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# Research Article
# Efficacy of 2-Chloro-3-Hydrazinylquinoxaline in Alleviating Indomethacin-Induced Gastric Ulcers: Insights from Animal Model Investigation

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## Abstract

**Background and Objective:** Nonsteroidal Anti-Inflammatory Drugs (NSAIDs) are widely used for pain and inflammation relief but are associated with remarkable risk, notably gastric damage. This damage substantially contributes to the development of stomach ulcers, a leading cause of morbidity. The current study investigated the gastroprotective effects of 2-Chloro-3-hydrazinylquinoxaline in a rat model of indomethacin-induced gastric. **Materials and Methods:** Thirty male Wistar rats were divided into five groups: Control, indomethacin only, indomethacin with varying doses of 2-Chloro-3-hydrazinylquinoxaline and indomethacin with Esomeprazole. The efficacy of 2-Chloro-3-hydrazinylquinoxaline in mitigating gastric ulcers was assessed through histopathological examinations and the measurement of inflammatory biomarkers. A one-way ANOVA was used for multiple comparisons, followed by Tukey's *post-hoc* test. **Results:** Indomethacin-induced ulcers resulted in notable epithelial damage and blood streaks on the gastric mucosa. In contrast, treatment with 2-Chloro-3-hydrazinylquinoxaline significantly ($p<0.05$) reduced ulcer formation compared to the indomethacin-only group. The 2-Chloro-3-hydrazinylquinoxaline-treated group exhibited restoration of normal epithelial tissue and minimal presence of inflammatory cells, findings that were comparable to those in the control and Esomeprazole-treated groups. Additionally, 2-Chloro-3-hydrazinylquinoxaline significantly ($p<0.05$) decreased inflammatory biomarkers while enhancing levels of gastroprotective mediator compared to indomethacin alone. **Conclusion:** These findings highlight the gastroprotective properties of 2-Chloro-3-hydrazinylquinoxaline, demonstrating its potential efficacy in treating NSAID-induced gastric ulcers.

Key words: NSAIDS, indomethacin, esomeprazole, gastric ulcer, 2-Chloro-3-hydrazinylquinoxaline, inflammatory biomarkers, ELISA

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

## INTRODUCTION

A peptic or gastric ulcer refers to a condition characterized by the erosion or damage of the stomach lining or mucosa and the duodenum, where the damage extends beyond the superficial layers into the deeper muscle tissue. This erosion is primarily attributed to the synthesis of gastric acid in the stomach environment[1]. Peptic ulcer disease continues to impose a substantial health burden globally and serves as a critical cause of hospital admissions worldwide. If left untreated, these ulcers can result in serious complications, including ulcer haemorrhage, perforation, penetration and gastric outlet obstruction, which may pose life-threatening risks and potentially lead to fatalities[2]. The management and timely detection of gastric ulcers present considerable hurdles in medical practice, necessitating comprehensive approaches to diagnosis and treatment. Addressing these challenges effectively is crucial to mitigating the adverse outcomes associated with gastric ulcers and improving patient outcomes[3-5].

