Gastroprotective and Antioxidant Effects of Montelukast on Indomethacin-Induced Gastric Ulcer in Rats

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Received January 12, 2007; Accepted July 23, 2007

Abstract. Montelukast, a selective reversible cysteinyl leukotriene D4-receptor (LTD4 receptor) antagonist, is used in the treatment of asthma. We have investigated alterations in the glutathione (GSH) and activity levels of antioxidative enzymes [superoxide dismutase (SOD), catalase (CAT), glutathione S-transferase (GST), and glutathione reductase (GR)] and myeloperoxidase (MPO), as markers of the ulceration process following oral administration of montelukast, lansoprazole, famotidine, and ranitidine, respectively, in rats with indomethacin-induced ulcers. In the present study, we found that 1) montelukast, lansoprazole, famotidine, and ranitidine all reduced the development of indomethacin-induced gastric damage, with this reduction occurring at a greater magnitude for montelukast, famotidine, and lansoprazole than for ranitidine; 2) montelukast and ranitidine both alleviated increases in the activity levels of CAT and GST enzymes resulting from gastric injury; 3) montelukast and ranitidine both ameliorated depressions in the GSH and activity levels of SOD and GR enzymes caused by indomethacin administration; and 4) all doses of montelukast, lansoprazole, and ranitidine decreased amplification of MPO activity resulting from induced gastric injuries. These results suggest that the gastroprotective effects of montelukast on indomethacin-induced ulcerations can be attributed to its ameliorating effect on oxidative damage and MPO activity.

Keywords: montelukast, indomethacin, gastroprotective effect, myeloperoxidase, antioxidant enzyme

Introduction

Several drugs are currently used in the treatment of asthma, including sodium cromoglycate, sodium nedocromil, theophylline, and oral or inhaled glucocorticoids (1). However, montelukast is a new anti-inflammatory drug that interferes directly with leukotriene production (5-lipoxygenase inhibitors) and/or reception (leukotriene receptor antagonists) (2). As leukotrienes are important mediators of asthma, this selective reversible cysteinyl leukotriene D4 (LTD4)-receptor antagonist helps treat the illness by reducing airway eosinophilic inflammation (3). Both leukotriene pathway modifiers and LTD4-receptor antagonists (4) have been shown to improve asthma control. Therefore, montelukast provides a significant improvement in chronic asthmatic symptoms and maintains a highly safe profile (5). It was reported by Wallace et al. that leukotriene C4 (LTC4)/LTD4-receptor antagonists ameliorate the ethanol-induced gastric mucosal damage (6, 7). However, it has been shown that LTC4 is not the exclusive mediator of gastric injury caused by indomethacin (8). Cysteinyl leukotrienes, leukotrienes C4, D4, and E4 (LTC4, LTD4, and LTE4), are secreted mainly by eosinophils, mast cells, monocytes, and macrophages and perform a number of pathogenic actions during periods of inflammation (9, 10). It has been reported by Konturek et al. (11) that exogenous LTC4 causes only mild cases of gastric mucosal injury, but greatly augments the mucosal lesions induced by various noxious agents such as ethanol, taurocholate, and aspirin or by stress. These lesions are accompanied by significant
increases in the mucosal generation of LTC₄. On the other hand, LTC₄-receptor antagonists have been reported to reduce the extent of gastric damage (12). Similarly, Peskar (8) has reported that ethanol stimulates the generation of LTC₄ and 15-hydroxyicosatetraenoic acid in rat gastric mucosa and that a number of drugs show gastroprotective effects by dose-dependently inhibiting the stimulatory action of ethanol on the gastric mucosa.

