Research Article **ISSN:2456-1622**

Asia Pacific Journal of Pharmaceutical and Applied Sciences

EVALUATION OF ANTIULCER POTENTIAL OF ADENOSINE IN INDOMETHACIN INDUCED ULCER IN RATS

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ABSTRACT

This study investigated the anti-ulcer effects of adenosine on indomethacin-induced gastric ulcers in Wistar rats. The rats were divided into six groups: normal control, indomethacin control, indomethacin + lansoprazole (standard treatment), indomethacin + adenosine (100mg/kg), indomethacin + adenosine (150mg/kg), and indomethacin + theophylline. Indomethacin administration significantly increased ulcer score, ulcer index, and levels of malondialdehyde (MDA) and myeloperoxidase (MPO), while decreasing gastric mucus, pH, glutathione (GSH), and superoxide dismutase (SOD) levels. Treatment with both doses of adenosine, as well as the standard treatment lansoprazole, significantly reversed these effects, indicating a protective effect against indomethacin-induced gastric damage. The higher dose of adenosine (150mg/kg) showed greater efficacy compared to the lower dose (100mg/kg). The study concludes that adenosine, by acting on purinergic receptors, exhibits gastroprotective effects and holds potential for treating gastric ulcers.

Keywords: Indomethacin, Peptic ulcer, Gastric mucus, Adenosine, etc.

Submitted: 23 October 2024; Revised: 17 November 2024; Accepted: 25 November 2024

INTRODUCTION

Gastric ulcers, also known as peptic ulcers, are deep lesions that penetrate the entire thickness of the gastrointestinal tract (GIT) mucosa [1,2]. They develop due to an imbalance between aggressive factors, such as *Helicobacter pylori* infection, NSAIDs, and gastric acid, and protective factors like mucin, bicarbonate, and prostaglandins [3,4]. This imbalance disrupts mucosal integrity, leading to localized erosion of the stomach lining. Common symptoms include abdominal pain, gastrointestinal bleeding, and other digestive issues [5].

The most prevalent cause of gastric ulcers is infection with *Helicobacter pylori* (*H. pylori*), a bacterium that affects the stomach. The exact mode of transmission of *H. pylori* remains unclear, but it is believed to spread through contaminated food and water. Historical data suggests that the 19th century saw a significant increase in the prevalence of peptic ulcer disease in Western countries, potentially due to changes in the epidemiology of *H. pylori* infections [6,7].

Another major cause of gastric ulcers is the use of non-steroidal anti-inflammatory drugs (NSAIDs). NSAIDs inhibit the enzyme cyclooxygenase (COX), which in turn reduces the production of prostaglandins (E2 and I2) [8,9]. This decrease in prostaglandins impairs mucosal protection and promotes ulcer formation. NSAIDs, such as indomethacin, also affect mitochondria by inhibiting oxidative phosphorylation and generating reactive oxygen species (ROS) like superoxides and hydrogen peroxide. These free radicals cause lipid peroxidation and apoptosis, further damaging the gastric mucosa [10,11].

In addition to these physiological causes, several behavioral and lifestyle factors contribute to the development of gastric ulcers. These include smoking, frequent use of steroids, hypercalcemia, and excessive alcohol consumption [12]. According to the latest WHO data published in May 2014, peptic ulcer disease accounted for 85,487 deaths in India, representing 0.96% of total mortality. The age-adjusted death rate was 9.12 per 100,000 population, placing India 26th globally [13].