Gastric ulcers arise from an imbalance between factors that impact the stomach's well-being and those that offer protective mechanisms. This intricate balance between destructive and defensive elements plays a pivotal role in the onset of gastric ulcers[6]. Several risk factors contribute to the development of peptic ulcers, including heightened gastric acid secretion, alcohol consumption, irregular motility, smoking, *Helicobacter pylori* infection and the use of Non-Steroidal Anti-Inflammatory Drugs (NSAIDs)[7]. *Helicobacter pylori* infection and the use of NSAIDs represent the primary risk factors associated with the development of peptic ulcer disease. Research indicates that individuals with these risk factors are twice as likely to develop peptic ulcer disease[8]. Tobacco use is linked to various detrimental effects on the gastrointestinal tract, including reduced secretion of bicarbonate, decreased mucous blood flow, increased inflammation and increased susceptibility to complications from *H. pylori* infection. Nicotine, found in tobacco smoke, stimulates the production of gastric acid, contributing to its classification as a risk factor for peptic ulcer disease. Similarly, alcohol consumption poses risks, as chronic alcohol intake can impair the gastric mucosal barrier by inhibiting COX-1 receptor enzymes, thereby diminishing the production of cytoprotective prostaglandins[8-10]. The manifestations of inflammation in the gastrointestinal tract can result in persistent tissue injury over time. Furthermore, an alteration in the levels of proinflammatory cytokines, such as TNF-α, IFN-γ, IL-1, IL-6 and IL-12, as well as anti-inflammatory cytokines like IL-4, IL-10 and IL-11, is thought to play a critical role in regulating the inflammatory response[11]. Specific NSAIDs, including COX-2 inhibitors, ibuprofen and naproxen, are associated with gastrointestinal complications, although the extent of risk differs among them. Blocking COX-1 in the gastrointestinal tract diminishes the secretion of prostaglandins and their protective effects on the gastric mucosa, heightening the risk of mucosal injury. Additionally, inhibiting COX-2 may contribute to mucosal damage[11,12].

In Saudi Arabia, there is widespread and significant daily usage of NSAIDs. Furthermore, *Helicobacter pylori* infection is notably prevalent among the Saudi population afflicted with gastric ulcers[13,14]. On the contrary, crucial protective factors include prostaglandin production, mucus secretion, bicarbonate generation and ensuring consistent blood flow to the tissue[7,15]. Disrupting the equilibrium between harmful factors and protective mechanisms in the gastrointestinal mucosa can result in gastric irritation and ulcer formation. Prolonged anxiety, stress, surgical shock, burns and trauma are all factors that contribute to this imbalance. Oxygen-derived free radicals are recognized to play a role in developing gastric damage induced by various human factors[6]. Indomethacin is favored as the primary option for establishing an experimental ulcer model owing to its increased ulcer-causing potential in comparison to other NSAIDs. Its mechanism involves inhibiting both Cyclooxygenase-1 (COX-1) enzymes, thereby suppressing prostaglandin synthesis. Studies suggest that indomethacin induces gastric injury by impeding the production of Prostaglandin E-2 (PGE-2), bicarbonate and mucus derived from COX-1 enzymes. Furthermore, it stimulates gastric acid secretion, raises levels of oxidant parameters and reduces antioxidant parameters[16-18]. Typical synthetic medications for treating NSAID-induced gastric ulcers, including cimetidine, misoprostol, ranitidine and omeprazole, are commonly employed. It is crucial to note that each of these medications carries a range of side effects, varying from mild to severe[19,20]. The quinoxaline structure facilitates several activities, serving as a precursor for synthesizing numerous compounds with a wide range of applications[21]. Quinoxaline compounds exhibit diverse applications and possess various biological properties beneficial in cancer therapy, antimicrobial treatment and anti-inflammatory interventions[22,23]. This study aimed to investigate the gastroprotective effects of 2-Chloro-3-hydrazinylquinoxaline against indomethacin-induced gastric ulcers, using *in vivo* animal models to assess its potential therapeutic benefits.

## MATERIALS AND METHODS

**Study area:** The study was conducted over three consecutive days in August, 2022 at the Faculty of Pharmacy, King

Abdulaziz University, Jeddah, Saudi Arabia. During this period, experimental procedures were conducted on laboratory rats to assess the gastroprotective effects of 2-Chloro-3-hydrazinylquinoxaline against indomethacin-induced gastric ulcers. The rats were administered indomethacin on the 1st day to induce gastric ulceration. Following this, they were treated with 2-Chloro-3-hydrazinylquinoxaline as the test compound. The animals were monitored closely for any signs of distress or changes in behavior throughout the study. On the 3rd day, the rats were humanely euthanized, adhering to ethical guidelines for animal research. Subsequently, their gastric tissues were collected for further analysis, including the evaluation of histopathological changes, inflammatory biomarker levels and the extent of mucosal protection provided by the compound. The collected data were then processed for statistical analysis to determine the efficacy of 2-Chloro-3-hydrazinylquinoxaline in mitigating indomethacin-induced gastric damage.