Non-steroidal anti-inflammatory drugs (NSAIDs) are widely used in the treatment of pain, fever, and inflammation. However, these drugs carry various side effects, especially those occurring in the gastrointestinal tract, such as gastric mucosal erosions, ulcerations, bleeding, and perforations. There is also an increased risk of bleeding from preexisting peptic ulcers (13). It is widely known, however, that NSAIDs produce beneficial effects through their ability to block cyclooxygenase (COX), thereby inhibiting prostaglandin (PG) production (14). The main consequence of a decreased PG level is its contribution to the pathogenesis of gastro-duodenal mucosal ulcerations (15). Other major factors include a decrease in the secretion of mucus, an inhibition of bicarbonate secretion, a reduction in the mucosal blood flow, an alteration in microvascular structures and microvascular injury (16), neutrophil infiltration, and an increase in acid and pepsinogen secretion (13). One of the major causes of mucosal lesions associated with NSAID-induced gastrointestinal damage also plays a vital role in reactive oxygen species (ROS) like superoxide radical anions and hydroxyl radicals (17, 18). These mechanisms, in combination with those related to PG suppression, lead to microvessel occlusion and subsequent hyper production of reactive oxygen metabolites. In particular, indomethacin causes gastric erosions with increased lipid peroxidation and decreased glutathione peroxidase activity (19). The presence of superoxide anions (O₂⁻) may be considered a reason for the gastric damage as well as other tissue damage. Thus, much attention has recently been focused on oxygen-derived free radicals. This mechanism, in combination with ROS, damages membrane proteins by inducing lipid peroxidation and attacking unsaturated fatty acids (20). The damage caused by the peroxidation of unsaturated fatty acids decreases the membrane’s permeability, while inhibiting the activities of enzymes and receptors and the activation of cells (20, 21). Therefore, antioxidant defense systems and their components, including antioxidant enzymes, foods, and drugs, are important in preventing the toxic and disease-causing effects of oxygen-derived free radicals. Oxygen-handling cells have antioxidant enzymes that are able to protect them. It has been shown that neutrophil infiltration into the gastric mucosal tissues is a critical process in the pathogenesis of a variety of gastric ulcers. A myeloperoxidase (MPO) assay has been in widespread use as an index of neutrophil infiltration in various gastric injuries (22, 23).

The purpose of the present study was to investigate the gastroprotective effects of montelukast (Fig. 1) against indomethacin-induced gastric injury in rats and to determine the relationship between this gastroprotective mechanism and MPO activity in the gastric tissues.

Materials and Methods

Chemicals

Montelukast and indomethacin were from Merck Sharp & Dohme (Istanbul, Turkey) and lansoprazole and famotidine were from Nobel A.Ş., Istanbul, Turkey. All other chemicals for laboratory experimentation were purchased from Sigma Chemical (Taufkirchen, Germany).

Animals

A total of 48 male, Sprague-Dawley rats, weighing 180 – 190 g, were provided by the Experimental Animal Laboratory of Ataturk University, Faculty of Medicine and Department of Pharmacology. The animals were grouped before the experiments and kept under standard conditions (24). Animal experiments were performed in accordance with the national guidelines for the use and care of laboratory animals and approved by the local animal care committee of Ataturk University.

Indomethacin-induced gastric damage

In these series of experiments, the effects of montelukast, lansoprazole, famotidine, and ranitidine on indomethacin-induced gastric damage were determined (25, 26). The protective effects of montelukast were compared with the H₂-receptor blockers, ranitidine and famotidine, and the proton pump inhibitor lansoprazole. Animals were divided into eight groups, each consisting of six rats. Montelukast (5, 10, and 20 mg/kg body weight doses, prepared by suspending in water), lansoprazole (30 mg/kg body weight) (27), famotidine...
was expressed as mm

2

sum of ulcerous areas and total stomach area calculated.

cellophane sheet was put on a millimeter paper and the ulcerous areas were drawn on a cellophane sheet. The ulcerous stomach was ingrained on a planar surface with small pins. Then the total areas of the stomach and ulcerous areas were measured to calculate the gastric damage score. For this purpose, the ulcerous stomach was ingrained on a planar surface with small pins. Then the total areas of the stomach and ulcerous areas were drawn on a cellophane sheet. The cellophane sheet was put on a millimeter paper and the sum of ulcerous areas and total stomach area calculated was expressed as mm