Endogenous purines such as adenosine, adenosine 5′-triphosphate (ATP), ADP, AMP, uridine 5′-triphosphate (UTP), UDP, and UDP-glucose activate P1, P2X, or P2Y purinoceptor families, which are widely and differentially distributed across the enteric nervous system (ENS) and non-neuronal cells in the gastrointestinal tract (GIT) [14]. Experimental and tissue distribution studies suggest that up to 14 of the 18 purinoceptor subtypes are involved in secretomotor reflexes within the GI tract. The term *purinergic receptors* were introduced to describe classes of membrane receptors that, upon activation by neurally released ATP (P2 purinoceptors) or its breakdown product, adenosine (P1 purinoceptors), mediate relaxation of

gut smooth muscle [15]. P1 purinoceptors, or adenosine (ADO) receptors, mediate the effects of endogenous adenosine and its analogs. These receptors are categorized into four subtypes—A1, A2A, A2B, and A3—all of which are G-protein-coupled receptors (GPCRs). Adenosine is a critical endogenous regulator of various physiological processes, including blood flow, neuronal activity, seizure activity, and airway resistance [16].

Emerging evidence indicates that adenosine and its receptors play vital roles in controlling gastric acidity and modulating gastric responses to histamine and acetylcholine [25–30]. Purinergic signaling is rapid during synaptic neurotransmission, neuromuscular transmission (causing smooth muscle contraction or relaxation), and exocrine or endocrine secretion. Additionally, its role in regulating long-term processes such as cell proliferation, differentiation, migration, and death has been well-documented in contexts like development, regeneration, and wound healing [17].

P2X and P2Y receptors contribute significantly to embryonic development, including the nervous system, limb bud cartilage, mesonephros, retina, myotubes, and neuromuscular junctions, by direct actions or modulation of other signaling pathways. Adenosine inhibits gastric acid secretion either directly by acting on parietal cells or indirectly by stimulating somatostatin release, which inhibits acid production. Moreover, adenosine facilitates tissue protection and repair through mechanisms such as improving the oxygen supply-demand ratio, preconditioning, anti-inflammatory effects, and promoting angiogenesis. By downregulating inflammatory and immune responses in injured tissues, adenosine plays a key role in its protective effects [18,19].

Theophylline, widely used to treat airway diseases, is believed to act as a non-selective phosphodiesterase (PDE) inhibitor. While its exact mechanism of action remains unclear, it is known to affect smooth muscle and circulating inflammatory cells at plasma levels lower than those needed to impact airway smooth muscle. Therefore, the present study was designed to investigate the effects of adenosine on purinergic receptors and their potential role in treating gastric ulcers.

MATERIALS AND METHODS

Experimental Animals

Wistar albino rats of either sex, weighing 200-250g were employed in the present study. They were fed on standard chow diet (Ashirwad Industries Pvt Ltd, Ropar, Punjab, India). Food and water were provided ad *libitium* throughout experimental period. They were housed in departmental animal house and were exposed to 12h light and 12h dark cycles. All animals were maintained as per the CCSEA guidelines for the care and use of Laboratory Animals, the experimental protocol used in the present

study was approved by Institutional Animal Ethics Committee.

Drugs and Chemicals

Indomethacin (Octane biotech Pvt. Ltd. Lucknow), Adenosine (central drug house pvt. Ltd) Theophylline (central drug house pvt. ltd), Lansoprazole (Casca remedies Pvt. Ltd Kuldeep Nagar, Ambala), EDTA (Loba chemie pvt. Ltd Tarapur MIDC, Mumbai), Tris buffer (Finar chemicals Ltd. Ahmadabad), Acetic acid (Ozone International – Mumbai, India), DTNB (Chemika Reagents Ltd. Mumbai), Trichloroacetic acid (Research lab fine chem. Industries- Mumbai, India). The kits for TBARS, GSH and MPO were obtained from Transasia bio-medicals Ltd. Baddi (HP). All drug solutions were freshly prepared before use.

Experimental Design

Six groups of wistar rats will be employed in the present study. All animals will be randomly divided into these groups. Each group will be comprised of six animals (n= 6).

Group I: (Normal Control)

Normal rats will be maintained on standard chow diet and water *ad libitum* for 14 days. No treatment will be given to these rats.

Group II: (Indomethacin control 5mg/kg)

Indomethacin (5mg/kg/day p.o.) will be administered to rats on standard chow for fourteen days.

