibata T, Nagasaka M, Nakamura M, et al. Is omeprazole or misoprostol superior for improving indomethacin-induced delayed maturation of granulation tissue in rat gastric ulcers? Digestion 2006;73(1):32–9.

Arroyo MT, Lanas A, Sáinz R. Prevention and healing of experimental indomethacin-induced gastric lesions: effects of ebrotidine, omeprazole and ranitidine. Eur J Gastroenterol Hepatol 2000;12(3):313–8.

Bauer RF, Bianchi RG, Casler J, Goldstin B. Comparative mucosal protective properties of misoprostol, cimetidine and sucralfate. Dig Dis Sci 1986;31(Suppl. 2):81S–5S.

Brooks PM, Day RO. Nonsteroidal antiinflammatory drugs – differences and similarities. N Engl J Med 1991;324:1716–25.

Brown GJ, Yeomans ND. Prevention of the gastrointestinal adverse effects of nonsteroidal anti-inflammatory drugs: the role of proton pump inhibitors. Drug Safety 1999;21(6):503–12.

Cooke CE. Disease management: prevention of NSAID-induced gastropathy. Drug Benefit Trends 1996;8(3):14–22.

Eastwood GL, Quimby GF. Effect of chronic aspirin ingestion on epithelial proliferation in rat fundus, antrum, and duodenum. Gastroenterology 1982;82:852–6.

Ehsanullah RB, Page MC, Tildesley G, Wood JR. Prevention of gastroduodenal damage induced by non-steroidal anti-inflammatory drugs: controlled trial of ranitidine. BMJ 1988;297:1017–21.

Ekstrom P, Carling L, Wtterhus S, Wingren PE, Anker-Hansen O, Lundegardh G, et al. Prevention of peptic ulcer and dyspeptic symptoms with omeprazole in patients receiving continuous non-steroidal anti-inflammatory drug therapy. Scand J Gastroenterol 1996;31(8):753–8.

Gabriel SE, Jaakkimainen RL, Bombardier C. The cost-effectiveness of misoprostol for nonsteroidal antiinflammatory drug-associated adverse gastrointestinal events. Arthritis Rheum 1993;36(4):447–56.

Hawkey CJ, Karrasch JA, Szczepanski L, Walker DG, Barkun A, Swannell AJ, et al. Omeprazole compared with misoprostol for ulcers associated with nonsteroidal antiinflammatory drugs. N Engl J Med 1998;338:727–34.

Jiranek GC, Kimmey MB, Saunders DR, Willson RA, Shanahan W, Silverstein E. Misoprostol reduces gastroduodenal injury from one week of aspirin: an endoscopic study. Gastroenterology 1989;96:656–61.

Kolbasa KP, Lancaster C, Olafsson AS, Gilbertson SK, Robert A. Indomethacin-induced gastric antral ulcers in hamsters. Gastroenterology 1988;95(4):932–44.

Konturek SJ, Radecki T, Pisatucki I, Brozozowski T, Drozdowicz D. Gastroprotective by colloidal bismuth subcitrate (De-Nol) and sucralfate. Role of endogenous prostaglandins. Gut 1987;28:201–5.

Kuwayama H, Matsuo Y, Eastwood GL. Effects of sucralfate, lansoprazole, and cimetidine on the delayed healing by hydrocortisone sodium phosphate of chronic gastric ulcers in the rat. Am J Med 1991;91(Suppl. 2A):15S–9S.

Lee M, Kallal SM, Feldman M. Omeprazole prevents indomethacin-induced gastric ulcers in rabbits. Aliment Pharmacol Ther 1996;10(4):571–6.

Sancar M, Hantash T, Okuyan B, Apikoglu-Rabus S, Cirakli Z, Gulluoglu MG, et al. Comparative effectiveness of Glycyrrhiza glabra vs. omeprazole and misoprostol for the treatment of aspirin-induced gastric ulcers. Afr J Pharm Pharmacol 2009;3(12):618–23.

Schmassmann A, Peskar BM, Stettler C, Netzer P, Stroff T, Flogerzi B, et al. Effects of inhibition of prostaglandin endoperoxide synthase-2 in chronic gastrointestinal ulcer models in rats. Br J Pharmacol 1998;123:795–804.

Segami T, Suzuki Y, Ito M. Simultaneous evaluation of gastric and duodenal ulcers-healing activities of anti-ulcer agents in rats. Biol Pharm Bull 1996;19(1):53–6.

Singh G. Recent considerations in nonsteroidal anti-inflammatory drug gastropathy. Am J Med 1998;1.05(Suppl. 1B):31–8.

Steen KS, Lems WF, Dijkmans BA. Effectiveness, safety and costs of measures for prevention of gastropathy due to the use of nonsteroidal antiinflammatory drugs. Ned Tijdschr Geneeskd 1999;143(32):1649–52.

Wang JY, Yamasaki S, Takeuchi K, Okabe S. Delayed healing of acetic acid-induced gastric ulcers in rats by indomethacin. Gastroenterology 1989;96:393–402.

Yeomans ND. A clinical approach to management of patients with non-steroidal anti-inflammatory gastropathy. Ital J Gastroenterol Hepatol 1999;31(Suppl. 1):89–92.

Yeomans ND, Tulassay Z, Juhasz L, Racz I, Horward JM, Van Rensburg CJ, et al. A comparison of omeprazole with ranitidine for ulcers associated with nonsteroidal antiinflammatory drugs. N Engl J Med 1998;338:719–26.

Wallace JL, McKnight GW. Characterization of a simple animal model for nonsteroidal anti-inflammatory drug induced antral ulcer. Can J Physiol Pharmacol 1993;71(7):447–52.

Zeller W, Weber H, Panoussis B, Bürge T, Bergmann R. Refinement of blood sampling from the sublingual vein of rats. Lab Anim 1998;32:369–76.