2

. The indomethacin group was compared with the healthy group. The protective effect of montelukast was compared with the results obtained from the indomethacin, lansoprazole, famotidine, and ranitidine groups. Ulcer index (UI) and % inhibition in ulcer index in relation to the indomethacin group were estimated from formulas:

\[ UI = \frac{\text{Ulcercated area (mm}^2\text{) / total stomach area (mm}^2\text{)}}{100} \]

\[ \% \text{Inhibition} = \frac{\text{UI}_{\text{treatment}}}{\text{UI}_{\text{control(indomethacin)}}} \times 100 \]

Biochemical investigation of stomach tissues

After the macroscopic analyses, catalase (CAT), glutathione S-transferase (GST), glutathione reductase (GR), superoxide dismutase (SOD), and MPO enzyme activities and the glutathione (GSH) levels in rat stomach tissues were determined. To prepare the tissue homogenates, stomach tissues were ground with liquid nitrogen in a mortar. The ground tissues (0.5 g each) were then treated with 4.5 ml of appropriate buffer. The mixtures were homogenized on ice using an ultra-turrax homogenizer for 15 min. Homogenates were filtered and centrifuged by using a refrigerated centrifuge at 4°C. Then, these supernatants were used for the determination of the enzymatic activities. All assays were carried out at room temperature in triplicate.

Biochemical estimations

SOD activity: Measurements were made according to Sun et al. (30). SOD estimation was based on the generation of superoxide radicals produced by xanthine and xanthine oxidase, which reacts with nitro blue tetrazolium (NTB) to form formazan dye. SOD activity was then measured at 560 nm by the degree of inhibition of this reaction and is expressed as millimole per minute per milligram tissue (mmol · min

−1

· mg tissue

−1

).

CAT activity: Decomposition of H

2

O

2

in the presence of CAT was followed at 240 nm (31). CAT activity was defined as the amount of enzyme required to decompose 1 nanomole of H

2

O

2

per minute, at 25°C and pH 7.8. Results are expressed as millimole per minute per milligram tissue (mmol · min

−1

· mg tissue

−1

).

GR activity: GR activity was determined spectrophotometrically by measuring the rate of NADPH oxidation at 340 nm (32). Results are expressed as the amount of enzyme that catalyzes the oxidation of 1 micromole of NADPH per minute per milligram tissue (µmol · min

−1

· mg tissue

−1

).

GST activity: Total GST activity was determined as described by Habig and Jakoby (33). Briefly, the enzyme activity was assayed spectrophotometrically at 340 nm in a 4-ml cuvette containing 0.1 M PBS (pH 6.5), 30 mM glutathione, 30 mM 1-chloro-2,6-dinitrobenzene, and tissue homogenate. Enzyme activity was expressed as nanomole per minute per milligram protein (nmol · min

−1

· mg tissue

−1

).

MPO activity: MPO activity was measured according to the modified method of Bradley et al. (34). The homogenized samples were frozen and thawed three times and centrifuged at 1500 × g for 10 min at 4°C. The supernatant was determined by adding 100 µl of the supernatant to 1.9 ml of 10 mM phosphate buffer (pH 6.0) and 1 ml of 1.5 mM o-dianisidine hydrochloride containing 0.0005% (wt/vol) hydrogen peroxide. The changes in absorbance at 450 nm of each sample were recorded in a UV–vis spectrophotometer. MPO activity in gastric tissues was expressed as micromole per minute per milligram tissue (µmol · min

−1

· mg tissue

−1

).

Total GSH determination: The amount of GSH in the gastric mucosa was measured according to the method of Sedlak and Lindsay (35). The mucosal surface of the stomach was collected by scraping, weighed, and homogenized in 2 ml of 50 mM Tris-HCl buffer containing 20 mM EDTA and 0.2 M sucrose, pH 7.5. Homogenate was immediately precipitated with 0.1 ml of 25% trichloroacetic acid and the precipitate was removed after centrifugation at 4200 rpm for 40 min at 4°C. The supernatant was used to determine GSH using 5,5'-dithiobis(2-nitrobenzoic acid). Absorbance was measured at 412 nm using a spectrophotometer. The results of the GSH level in the gastric mucosa were expressed as nanomole per milligram tissue (nmol · mg tissue

−1

).