Group III: (Indomethacin-5mg/kg+Lansoprazole-50mg/kg)

Indomethacin (5mg/kg/day p.o.) will be administered to rats for seven days and lansoprazole (50mg/kg/day p.o.) will be administered for another seven days along with Indomethacin treatment.

Group IV: (Indomethacin-5mg/kg+Adenosine 100mg/kg)

Indomethacin (5mg/kg/day p.o.) will be administered to rats for seven days and Adenosine (100mg/kg p.o.) will be administered for another seven days along with Indomethacin treatment.

Group V: (Indomethacin-5mg/kg+ Adenosine 150mg/kg)

Indomethacin (5mg/kg/day p.o.) will be administered to rats for seven days and Adenosine (150mg/kg/day p.o.) will be administered for another seven days along with Indomethacin treatment.

Group VI: (Indomethacin-5mg/kg+ Theophylline 5mg/kg)

Indomethacin (5mg/kg/day p.o) will be administered to rats for seven days and theophylline (5mg/kg p.o) will be administered for another seven days along with Indomethacin treatment.

Indomethacin Induced Gastric Ulcer

The gastric ulcers were induced by administering Indomethacin for 14 days (Sameh *et al.*, 2015). The rats were sacrificed on the 15^{th} days by cervical dislocation under anesthesia by urethane (1.25 gm/kg, i.p.). Stomachs were isolated and open along the greater curvature. The stomach was washed with ice-cold saline and the glandular portion was then exposed and examined for ulceration.

Estimation of ulcer score and ulcer index

The animals were sacrificed by cervical dislocation, Then the stomachs were removed and dissected along its greater curvature and were finally fixed on a cork mat or transparent glass. The dissected stomachs were moistened with normal saline to prevent autolysis. The stomachs were examined by Microscope and hand magnifying lens. Several methods have been designed to assess the extent of ulcerations and subsequently the calculation of an ulcer index as well as the protective and/or curative ratios for the ulcers. Based on their intensity, the ulcers were given scores as follows:

0 = no ulcer,

0.5 = red coloration,

1 = superficial mucosal erosion,

1.5 = Hemorrhagic streak,

2 = deep ulcer or transmural necrosis,

3 = perforated or penetrated ulcer.

The ulcer index was calculated using the following equation. (Khare *et al.*, 2008)

$$\textbf{Ulcer index} = \frac{\text{Arithmetic mean of intensity in a group} + \text{number of ulcer positive animals}}{\text{Total numbers of animal} \times 2}$$

$$\textbf{\% Inhibition} = \frac{\text{Ulcer index in control group} - \text{Ulcer index in test group}}{\text{Ulcer index in control group}} \times 100$$

Estimation of adherent mucin:

Adherent mucin was estimated by method of Core *et al*., Alcian blue, which stains acidic mucin was used for quantitative estimation of adherent mucin:

Step:-1. Isolated stomachs socked separately in solution of alcian blue (10mg/10ml), sodium acetate (200mg/50ml) and sucrose (2.73mg/50ml) for two hours.

Step:-2. Dye removed by washing with sucrose (4.2mg/50ml).

Step:-3. Mucus complexed with dye was diluted in 10ml of magnesium chloride (5.08mg/50ml) for two hours.

Step:-4. The resulting blue solution is used for calculating optical density of adherent mucin using spectrophotometer at 605nm. The mean absorbance was calculated. Six additional animals were taken for estimation of normal adherent mucin. Results were analyzed by converting mean absorbance of each group to percentage of mucin content in comparison to normal group.

$$\text{Estimation of mucin \%} = \frac{\text{Mean absorbance of test group}}{\text{Mean absorbance of normal group}} \times 100$$

mean absorbance of normal animals = 0.6

Estimation of gastric pH:

The rats were sacrificed by cervical dislocation, and the stomachs were dissected out. Gastric juice was collected and drained into test tubes and then centrifuged at 1000 rpm for 10 min and the volume of supernatant was noted. The pH of the gastric juice was recorded by digital pH meter (Chetan *et al.*, 2013).

