Possible Gastroprotective Impact of 3-hydrazinoquinoxaline-2-thiol on Gastric Ulcers in Rats

Hussam Daghistani, MD, PhD*,********** Yousef Almoghrabi, MD, PhD*,********** Taghreed Shamrani, MD, PhD*,********** Mohammed Alsieni, MD, PhD** Mohammed A Bazuhair, MD, PhD**,********** Mazen A. Ismail, MD*** Asim T. Sharif, MD*** Bayan Redwan, MD*** Samah Labban, MSc, PhD**** Turki M. Alharthi, MSc, PhD***** Deena Mohammed Bukhary, MSc, PhD****** Ohood S Alharbi, MD, PhD******* Malaz Gazzaz, MSc, PhD******** Motasim M Jawi, MD, PhD********* Waiel S. Halabi, MSc, PhD.********** Rawan Altalhi, MSc, PhD********** Mohanned Talal Alharbi, MD, PhD********** Hadeel Ahmed Alsufyani, MD, PhD********** Wafaa Alhazmi, MSc, PhD********** Khalil Alkuwaity, MSc, PhD**********,********** Jawahir A. Mokhtar, MD, PhD**********,**********,********** Mona Abdulrahman Alqarni, MD, PhD********** Noof R. Helmi, MSc, PhD********** Nabeel Hussain Alhussainy, MSc, PhD********** Abdulaziz Alsaedi, MD, PhD********** Bandar Hasan Saleh, MD, PhD**********,********** Abdelbagi Elfadi, MD, PhD ********** *********,********** Karem Ibrahem, MD, PhD**********,**********

*	Department of Clinical Biochemistry, Faculty of Medicine, King Abdulaziz University, Jeddah 21589 Saudi Arabia.
**	Clinical Pharmacology Department, Faculty of Medicine, King Abdulaziz University, Jeddah, 21589, Saudi Arabia.
***	Department of Medical Education, Faculty of Medicine, King Abdulaziz University, Jeddah 21589, Saudi Arabia.
****	Department of Physiology, Faculty of Medicine, Umm Al-Qura University, Makkah, Saudi Arabia 21955.
*****	Department of Clinical Laboratory Sciences, Faculty of Applied Medical Sciences, Umm Al-Qura University, Makkah, Saudi Arabia.
******	Department of Pharmaceutical Sciences, College of Pharmacy, Umm Al-Qura University, Makkah, Saudi Arabia.
*******	Department of Microbiology and Parasitology, Faculty of Medicine, Umm Al-Qura University, Makkah, Saudi Arabia.
********	Pharmaceutical Practices Department, College of Pharmacy, Umm Al-Qura University, Makkah, Saudi Arabia.
*********	Department of Physiology, College of Medicine, University of Jeddah, Jeddah 23890, Saudi Arabia.
**********	Department of Optometry, Faculty of Applied Medical Sciences, University of Jeddah, 23218, Jeddah, Saudi Arabia.
**********	Department of Biological Sciences , College of Science, University of Jeddah, Jeddah 23445, Saudi Arabia.
**********	Department of Basic Medical Sciences, Collage of Medicine, University of Jeddah, Jeddah, Saudi Arabia.
**********	Department of Clinical Physiology Faculty of Medicine, King Abdulaziz University, Jeddah, Saudi Arabia.
**********	Department of Medical Laboratory Sciences, Faculty of Applied Medical Sciences, King Abdulaziz University, Jeddah 21589, Saudi Arabia.
**********	Department of Clinical Microbiology and Immunology, Faculty of Medicine, King Abdulaziz University, Jeddah 21589, Saudi Arabia.
**********	Regenerative Medicine Unit, King Fahd Medical Research Center, King Abdulaziz University, Jeddah 21589, Saudi Arabia.
**********	Food, Nutrition and Lifestyle Unit, King Fahd Medical Research Centre, King Abdulaziz University, Jeddah 21551, Saudi Arabia.
**********	Centre of Research Excellence for Drug Research and Pharmaceutical Industries, King Abdulaziz University, Jeddah, Saudi Arabia
**********	Vaccines and Immunotherapy Unit, King Fahd Medical Research Center, King Abdulaziz University, Jeddah, 21589, Saudi Arabia.
**********	Department of Clinical Microbiology Laboratory, King Abdulaziz University hospital, Jeddah 21589, Saudi Arabia. E-mail: kaibrahem@kau.edu.sa