Statistical analyses

Data of enzyme activity, GSH level, and ulceration score were subjected to one-way analysis of variance.
(ANOVA), with the presence of negative (control, healthy group) and positive controls (ranitidine, lansoprazole, famotidine), by using SPSS 11.0 software. Differences among groups were attained using the LSD option and significance was declared at \( P<0.05 \), \( P<0.01 \), and \( P<0.005 \). The indomethacin group was compared to the control (healthy) group. Treated groups were compared to both indomethacin and control (healthy) groups.

Results

Gastroprotective effect of montelukast on indomethacin-induced gastric damage

The gastroprotective effect of 5, 10, and 20 mg/kg doses of montelukast on indomethacin-induced gastric damage was macroscopically determined in rats. Percent inhibition effects of montelukast are shown in Table 1. There were remarkable hyperemias in the stomachs of indomethacin-administered rats. In the groups treated with montelukast, lansoprazole, famotidine, and ranitidine, hyperemias were very slight compared to indomethacin-administered rats. The ulcer index in rats receiving montelukast at doses of 5, 10, and 20 mg/kg suspended in water were \( 2.98 \pm 0.88 \), \( 2.21 \pm 0.91 \), and \( 2.03 \pm 0.49 \), respectively.

In the indomethacin, lansoprazole, famotidine, and ranitidine groups, these numbers were found to be \( 7.29 \pm 0.43 \), \( 0 \pm 0 \), \( 0.16 \pm 0.08 \), and \( 2.54 \pm 0.76 \), respectively. Lansoprazole, famotidine, ranitidine, and 5, 10, and 20 mg/kg doses of montelukast reduced the ulcer areas at a rate of 100%, 97.8%, 65.2%, and 59.1%, 69.7%, and 72.2%, respectively, compared to the control (healthy) group. Treated groups were compared to both indomethacin and control (healthy) groups.

Comparison of enzyme activities in rat stomach tissues

In order to explore the effects of antioxidant defenses on the ulceration process in all gastric tissues, the antioxidant levels (SOD, CAT, GR, and GSH) were evaluated. The results are presented in Table 2 and show that SOD for indomethacin-administered groups was lower and CAT level was higher than those for the healthy rat group. However, as compared with CAT and SOD levels in montelukast and ranitidine-administrated gastric tissues, the opposite results were found for the levels of CAT and SOD activities in indomethacin-administered tissues. In contrast to indomethacin-administered tissues, all doses of montelukast and ranitidine increased SOD activity (\( P<0.05 \) and \( P<0.01 \)) and decreased CAT activity (\( P<0.05 \) and \( P<0.01 \)).

Table 1. Effects of different doses of the montelukast (MLK) and a single dose of famotidine (FAM), ranitidine (RAN), and lansoprazole (LAN) on indomethacin (IND)-induced gastric damage in rats

<table>
<thead>
<tr>
<th>Treatment</th>
<th>N</th>
<th>Dose (mg/kg body wt)</th>
<th>Ulcer index (UI)*</th>
<th>% Inhibition*</th>
</tr>
</thead>
<tbody>
<tr>
<td>IND + MLK</td>
<td>6</td>
<td>5</td>
<td>2.98 ± 0.88**</td>
<td>59.1</td>
</tr>
<tr>
<td>IND + MLK</td>
<td>6</td>
<td>10</td>
<td>2.21 ± 0.91**</td>
<td>69.7</td>
</tr>
<tr>
<td>IND + MLK</td>
<td>6</td>
<td>20</td>
<td>2.03 ± 0.49***</td>
<td>72.2</td>
</tr>
<tr>
<td>IND + FAM</td>
<td>6</td>
<td>25</td>
<td>0.16 ± 0.08***</td>
<td>97.8</td>
</tr>
<tr>
<td>IND + RAN</td>
<td>6</td>
<td>25</td>
<td>2.54 ± 0.76***</td>
<td>65.2</td>
</tr>
<tr>
<td>IND + LAN</td>
<td>6</td>
<td>30</td>
<td>0***</td>
<td>100</td>
</tr>
<tr>
<td>IND</td>
<td>6</td>
<td>25</td>
<td>7.29 ± 0.43***</td>
<td></td>
</tr>
<tr>
<td>Control (healthy)</td>
<td>6</td>
<td>—</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