**Drugs and chemicals:** The compounds, including 2-Chloro-3-hydrazinylquinoxaline, indomethacin, esomeprazole and Sodium Carboxymethyl Cellulose (CMC-Na), were sourced from Sigma-Aldrich in St. Louis, Missouri, USA. Additionally, the experimental setup incorporated several ELISA kits, namely the Rat TNF-α ELISA kit, Rat Interferon Alpha kit (Cat No. MBS267050), Rat Prostaglandin E2 (PGE2) ELISA kit (Cat No. MBS262150), Rat Mucin ELISA (Cat No. MBS1600651), Rat Interleukin 6 (IL-6) ELISA kit (Cat No. MBS269892), Rat Inducible Nitric Oxide Synthase (iNOS) ELISA kit (Cat No. MBS723326), ELISA kit (Cat No. MBS725633) and rat IL-1β ELISA kit (Cat No. MBS825017), all obtained from Sigma-Aldrich in St. Louis, MO, USA. Various commercially available chemicals, such as formalin, phosphate buffer and other necessary compounds, were selected with elevated purity grades. It is essential to highlight that every chemical used throughout the study met the standards of analytical grade.

Esomeprazole, commonly employed as a protective agent against gastric ulceration, was a reference drug in this study. This choice aligned with previous research studies on gastroprotective actions, which often utilize esomeprazole as a reference drug[24].

**Study design:** Thirty male Wistar rats, aged 10 weeks and weighing between 200 and 230 g, were procured from the animal facility at the Faculty of Pharmacy, King Abdulaziz University. These rats were housed in a controlled environment with a temperature maintained between 20-24°C and a 12 hrs light and 12 hrs dark cycle. They were provided with unlimited access to a standard diet and water and 1 week acclimation period was observed before the commencement of experiments. Thirty rats were randomly allocated to five groups, each consisting of six rats. Rats were divided into five groups for the study: Group 1 served as the negative control, receiving the vehicle (0.5% w/v carboxymethyl cellulose sodium, 10 mL/kg) orally. Group 2 acted as the positive control, receiving a single oral dose of indomethacin (30 mg/kg). Group 3 received a combination of indomethacin and 2-Chloro-3-hydrazinylquinoxaline at a dose of 30 mg/kg for three consecutive days, with indomethacin administered orally on the 3rd day followed by the last dose of 2-Chloro-3-hydrazinylquinoxaline one hour later. Similarly, Group 4 received a combination of indomethacin and 2-Chloro-3-hydrazinylquinoxaline at 60 mg/kg for three consecutive days. Group 5 was administered esomeprazole (30 mg/kg) orally for three consecutive days, followed by oral administration of indomethacin (30 mg/kg) on the third day, with the last dose of esomeprazole given one hour later. After 4 hrs of indomethacin administration, rats in the treatment groups were euthanized for subsequent analysis[15].

**Induction of gastric ulcers:** The onset of gastric ulcers was noted after the administration of indomethacin. On the second day of the experiment, a 24 hrs fasting period was enforced permitting access solely to water. On the third day, all groups, except the control group, received intragastric administration of indomethacin at a dose of 30 mg/kg, dissolved in a 0.5% solution of Sodium Carboxymethyl Cellulose (CMC-Na).

**Analysis of histopathology:** The research utilized a grading scale ranging from 0 to 4 to assess histopathological alterations. This scale, employed by a histopathologist who was unaware of the particular treatments administered, considered various factors including edema in the gastric mucosa, infiltration of inflammatory cells, gastric hemorrhage and necrosis[15].