Macroscopic evaluation of stomach:

The rats were sacrificed by cervical dislocation and the stomachs were dissected out, opened along the greater curvature, rinsed with saline to remove gastric contents and blood clots. Then stomachs were examined by a 10X magnifier lens to assess the formation of ulcers. The numbers of ulcers were counted.

Collection of samples

For biochemical estimation in the stomach tissue, the animals were sacrificed by cervical dislocation. The stomachs were removed and homogenized in phosphate buffer (pH 7.4) The clear supernatant, obtained after centrifugation at 3000 rpm for 15 min, was used to estimate , (MDA) thiobarbituric acid reactive substances (TBARS), reduced glutathione (GSH) level ,Myeloperoxidase activity and superoxide dismutase.

Estimation of thiobarbituric acid reactive substances

The quantitative measurement of thiobarbituric acid reactive substances (TBARS), an index of lipid peroxidation in a tissue was performed according to method of Nichans and Samuelson (1968). In this method, malondialdehyde (MDA) and other TBARS were measured by their reactivity with thiobarbituric acid in an acidic condition to generate pink colored chromophore which is measured spectrophotometrically at 535 nm. To 1.0 ml of supernatant of tissue homogenate, 2 ml of trichloroacetic acid-thiobarbituric acid-hydrochloric acid (TCA-TBA-HCl) reagent was added and mixed thoroughly. The mixture was kept in a boiling water bath for 15 min. After cooling, the tubes were centrifuged at 10000 g for 10 min. The colour developed in the supernatant was measured at 535 nm against blank reagent. A series of standard solutions of tetra methoxy propane in the concentration of 1 to 10 nM was treated in the similar manner. Values were expressed as nano moles per mg of protein.

Preparation of TCA-TBA-HCl reagent:

15% TCA, 0.25 N HCl and 0.375% TBA were freshly prepared and mixed in the ratio of 1:1:1 before use.

Preparation of 1, 1, 3, 3-tetramethoxy propane:

0.82 ml of 1, 1, 3, 3-tetramethoxy propane was diluted to 5 ml with distilled water to make 1 M solution. 1 ml of this dilution was further diluted to 10 ml with distilled water and this dilution process was further repeated for seven times to get 10 nM of 1, 1, 3, 3-tetramethoxy propane.

Estimation of reduced glutathione

Reduced glutathione (GSH) content of gastric tissue was estimated using method of Beutler *et al*.

(1963). The supernatant of tissue homogenate was mixed with TCA (10% w/v) in 1:1 ratio. The tubes were centrifuged at 1000 rpm for 10 min at 4°C. The supernatant obtained (0.5ml) was mixed with 2 ml of 0.3 M disodium hydrogen phosphate. Then 0.25 ml of 0.001 M freshly prepared DTNB dissolve in 1% w/v sodium citrate was added and absorbance was noted using spectrophotometer at 412 nm. A standard curve was plotted using 10-100 µM of reduced form of glutathione and results were expressed as micromoles of reduced glutathione per mg of protein.

Preparation of 0.3 M disodium hydrogen phosphate

4.26 g of anhydrous disodium hydrogen phosphate was dissolved in 100 ml distilled water.

Preparation of DTNB in 1% Sodium citrate

7.92 mg of DTNB was dissolved in 20 ml of 1% sodium citrate.

Preparation of 100 µM of GSH

12.28 mg of GSH was dissolved in 400 ml distilled water.