ABSTRACT

NSAIDs are widely utilized worldwide, serving as a common pharmacological option for pain and inflammation relief. Despite their extensive use, NSAIDs come with notable adverse effects, with gastric damage being a prominent concern. This side effect is recognized as the primary issue linked to NSAID usage. Consequently, NSAIDs significantly contribute to the prevalent problem of stomach ulceration, ranking as the fourth leading cause of morbidity in the medical field. In our study, we employed experimentalmodel of indomethacin (IDMN)-induced gastric ulcer in evaluating the influence of 3-hydrazinoquinoxaline-2-thiol (3HQE) on gastroprotection inrat model. Thirty male Wistar rats, with weights ranging from 200 to 230 grams, were randomly allocated to groups into 5 group (n = six) as follows: Group1,control, Group 2,IDMN only, IDMN 30mg/kg, Group3 (IDMN and 3HQE 30mg/kg), Group4 (IDMN with 3HQE 60mg/kg), and Group5 (IDMN with esomeprazole (EZE)30mg/kg). The activity of 3HQE in gastric ulcers (GU) due to the administration of IDMN inrats was assessed according to gastric morphology, and inflammatory biomarkers. IDMN-induced GU led to epithelialdamage andblood streaks on the gastric-mucosa. However, treatment with IDMN+ 3HQE (60mg/kg) drastically reduced the ulcer in comparison with the IDMN only group. Inflammatorycells were detected in the IDMNgroup, while the 60mg/kg 3HQE-treated group displayed arestoration of thenormal epithelialtissue and minimise theinflammatory cells, that is almostsimilar to thecontrol and esomeprazole-treated groups. In addition, At dosages of both 30 and 60mg/kg, 3HQE exhibited significant anti-inflammatory properties, resulting in a marked reduction in inflammatory markers such as TNF-α,IL-6,IL-1β,IFN-gamma, and INOs, in comparison with the group treated solely with IDMN. This decrease was statisticallysignificant, withp-values less than 0.0001 for TNF-α and IL-6, and less than 0.0002 for IL-1β, IFN-gamma, and INOs. Furthermore, prior administration of 3-hydrazinoquinoxaline-2-thiol at both 30 and 60 mg/kg doses demonstrated notable increases in the concentrations of protective gastric factors, such as PGE2 and mucin, compared to the group exposed only to indomethacin. These increases were found to be statisticallysignificant, with p-values less than 0.0001 for PGE2 and less than 0.0003 for mucin. This study is noteworthy by Provide strong clinical evidence highlighting its remarkable gastroprotective properties of 3HQE. The inaugural application of this drug has revealed, for the-first-time, its activity in gastroprotection for treating GU due to IDMN. However, additional assays are required to assess these outcomes further.

Keywords: GU, NSAIDS, IDMN, inflammatory biomarkers, 3HQE.

INTRODUCTION

Inflammation is an adaptive physiological reaction activated by diverse factors such as infections, exposure to detrimental chemicals, and immune responses. It is activated in the presence of tissue damage, and this response is evident via observable-symptoms like swelling,redness,warmth, and pain, which may ultimately result in a reduction in the functionality of the tissue[1,2]. The process of the inflammation includes the widening of blood-vessels and increased activity of WBCs, along with releasing of inflammatory-mediators[3]. Persistent immune system activation and ongoing contribution in releasing inflammatory mediators significantly to the development of a variety severe medical conditions in chronic inflammation[4]. NSAIDs are extensively employed on a global scale, constituting a ubiquitous pharmacological choice for alleviating pain and inflammation. However, amidst their widespread use, the adverse effects of NSAIDs, particularly gastric damage emerge. This side effect is acknowledged as the foremost concern associated with NSAID usage[5]. NSAIDs are a significant contributor to the prevalent issue of stomach ulceration, standing as the fourth leading cause of morbidity in the medical domain[5,6]. Studies conducted in SA have revealed the preevalence of pepptic ulcers among the surveyed individuals. The findings indicate that 21.9% of respondents had peptic ulcers, with 16.2% presenting GU and 5.6% displaying duodenale ulcers. This prevalence is linked to the lengthened use of NSAIDs in 33.3% of cases and *H.pylori*-pathogen among24.2% of cases[7].

Gastric ulcers result from non-malignant lesions on the mucosal epithelium due to abundant gastric acid and heightened pepsin aggressiveness. Gastric ulcers, a widespread gastrointestinal disease, pose life-threatening risks with a 10% global prevalence. Control and early detection are significant challenges, leading to approximately 15 deaths per 15,000 cases annually worldwide[8]. Imbalances in factors affecting the mucous membrane's epithelium are common causes of gastric ulcers. As previously noted, the occurrence of peptic ulcers results from mucosal damage when there is a disturbance in the equilibrium between potent elements (*H. pylori* infection, acid, alcohol, NSAIDs, bile salts, and pepsin) and protective mechanisms (prostaglandins, mucosal blood flow, mucous, bicarbonate, and epithelial renewal)[9].