**Inflammatory biomarker assessment:** In assessing inflammatory biomarkers, gastric tissue homogenates underwent comprehensive analysis to measure PGE2 activity using the Rat Prostaglandin E2 (PGE2) ELISA kit (Cat No. MBS262150, St. Louis, Missouri, USA). Additionally, evaluation of IL-6, TNF-α, IFN-γ and Rat IL-β in the supernatant was performed utilizing their respective ELISA kits (Cat No. MBS269892, MBS2507393, MBS267050, MBS825017). Measurement of mucin protein levels was carried out with the Rat MUC1 ELISA kit (Cat. No. MBS1600651, St. Louis, Missouri,

USA). All procedures strictly followed the manufacturer's protocols and the kits utilized were obtained from Sigma-Aldrich in St. Louis, Missouri, USA.

**Ethical consideration:** The animal procedures conducted in this study strictly followed the approved protocols established by the Research Ethics Committee of the Faculty of Pharmacy at King Abdulaziz University, KSA (Reference No. "PH-1444-55").

**Statistical analysis:** The study data are depicted as Mean±Standard Deviation (SD). A one-way ANOVA was employed to analyze multiple comparisons, followed by Tukey's *post-hoc* test. Statistical significance was defined as a probability value (p) less than 0.05. All statistical computations were conducted using GraphPad Instat software version 3. Graphs were constructed using GraphPad Prism software version 8 (GraphPad Software, Inc., USA).

## RESULTS

**Impact of 2-Chloro-3-hydrazinylquinoxaline on the histopathological features of rat:** Histological examination was utilized to evaluate the effects of indomethacin, 2-Chloro-3-hydrazinylquinoxaline and esomeprazole on the stomach mucosa of rats. The gastric epithelial tissue in the negative control group (Fig. 1a) displayed its typical histological structure, devoid of inflammatory cells. Conversely, Fig. 1b showed inflammatory cells in rats exposed to indomethacin, with evident damage to the epithelial lining and focal ulceration. Histopathological analysis revealed less severe damage to the mucosal layer's lining epithelium, along with a reduction in inflammatory cells and hemorrhage in the 30 mg/kg treatment group (Group 3) compared to the indomethacin-treated group (Group 2) (Fig. 1c). However, the group treated with 60 mg/kg of 2-Chloro-3-hydrazinylquinoxaline (Group 4) displayed restoration of normal epithelial tissue and minimal inflammatory cells, akin to the control (Group 1) and esomeprazole-treated groups (Fig. 1d-e). This suggested that 2-Chloro-3-hydrazinylquinoxaline may act as an anti-ulcer agent for the stomach, with higher doses correlating with greater improvements in rat stomach histology.

**Effect of pre-treatment with 2-Chloro-3-hydrazinylquinoxaline on inflammatory markers (TNF-α, IL-6, IL-1β, IFN-γ and INOs):** In the experimental rat ulcer model, the efficacy of 2-Chloro-3-hydrazinylquinoxaline was investigated by evaluating its impact on molecular markers, including TNF-α, IL-6, IL-1β, IFN-γ and INOs. The study utilized molecular markers as indicators of inflammatory response in gastric tissues. Notably, exposure to indomethacin induced a pronounced pro-inflammatory reaction, as evidenced by a substantial elevation in concentrations of TNF-α, IL-6, IL-1β, IFN-gamma and INOs compared to the control group devoid of indomethacin administration.

However, pre-administration of either esomeprazole at a dose of 30 mg/kg or 2-Chloro-3-hydrazinylquinoxaline at doses of 30 and 60 mg/kg exhibited a significant anti-inflammatory effect. This effect was demonstrated by a marked reduction ($p<0.0001$) in the concentrations of TNF-α, IL-6, IL-1β, IFN-gamma and INOs compared to the indomethacin-treated group. These findings underscore the potential of 2-Chloro-3-hydrazinylquinoxaline in modulating molecular mediators involved in inflammatory processes, akin to the effects observed with esomeprazole treatment. As depicted in Fig. 2(a-e), the results suggest a comparable efficacy between 2-Chloro-3-hydrazinylquinoxaline and esomeprazole in mitigating inflammatory responses.