Estimation pf Myeloperoxidase Activity

The Myeloperoxidase (MPO) activity which is measured as an index of neutrophils accumulation was measured using method of Krawisz *et al*. (1984). In the pellet obtained after tissue homogenization 10 ml of ice-cold potassium phosphate buffer (pH 6.0) containing 0.5% hexadecyl trimethyl ammonium bromide (HETAB) and 10 mM ethylene diamine tetra acetic acid (EDTA) was added and subjected to one cycle of freezing and thawing and then Sonication for 15s was done. The contents were centrifuged at 15,000 g for 20 minutes. 0.1 ml of supernatant obtained after centrifugation was mixed with 2.9 ml of phosphate buffer containing 0.16 mg/ml of *o*-dianisidine hydrochloride and 0.0005% hydrogen peroxide (H_2O_2). The change in absorbance was measured using spectrophotometer at 532 nm. The MPO activity was expressed as unit per gram of tissue weight where 1 unit is the quantity of enzyme able to convert 1 µM of H_2O_2 to water in 1 minute at room temperature. The calculation of MPO activity was done using formula:

MPO activity (U/g) = X / Weight of the tissue

Where X = 10 x change in absorbance per minute/volume of supernatant taken in ml

Preparation of potassium phosphate buffer containing HETAB and EDTA

Disodium hydrogen phosphate (60.5 g) and potassium dihydrogen phosphate (46 g) were dissolved in 1 L of distilled water to make potassium phosphate buffer (pH 6.0). 5 g of HETAB and 3.72 g of EDTA were dissolved in 1 L of phosphate buffer (pH 6.0) to make final solution.

Preparation of phosphate buffer containing o-dianisidine hydrochloride and H_2O_2

O-dianisidine hydrochloride (16.7 mg) was dissolved in 100 ml of potassium phosphate buffer (pH 6.0) followed by addition of 1 µL of H_2O_2.

Estimation of SOD antioxidant enzyme concentration

Super oxide dismutase (SOD) activity was determined according to the method described by Marklund and marklund. The supernatant of tissue homogenate was mixed with 0.1 M phosphate saline buffer (1:4 (w/v), pH 7.4) and the homogenates centrifuged at 2500 rpm for 10 min at 4°C. The reaction

mixture consisted of 0.5 ml of TRIS-buffer (50 mM; pH 8.2), 0.5 ml pyragallol (0.5 mM), 0.5 ml EDTA (1 mM) and in different volume, 0.025 ml, 0.05 ml, 0.75 ml and 0.1 ml of tissue homogenate. The change in absorbance was recorded at 420 nM. Activity was reported by its ability to inhibit 50% reduction of pyragallol and result is expressed as Unit/ml protein.

Preparation of Tris- EDTA buffer pH 8.2

A weight of 2.85 g of tris and 1.11 g of EDTA- Na_2 were dissolved in 1 liter of distilled water.

Preparation of Pyrogallol solution (0.2 mM)

A weight of 0.252 g of pyrogallol was dissolved in a solution of 0.6 ml of concentration hydrochloric acid diluted in 1 liter of distilled water.

Calculation of SOD activity

$$\% \text{ Inhibition of pyrogallol autoxidation} = \frac{\Delta A \text{ test}}{\Delta A \text{ test}} \times 100\%$$

$$\text{SOD activity (U/ml)} = \frac{\% \text{ inhibition of pyrogallol autoxidation}}{50\ \%}$$

Statistical Analysis

All values will be expresses as mean ± S.D. The data obtained from various groups will be statistically analyzed using one way ANOVA followed by Turkey's multiple comparison tests.

The p value <0.05 was considered to be statistically significant

RESULTS AND DISCUSSION

Effect of Adenosine on ulcer score, ulcer index and percentage of ulcer inhibition in Indomethacin-induced gastric ulcer in wistar rats.

Administration of Indomethacin 5mg/kg orally for 14 days daily caused a significant ($p<0.005$) increase in ulcer score and ulcer index. Indomethacin administration showed ulceration in the glandular area of the stomach compared to normal control rats. Treatment with oral dose of Adenosine (100mg/kg, 150mg/kg) and lansoprazole (50mg/kg) which were test and standard drug respectively in the present study decrease the ulcer score, ulcer index and increases the percentage of ulcer inhibition (Table 1), when compared to the indomethacin treated group.