Groups treated with three doses of MLK or one dose of FAM, RAN, or LAN were compared with both the IND group (**\( P<0.01 \) and ***\( P<0.005 \), as compared with the IND group) and healthy group (\( *P<0.05 \) and \( **P<0.005 \), as compared with the healthy group). *Mean damage index ± S.E.M. of six animals in each group. Ulcer index (UI): Ulcerated area / Total stomach area × 100. % Inhibition in ulcer index in relation to the indomethacin group. N: the number of rats.
can be seen from this figure, the injection of indomethacin increased MPO activity in comparison to healthy rat tissues. Both doses of montelukast ($P<0.01$) and ranitidine ($P<0.05$) significantly decreased the level of MPO activity.

**Discussion**

The present study macroscopically evaluated the gastroprotective effects of three doses (5, 10, and 20 mg/kg body weight) of montelukast on indomethacin-induced gastric damage in rats and compared these with the effects of three positive controls [lansoprazole (30 mg/kg body weight), famotidine (25 mg/kg body weight), and ranitidine (25 mg/kg body weight)]. Oral administration of the 25 mg/kg body wt. dose of indomethacin induced significant gastric damage in the rats. However, while montelukast, lansoprazole, famotidine, and ranitidine reduced the development of indomethacin-induced gastric damage, this reduction occurred at a greater magnitude for montelukast, famotidine, and lansoprazole than for ranitidine, an H$_2$-receptor blocker (Table 1). Sener et al. (36) have shown the gastroprotective effects of montelukast in alendronate-induced lesions of the rat gastric mucosa. They reported that the gastroprotective effect of montelukast is related to its effects on the antioxidants and MPO activity. On the other hand, there remains no report about the gastroprotective effects of montelukast on NSAID-induced gastric ulcers. NSAIDs, like indomethacin, cause mucosal damage by interfering with PG synthesis, thus increasing acid secretion and the back diffusion of H$^+$ ions, and resulting in overproduction of leukotrienes and other products of the 5-lipoxygenase pathway (37, 38). While it is generally accepted that the ulcerogenic activity of NSAIDs is related to their ability to inhibit endogenous PG synthesis by blocking COX-1 and COX-2 (37), recent studies have shown that the inhibi-

<table>
<thead>
<tr>
<th>Treatment</th>
<th>N</th>
<th>Dose (mg/kg body wt)</th>
<th>CAT activity (mmol·min$^{-1}$·mg tissue$^{-1}$)</th>
<th>SOD activity (mmol·min$^{-1}$·mg tissue$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>IND + MLK 6 5</td>
<td>115.6 ± 0.4$^*$ &amp; 119.5 ± 0.8$^{**}$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IND + MLK 6 10</td>
<td>112.7 ± 0.4$^*$ &amp; 123.2 ± 0.8$^{**}$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IND + MLK 6 20</td>
<td>107.3 ± 1.2$^*$ &amp; 126.6 ± 0.4$^{**}$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IND + LAN 6 30</td>
<td>127.0 ± 0.7$^{<strong>}$ &amp; 115.9 ± 0.3$^{</strong>}$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IND + MLK 6 30</td>
<td>ND</td>
<td>ND</td>
<td></td>
<td></td>
</tr>
<tr>
<td>IND + RAN 6 25</td>
<td>57.7 ± 0.9$^{<strong>}$ &amp; 119.6 ± 0.9$^{</strong>}$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IND 6 25</td>
<td>119.9 ± 0.9$^{<strong>}$ &amp; 83.6 ± 0.7$^{</strong>}$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control (healthy) 6</td>
<td>—</td>
<td>71.7 ± 0.5</td>
<td>126.3 ± 0.4</td>
<td></td>
</tr>
</tbody>
</table>