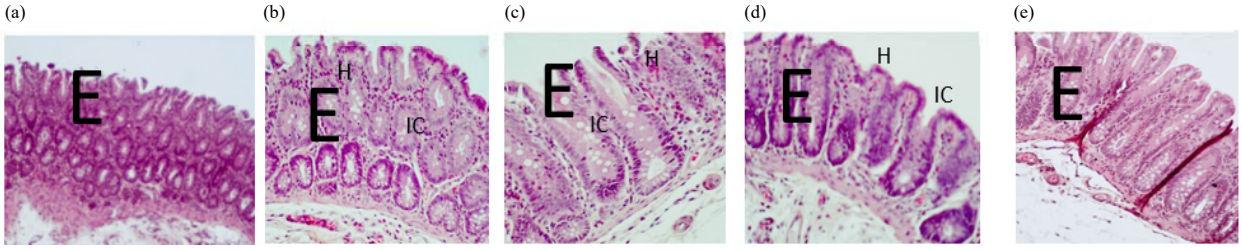


Fig. 1(a-e): Depict histopathological examination of epithelial tissue, (a) Negative control, (b) Ulcer control (indomethacin only), (c) Low dose (30 mg/kg) of 2-Chloro-3-hydrazinylquinoxaline, (d) High dose (60 mg/kg) of 2-Chloro-3-hydrazinylquinoxaline and (e) Esomeprazole (30 mg/kg)
Compared to B, C, D and E show restoration of normal E: Epithelial layer, reduced H: Hemorrhage and fewer IC: Inflammatory cells