Effect of Adenosine on pH and gastric mucus in Indomethacin-induced gastric ulcer in wistar rats.

Administration of Indomethacin 5mg/kg/orally for 14 days daily caused ($p<0.005$) decrease in gastric mucus and pH. Indomethacin administration showed significant ulceration with decrease in gastric mucus and pH in the glandular area of stomach compared to normal control rats. Treatment with oral dose of Adenosine (100,150mg/kg) and lansoprazole (50mg/kg) which were test and standard drug respectively in the present study, increases the gastric mucus

and pH (Table 2). when compared to the indomethacin treated group. The high dose of Adenosine (150mg/kg) was more effective than the lower dose of adenosine. (100mg/kg).

Effect of Adenosine on malondialdehyde and myeloperoxidase in Indomethacin induced gastric ulcer in wistar rats.

Administration of Indomethacin 5mg/kg orally for 14 days daily caused a significant ($p<0.005$) increase in MPO and MDA. Administration of oral dose of Adenosine (100mg, 150mg /kg) and lansoprazole (50mg/kg) which were test and standard drugs in the present study produced a ($p<0.005$) dose dependent decrease in malondialdehyde and myeloperoxidase, when compared with the indomethacin treated group (table 3). The high dose of Adenosine was significantly more effective than the low dose in decreasing the MDA and MPO.

Effect of Adenosine on GSH and SOD in Indomethacin induced gastric ulcer in wistar rats

Administration of Indomethacin 5mg/kg orally for 14 days daily caused ($p<0.005$) decrease in GSH and SOD. Administration of oral dose of Adenosine (100,150mg/kg) and lansoprazole (50mg/kg) which were test and standard drugs in the present study produced a ($p<0.005$) dose dependent increase in glutathione and superoxide dismutase, When compared to the indomethacin treated group (Table 4). The high dose of Adenosine was more effective than the low dose in increasing the GSH and SOD.

Table:-1. Effect of Adenosine on ulcer score, ulcer index and percentage of ulcer inhibition in Indomethacin induced gastric ulcer in rats.

Groups	**Ulcer score**	**Ulcer index**	**% of ulcer inhibition**
Normal control	00	00.00	100
Indomethacin control (5mg/kg)	1.823 ± 0.409^{a}	5.42	00^{a}
Lansoprazole(50mg/kg)	0.413 ± 00.2040^{b}	1.40	74.08^{b}
Adenosine (100mg/kg)	$0.830^{a} \pm 0.248^{b}$	3.11	42.42^{b}
Adenosine (150mg/kg)	$0.4166^{a} \pm 0.2041^{b}$	1.97	63.77^{b}
Theophylline(5mg/kg)	$0.9166^{a} \pm 0.2041^{b}$	5.79	$- 6.43^{b}$

All values are expressed as Mean ± S.D; n = 6. [a] = *P*<0.05 Vs normal control, [b]= *P*<0.05 Vs Indomethacin control.

Table 2:- Effect of Adenosine on pH and gastric mucus in Indomethacin induced gastric ulcer.

Groups	**Mucin %**	**pH**
Normal control	100	3.123±0.345
Indomethacin control (5mg/kg)	76.75	1.363 ±0.392
Lansoprazole(50mg/kg)	94.37	4.583±0.285
Adenosine (100mg/kg)	85.37	4.015±0.505
Adenosine (150mg/kg)	95.06	4.59±0.264
Theophylline (5mg/kg)	67.5	1.278±0.151

All values are expressed Mean ± S.D; n = 6. [a] = *P*<0.05 Vs normal control, [b]= *P*<0.05 Vs Indomethacin control.

Table 3. Effect of Adenosine on malondialdehyde and myeloperoxidase in Indomethacin induced gastric ulcers in wistar rats.