Groups treated with three doses of MLK or one dose of LAN, FAM, or RAN were compared with both the IND group ($^*P<0.05$ and $^{**}P<0.01$, as compared with the IND group) and healthy group ($^#P<0.05$ and $^{##}P<0.01$, as compared with the healthy group). Results are means ± S.E.M. of three measurements. N: the number of rats.
tion of the PG secretion is not the only significant pathogenic factor for the mucosal damage (39 – 41). 
ROS also plays an important role in the mucosal damage caused by ethanol, indomethacin, and other agents (26, 42 – 45). Recent studies have shown that pro-oxidants expeditiously block the antioxidant systems of mucosal cells, causing ROS formation and oxidative damage (26, 44, 45).

Organisms do, however, have both enzymatic and non-enzymatic defense mechanisms against the toxicity and tissue damage of ROS (43). SOD, GST, and CAT are some of the antioxidant enzymes that contribute to the enzymatic defense mechanisms. However, our results support previous findings that SOD activity in rat stomach tissues is decreased by NSAIDs (26, 43 – 46). In our study, SOD activity was inhibited by indomethacin (P<0.01), while all doses of montelukast, lansoprazole, and ranitidine increased SOD activity near the levels of the control groups (P<0.05 and P<0.01) (Table 2). These results indicate that SOD plays an important role in eliminating gastric damage by partially preventing oxidative damage. However, SOD destroys the highly reactive radical superoxide (O$_2^-$) by converting it into the less reactive peroxide (H$_2$O$_2$), which can be destroyed by a CAT reaction.

CAT is a highly reactive enzyme that reacts with H$_2$O$_2$ to form water and molecular oxygen. It can also form methanol, ethanol, formic acid, or phenols by donating hydrogen. In the present study, we established that all doses of montelukast and ranitidine decreased CAT activity (Table 2), which had been increased by indomethacin (26, 44, 45). On the other hand, Kaleli et al. (47) reported that montelukast has a decreased effect on the CAT activity and an increased effect on the SOD activity in the sera of patients with bronchial asthma. The effects of CAT on activities of COX enzymes were investigated in another study. In this study, Chen et al. (48) suggested that CAT stimulated the expression of mRNA and proteins for COX-2 in the rats’ aortic smooth muscle cells, despite not affecting the expression of...
those for COX-1. That is, CAT exerts a biphasic effect on PG synthesis and enhances PG production when externally administered at low concentrations. This reflects a close relationship between CAT activity and PG levels. In addition, increases in CAT activity may be related to levels of other antioxidants. However, in all cases, one of the factors causing formation of an indomethacin-induced gastric ulceration process is probably an augmentation of CAT activity, as identified by the results of the present experiment. That is to say, an increase in CAT activity can be considered a marker of the NSAID-induced ulceration process.

GSTs comprise a multiage family of isoenzymes responsible for the detoxification of xenobiotics in aerobic organisms. Endogenous substrates include the toxic products of tissue damage, including both the hydroperoxide products of oxidative damage (including lipid peroxides) and aromatic xenobiotics. GSTs are catalysts of reactions in which reduced GSH acts as a nucleophile, conjugating with and facilitating removal or reduction of a second substrate. In all organisms with GST activity, multiple forms of the enzyme (which may or may not be tissue-specific) have been discovered (21). In the present study, it was determined that GST activity was elevated by montelukast in indomethacin-induced gastric tissues (Fig. 2). Montelukast may be seen as a chemopreventive agent that activates the GST enzyme. GSH and GSH-bound enzymes in tissues, particularly glutathione peroxidase (GPx) and GST, have been proposed as potential chemopreventive agents for their antioxidant and detoxification properties (49).