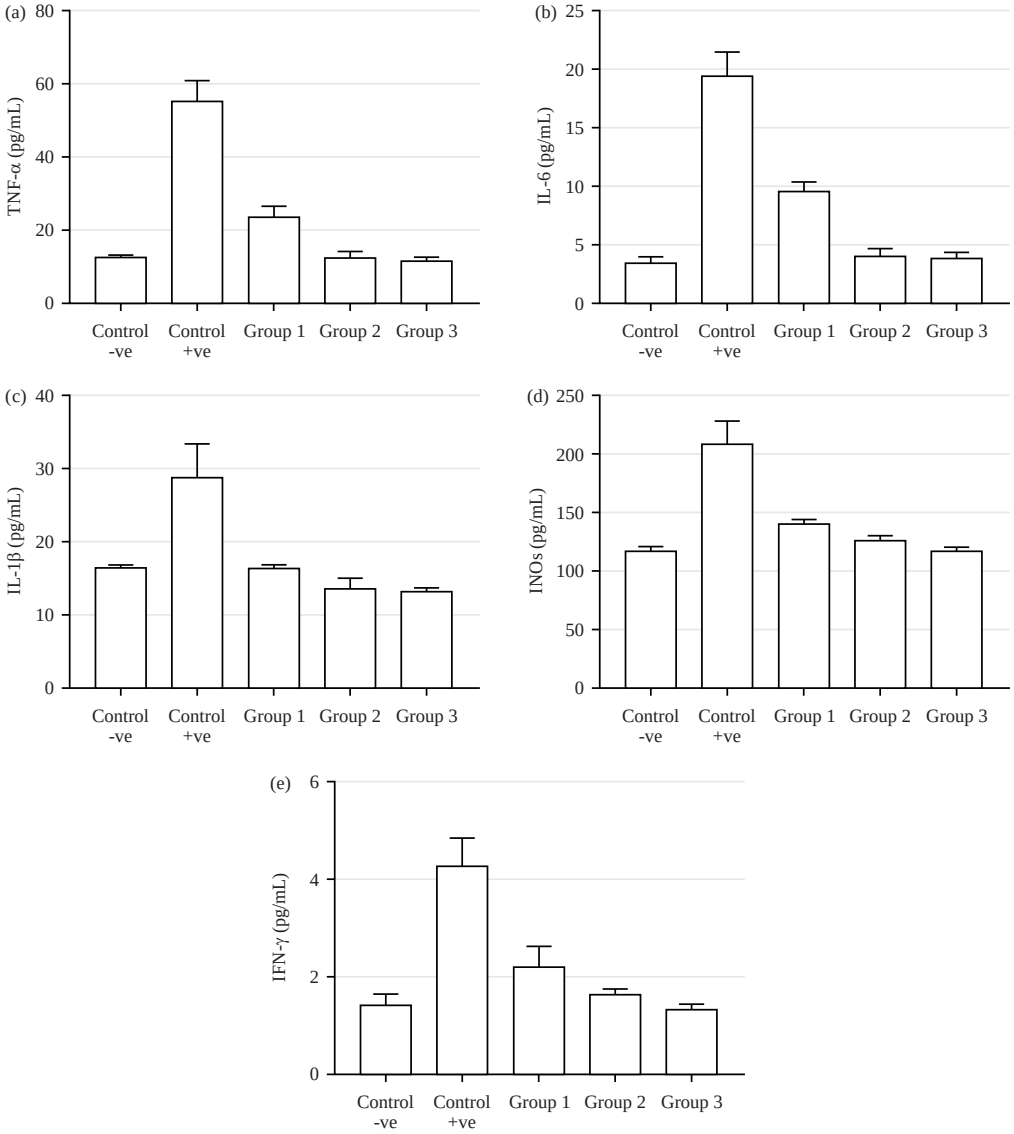


Fig. 2(a-e): Illustrates the impact of pre-treatment with 2-Chloro-3-hydrazinylquinoxaline on molecular markers, (a) TNF-α, (b) IL-6, (c) IL-1β, (d) INOs and (e) IFN-gamma in rats with indomethacin-induced gastric ulcers

Results presented as Mean±SD (n = 6) and demonstrate significant differences ($p<0.05$) compared to both control and indomethacin groups

**Effect of pre-treatment with 2-Chloro-3-hydrazinylquinoxaline on the concentrations of PGE2 and mucin:** Figure 3(a-b) shows a significant decline in mucin and prostaglandin levels after indomethacin administration compared to the negative control (untreated group). Conversely, pretreatment with esomeprazole led to a considerable ($p<0.0001$) increase in PGE2 and mucin concentrations relative to the indomethacin-exposed group. Additionally, pretreatment with 2-Chloro-3-hydrazinylquinoxaline at doses of 30 and 60 mg/kg exhibited remarkable ($p<0.0001$) elevations in PGE2 and mucin concentrations in a dose-dependent manner when compared to the indomethacin-exposed group. These results suggest a comparable efficacy between 2-Chloro-3-hydrazinylquinoxaline and esomeprazole in augmenting the concentration of both PGE2 and mucin.

## DISCUSSION

In this study, pre-treatment with 2-Chloro-3-hydrazinylquinoxaline significantly reduced the formation of indomethacin-induced gastric ulcers, as demonstrated by improved epithelial restoration and reduced inflammation. Notably, this compound decreased levels of inflammatory

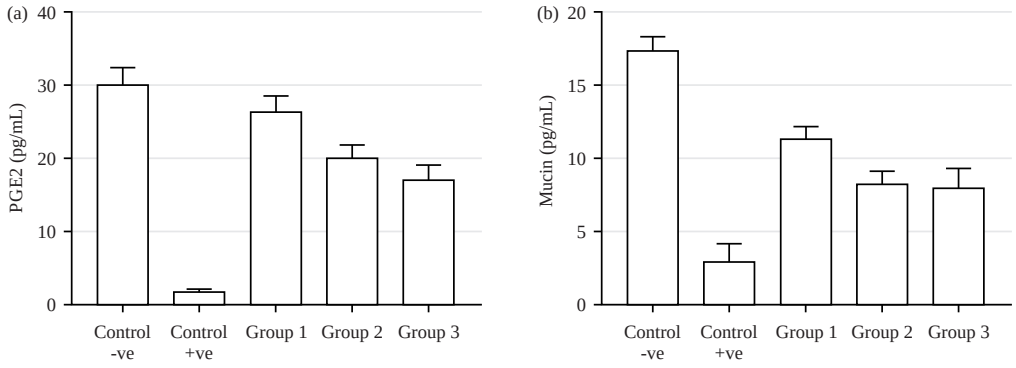


Fig. 3(a-b): Impact of pre-treatment with 2-Chloro-3-hydrazinylquinoxaline on the concentrations of key gastric components in rats with indomethacin-induced gastric ulcers, (a) PGE2 levels and (b) Mucin concentrations showing a marked elevation in the pre-treatment group relative to the control and indomethacin-only groups