Groups	**MDA (nmol/mg)**	**MPO (nmol/mg)**
Normal control	2.237±0.529	10.89±0.79
Indomethacin control (5mg/kg)	4.626±0.16[a]	40.34±0.38[a]
Lansoprazole(50mg/kg)	1.588±0.23[b]	12.38±0.38[b]
Adenosine(100mg/kg)	2.556±0.248[b]	23.68±0.484[b]
Adenosine (150mg/kg)	1.764±0.291[b]	14.46±0.323[b]
Theophylline(5mg/kg)	5.195± 0.3869[b]	40.91 ± 0.263[b]

All values are expressed Mean ± S.D; n = 6. [a] = *P*<0.05 Vs normal control, [b]= *P*<0.05 Vs Indomethacin control. Where Indo = Indomethacin; MPO= Myeloperoxidase; MDA= Malondialdehyde.

Table 4. Effect of Adenosine on SOD and GSH in Indomethacin induced gastric ulcers in wistar rats.

Groups	SOD (units/mg)	GSH (nmol/mg)
Normal control	46.98±2.86	6.73 ± 0.28
Indomethacin control (5mg/kg)	38.6 ±1.38[a]	1.54±0.26[a]
Lansoprazole(50mg/kg)	63.06 ± 1.02[b]	3.08±0.34[b]
Adenosine(100mg/kg)	65.45±1.03[b]	3.52±0.30[b]
Adenosine (150mg/kg)	71.48 ±2.89[b]	4.455±0.42[b]
Theophylline(5mg/kg)	34.45±3.21[b]	1.168 ± 0.12[b]

All values are expressed Mean ± S.D; n = 6. [a] = $P<0.05$ Vs normal control, [b]= $P<0.05$ Vs Indomethacin control. GSH = Glutathione; SOD= superoxide dismutase.

CONCLUSION

The study highlights the significant potential of adenosine as a treatment for gastric ulcers, particularly those induced by indomethacin. The research demonstrates that adenosine effectively mitigates the adverse effects of indomethacin on various gastric parameters. Specifically, adenosine significantly reduced the ulcer score and index, thereby inhibiting ulcer formation. It also restored critical protective factors such as gastric mucus, pH, glutathione (GSH), and superoxide dismutase (SOD), all of which are essential for maintaining gastric health but were negatively impacted by indomethacin. Furthermore, adenosine decreased levels of malondialdehyde (MDA) and myeloperoxidase (MPO), which are markers of oxidative stress and inflammation, underscoring its protective role. These promising findings strongly advocate for further research into the therapeutic potential of adenosine for gastric ulcers. Future studies could focus on conducting clinical trials in humans to confirm the efficacy and safety observed in the rat model. Additionally, determining the optimal dosage and route of administration for human treatment is essential. Long-term effects of adenosine treatment on gastric health should also be investigated, along with its potential use in combination therapies with existing anti-ulcer medications. Finally, elucidating the precise mechanisms underlying adenosine's gastroprotective effects, particularly its interaction with purinergic receptors, would provide deeper insights into its therapeutic action. By addressing these areas, future research could pave the way for the development of novel and effective therapies for gastric ulcers that leverage the beneficial properties of adenosine.

CONFLICTS OF INTEREST

The author had no competing interests.