Reduced GSH plays pleiotropic roles, including maintaining cells in a reduced state, serving as an electron donor for certain antioxidative enzymes (e.g., glutathione peroxidase) and forming conjugates with some harmful endogenous and xenobiotic compounds via catalysis of GST. GSH levels are maintained by two systems. One is a process of synthesizing de novo from building blocks, glutamate, cysteine, and glycine via two ATP-consuming steps involving c-glutamylcysteine synthetase (cGCS) and glutathione synthetase. The other constitutes a recycling system involving GR, a flavoprotein, and reducing oxidized glutathione (GSGG) back to GSH in an NADPH-dependent manner (50). Thus, GR indirectly participates in the protection of cells against oxidative stress. The enzymatic activity of GR has been investigated in various tissues under physiologic and pathologic conditions (51).

The gastroprotective effects of the montelukast may be related to alterations in the GSH levels and GR activity of rat gastric tissues because of the alterations in these antioxidant parameters occurring in indomethacin-treated rats. In the present study, we determined that treatment with montelukast in all doses (Table 3) increases those GSH levels in the rat stomach that had previously been decreased by indomethacin (Table 3). Moreover, montelukast alleviated indomethacin-induced GR activities at all doses. The present data reveals that GSH levels are decreased in the ulceration process, while ulcerated tissues respond to this by amplifying GR activity. Similar results have been reported by other researchers (36, 45, 47, 52 – 54).

The MPO assay has had widespread use as an index of neutrophil infiltration in various gastric injuries (23, 26, 44, 45, 55, 56). As shown in Fig. 3, the MPO activity in indomethacin-administered rat stomach tissues increases in comparison with that occurring in the tissues of healthy rats (P<0.05). The increase in this enzyme activity level may be associated with increases in the levels of neutrophil infiltration and H2O2 in those gastric damaged tissues administered with indomethacin. The activity levels of these enzymes were alleviated by each dose of montelukast, acting counter to the indomethacin. Similarly, lansoprazole and ranitidine also increased MPO activity. It has been reported that the release of MPO from gastric cells is another indication of the degree of ulceration, with NSAIDs such as indomethacin also exerting their effects via inhibition of MPO pathways (23, 26, 44, 45, 55, 56). Tissue MPO activity is a sensitive and specific marker of acute inflammation and reflects polymorphonuclear cell infiltration into the parenchyma. The effect of montelukast on decreasing MPO activity may be related to its gastroprotective ability.

In conclusion, this experiment showed that indomethacin successfully induced ulcers in rat stomachs, while the experimental drugs (montelukast, lansoprazole, famotidine, and ranitidine) all reduced them. Moreover, montelukast, famotidine, and lansoprazole all effected greater reductions than did ranitidine. The levels of the anti-oxidant system enzymes (SOD, GR, GST, and CAT) and GSH were adversely affected by ulcer induction. However, montelukast, lansoprazole, famotidine, and ranitidine alleviated the adverse effects of ulceration on these enzymes and the GSH. The gastroprotective properties of montelukast may be related to its positive effects on the antioxidant system and MPO activity of rats affected by indomethacin-induced gastric ulcers. Steroidal drugs are very important in the treatment of asthma. Therefore, adjunctive treatment of montelukast with inhaled corticosteroids improves the asthmatic conditions. However, asthma treatment with steroidal drugs requires a long-term application and carries some limitations such as gastrointestinal compliance. The effect of corticosteroids on ulcer healing is similar to that of the PG synthetase inhibitor indo-
methyla-

We argue that combining montelukast with cortico-
steroids in asthma treatment can provide a greater
efficacy as well as remove the need for use of an
additional gastroprotective agent to reduce steroid-
related stomach damage. Thus, montelukast can reduce
the cost of asthma treatments that require chronic drug
application.

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Antiulcer and Antioxidant Activity of Montelukast