(a) Demonstrating a significant increase in the pre-treatment group compared to the indomethacin group and (b) Showing a marked elevation in the pre-treatment group relative to the control and indomethacin-only groups. Data presented as Mean±SD (n = 6) and indicate statistical significance ($p<0.05$) compared to both control and indomethacin groups

biomarkers (TNF-α, IL-6, IL-1β, iNOS and IFN-gamma) while enhancing gastroprotective mediators such as PGE2 and mucin, which are essential in maintaining gastric mucosal integrity. The findings showed that the gastroprotective effects of 2-Chloro-3-hydrazinylquinoxaline are comparable to esomeprazole, a standard treatment for NSAID-induced gastric ulcers.

The NSAIDs elevate gastric and peptic ulcer occurrence by 15%, with indomethacin being a notable choice for inducing experimental ulcer models in animals[16,25]. Current therapies for gastric ulcers have drawbacks including side effects and limited efficacy[26], driving research toward the development of safe and effective plant-derived anti-ulcer medications.

The widely used NSAID-induced ulcer model hinges on disturbing the balance between anti-inflammatory and pro-inflammatory agents at the injury site. This disruption entails decreased prostaglandin E2 levels and increased secretion of IL-6 and TNF-α by epithelial cells[17,27,28]. Moreover, oral administration of NSAIDs results in the release of intercellular adhesion molecule-1 in the vascular endothelial cells of the gastric mucosa. As a result of this mechanism, increased neutrophil infiltration occurs, driven by their attachment to vascular endothelial cells under the influence of inflammatory cytokines such as Tumor Necrosis Factor α (TNF-α) and Interleukin-1 (IL-1)[29,30]. Locally, NSAIDs exert a direct effect on the gastric mucosa by disrupting oxidative phosphorylation. This disruption impairs electron transport within the mitochondrial membrane, causing the release of cytochrome C. The cytochrome C subsequently generates reactive oxygen species, which are crucial in activating proteases and initiating the process of apoptosis[30-32].

In the current study, the administration of indomethacin caused histopathological alterations and changes in the inflammatory biomarkers, mucin and prostaglandin content in rat gastric tissues. However, pre-treatment with 2-Chloro-3-hydrazinylquinoxaline at different doses reduced injuries compared to the indomethacin-exposed group. The highest dose of 2-Chloro-3-hydrazinylquinoxaline showed minimal injuries with no observable histopathological changes. Importantly, 2-Chloro-3-hydrazinylquinoxaline enhanced mucin levels at all tested doses. These findings were consistent with previous research showing that pre-treatment with tetramethylpyrazine increased mucin concentration and decreased TNF-α and IL-6 levels. Additionally, the tetramethylpyrazine-treated group exhibited improvements in histopathological changes compared to the peptic ulcer group induced by indomethacin[15].

Gastric ulceration occurs due to an increased concentration of reactive oxygen species (ROS), including hydrogen peroxide, hydroxyl radicals and superoxide anions. This oxidative stress within the gastric tissue is a key factor in developing gastric bleeding and subsequent ulcer formation[33]. Additionally, indomethacin-induced oxidative stress contributes to mitochondrial respiration uncoupling, leading to inflammation and the production of pro-inflammatory cytokines such as TNF-α and IL-6. These cytokines further enhance the expression of adhesion molecules like ICAM-1, crucial in initiating and perpetuating injury and inflammation within the gastric tissue[15,28]. In this study, pretreatment with 2-Chloro-3-hydrazinylquinoxaline significantly reduced ($p<0.0001$) the levels of Inducible Nitric Oxide Synthase (iNOS), TNF-α, IL-1 and IL-6