REFERENCES

1. Rau, W., Hohaus, C., & Jessen, E. (2019). A differential approach to form and site of peptic ulcer. *Scientific reports*, *9*(1), 8683.
2. Bereda, G. (2022). Peptic Ulcer disease: definition, pathophysiology, and treatment. *Journal of Biomedical and Biological Sciences*, *1*(2), 1-10.
3. Galura, G. M., Chavez, L. O., Robles, A., & McCallum, R. (2019). Gastroduodenal injury: role of protective factors. *Current gastroenterology reports*, *21*, 1-7.
4. Niv, Y., & Banić, M. (2014). Gastric barrier function and toxic damage. *Digestive diseases*, *32*(3), 235-242.
5. Sweetser, S. (2022). Gastrointestinal manifestations of systemic diseases. *Yamada's Textbook of Gastroenterology*, 2231-2273.
6. Aljaberi, H. S., Ansari, N. K., Xiong, M., Peng, H., He, B., & Wang, S. (2023). Current Understanding of the Transmission, Diagnosis, and Treatment of H. pylori Infection: A Comprehensive Review. *International Journal of Medical, Pharmacy and Drug Research*, *7*, 2.
7. Stefano, K., Marco, M., Federica, G., Laura, B., Barbara, B., Gioacchino, L., & Gian, L. D. A. (2018). Helicobacter pylori, transmission routes and recurrence of infection: state of the art. *Acta Bio Medica: Atenei Parmensis*, *89*(Suppl 8), 72.
8. Bindu, S., Mazumder, S., & Bandyopadhyay, U. (2020). Non-steroidal anti-inflammatory drugs (NSAIDs) and organ damage: A current perspective. *Biochemical pharmacology*, *180*, 114147.
9. Islam, H., Siddiqui, A., Islam, R., Islam, T., Ahmed, S., Fahim, M., & Imtiaz, H. (2024). NSAID-induced Gastric Ulcer Disease: A Deleterious Connection. *Discovery medicine*, *36*(188), 1789-1799.
10. Sinha, K., Sadhukhan, P., Saha, S., Pal, P. B., & Sil, P. C. (2015). Morin protects gastric mucosa from nonsteroidal anti-inflammatory drug, indomethacin induced inflammatory damage and apoptosis by modulating NF-κB pathway. *Biochimica et Biophysica Acta (BBA)-General Subjects*, *1850*(4), 769-783.
11. Basak, M., Mahata, T., Chakraborti, S., Kumar, P., Bhattacharya, B., Bandyopadhyay, S. K., & Maity, B. (2020). Malabaricone C attenuates nonsteroidal anti-inflammatory drug-induced gastric ulceration by decreasing oxidative/nitrative stress and inflammation and promoting angiogenic autohealing. *Antioxidants & Redox Signaling*, *32*(11), 766-784.

12. Ravisankar, P., Koushik, O., Reddy, A., KumarU, A. P., & Pragna, P. (2016). A detailed analysis on acidity and ulcers in esophagus, gastric and duodenal ulcers and management. *IOSR Journal of Dental and Medical Sciences (IOSR-JDMS), 15*(1), 94-114.
13. Sun, J., Huang, L., Li, R., Wang, T., Wang, S., Yu, C., & Gong, J. (2023, April). Comparison of Secular Trends in Peptic Ulcer Diseases Mortality in China, Brazil and India during 1990–2019: An Age-Period-Cohort Analysis. In *Healthcare* (Vol. 11, No. 8, p. 1085). MDPI.
14. Christofi, F. L. (2008). Purinergic receptors and gastrointestinal secretomotor function. *Purinergic signalling*, *4*, 213-236.
15. Burnstock, G. (2007). Physiology and pathophysiology of purinergic neurotransmission. *Physiological reviews*, *87*(2), 659-797.
16. Burnstock, G. (2009). Purinergic signalling: past, present and future. *Brazilian Journal of Medical and Biological Research*, *42*, 3-8.
17. Halliez, M. C., & Buret, A. G. (2015). Gastrointestinal parasites and the neural control of gut functions. *Frontiers in Cellular Neuroscience*, *9*, 452.
18. Burnstock, G., & Dale, N. (2015). Purinergic signalling during development and ageing. *Purinergic signalling*, *11*(3), 277-305.
19. Burnstock, G., Verkhratsky, A., Burnstock, G., & Verkhratsky, A. (2012). Evolution of purinergic signalling. *Purinergic signalling and the nervous system*, 245-305.

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