compared to rats treated with indomethacin, resulting in a notable decrease in inflammation. These findings were consistent with previous research of Shaik and Eid[34] demonstrated the ability of Piceatannol to attenuate the production of inflammatory cytokines. The ongoing investigation highlights that gastric injuries resulting from NSAID usage primarily occur due to the inhibition of Cyclooxygenase (COX) enzymes. This inhibition hinders the synthesis of Prostaglandin E2 (PGE2), which is crucial for gastric protection. The PGE2 plays a central role in promoting mucus production and improving gastric blood flow, thereby initiating a protective mechanism in the stomach[35].

In the current study's extensive analysis, a clear decrease in gastric Prostaglandin E2 (PGE2) and mucin levels was convincingly demonstrated following exposure to indomethacin. However, it is important to note that pretreatment with 2-Chloro-3-hydrazinylquinoxaline partially alleviated this effect, indicating its potential to mitigate the adverse impacts induced by indomethacin on gastric PGE2 and mucin concentrations. Existing literature acknowledges the healing properties of Prostaglandin E2 (PGE2), particularly its role in inducing angiogenesis by stimulating Vascular Endothelial Growth Factor (VEGF) production in fibroblasts, thus promoting cell proliferation. Moreover, recent research involving rats supported these findings, showing an increase in gastric mucin content, reduced neutrophil infiltration (as evidenced by decreased myeloperoxidase activity) and a decline in elevated serum nitric oxide levels[36,37].

The study provides compelling evidence for the gastroprotective effects of 2-Chloro-3-hydrazinylquinoxaline, suggesting that this compound could be developed as a novel therapeutic agent for the prevention and treatment of NSAID-induced gastric ulcers. Its ability to both reduce inflammation and promote epithelial healing positions it as a dual-action agent that could improve treatment outcomes for patients using NSAIDs. Clinically, this compound could be integrated into treatment regimens for patients at high risk of gastric ulcers, particularly those requiring long-term NSAID use for chronic conditions such as arthritis. Its dual action on inflammation and mucosal protection could also make it suitable for use in combination therapies with proton pump inhibitors like esomeprazole.

Further studies are necessary to validate these findings, particularly in larger animal models and clinical trials. Additional investigations should also explore the long-term safety profile of 2-Chloro-3-hydrazinylquinoxaline, including its potential interactions with other common medications. Moreover, the underlying molecular mechanisms of its gastroprotective effects should be elucidated to optimize its therapeutic potential.

One of the key limitations of this study is the use of a single animal model, which may not fully capture the diversity of gastric ulceration mechanisms in humans. The study also did not explore the compound's pharmacokinetics or long-term safety profile, which are critical for clinical translation. Additionally, while the findings are promising, further validation is needed to confirm the reproducibility of these effects across different NSAID-induced ulcer models.

## CONCLUSION

In summary, this study represented the first instance where the protective effects of 2-Chloro-3-hydrazinylquinoxaline against indomethacin-induced gastric ulcers in rats have been demonstrated. This observed protection is believed to be linked, at least partially, to the antioxidant and anti-inflammatory characteristics of the compound. Nonetheless, it is crucial to emphasize that further thorough investigations are necessary to thoroughly explore the underlying mechanisms and validate these initial results.

## SIGNIFICANCE STATEMENT

This study aimed to investigate the gastroprotective effects of 2-Chloro-3-hydrazinylquinoxaline against NSAID-induced gastric ulcers in a rat model. The study findings demonstrate that 2-Chloro-3-hydrazinylquinoxaline significantly reduced ulcer formation, restored normal epithelial tissue and decreased inflammatory biomarkers. These results highlight the compound's potential as a novel treatment for NSAID-induced gastric damage, expanding the understanding of gastroprotection mechanisms.

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