Inflammation can trigger the liberation of inflammatory agents, likeinterleukin-1β (IL-1β),interleukin-6 (IL-6),C-reactive protein (CRP),and tumor necrosis factor-alpha(TNF-α)[10]. Significantly, these mediatores are present in both acuteandchronic inflammmatory-cases. It is worth mentioning that previous research on indomethacin (belongs to the NSAIDs family)-induced ulcers has revealed its inhibition of prostaglandin-E2 (PGE2) production and angiogenesis. Additionally, it stimulates free radical generation, and prompts the enabling of cytokines associated with pro-inflammatory-processes[11-13].

Currently, medications to address inflammation/pain include NSAIDs and antinociceptive agents. However, these medicines often carry various adverse and toxic side effects[14]. IDMN in particular, is recognized for its elevated likelihood of causing ulcers in comparison to other NSAIDs, rendering it the favored option for experimentally inducing gastric ulcers[11]. IDMN is preferred as the primary choice for establishing an experimental-ulceer-moddel becauseof its heightened ulcerogenic possibility compared to other NSAIDs. Its mechanism involves the inhibition of COX-1 enzymes, thereby suppressing prostaglandin synthesis. Literature reports indicate that IDMN inducces gasstric injurey by hindering the production of prostaglandin E-2(PGE-2), bicarbonate, and mucus derived from COX-1 enzymes. Additionally, it promotes gastric acid secretion, increases oxidant parameters, and decreases antioxidant parameters[11,16,17].

Various non-natural antiulcer medications including cimetidine,misop rostol,ranitidine and omeprazole are currently used to treat NSAID-causing GU. However, it is crucial to acknowledge that these drugs comes with a range of side-effects, varying from light to heavy[18,19]. This has prompted the exploration of different anti-ulcer agents that are safe, easily available, and not expensive.

Quinoxaline compounds exhibit a broad spectrum of uses, showcasing Wide range of biological characteristics with notable implications in cancer-therapy. Moreover, they are crucial in crafting antimicrobial agents tailored to target bacteria, fungi, and viruses. In addition, it shows good activity as an anti-inflammatory agent[20]. The *Chromolaena odorata* plant has been documented to naturally contain 2,3-dimethylquinoxaline (DMQ)[21]. The quinoxaline structural framework facilitates the feasibility of these activities. Serving as a precursor, the quinoxaline structure enables the synthesis of many new compounds with varied applications[20]. By assessing the biological properties of various quinoxalines *in vitro*, we have ventured into understanding their potential[22]. Extending this understanding, we posit that 3-hydrazinoquinoxaline-2-thiol exhibits gastroprotective effects against gastric ulcers induced by indomethacin. Our aim is to thoroughly explore the potential gastroprotective properties of 3-hydrazinoquinoxaline-2-thiol versus IDMN-caused GU in rats, utilizing an animal experimental-model specifically designed for studying gastric ulcers. We assessed the effectiveness of 3-hydrazinoquinoxaline-2-thiol in alleviating GU caused via IDMN administration among rats by evaluating gastric morphology, and inflammatory biomarkers.

MATERIALS AND METHODS

Agents and chemicals

3HQE, IDMN, EZE, and carboxymeethyl-celluloose-sodum(CMC-Na),were sourced fromSigma-Aldrich,USA. Additionally, different ELISA kits were employed, such as the RatTNF-αELISA-Kit, Rat-InterferonAlpha-kit (Cat-No.MBS267050), Rat-Prostaglandin E2(PGE2)-ELISA Kit (Cat-No.MBS262150), Rat-MucinELISA-(Cat-NoMBS1600651), Rat-Interleukin-6(IL-6) ELISA-Kit-(Cat-No. MBS269892),Rat-Inducible-Nitric-Oxide-Synthase-(INOs) Elisa-kit-(Catalog-Number:MBS723326), and RatIL-β1ELISA Kit-(Catalog #MBS825017), all provided from Sigma-Aldrich in USA.

The stuudy alsoutilized commercially-available-chemicals such as formalin, phosphate bufffer, and othir essential agents.

Animal

The animal-related aspects of this study conformed to the approved protocols well-established by the Reseearch-Ethiics-Committee-of-the Facultey of Pharmmacy at King-Abdulaziz-University, under RN (PH-1444-56). Thirty malee-Wisstar-ratss, aged ten Ws and weighing between 200-230g, were obtained from the animal-facility at the Faculty of Pharmacy,King-Abdulaziz-University. Ratss were housed in a well-controllled-environmente, maintaining a temperaturee-rangge of 20-24°C and a 12-hour lighht and 12-hour darkk cyclee. Rats had freee-accesss to a standardd diet/water[13]. A one-week acclimation period was provided for the rats to adapt to the experimental facility conditions before initiating the experiments. The animals were induced into unconsciousness via intramuscular-injections of 2mg/ml chlorpromazine chloride obtained fromsigma, with 50µl injected into each femur.

30-rats were distributed at random into five groups, each consisting of 6-rats, and received the following treatments:

Control-Group-1: thisgroup received orally 0.5%w/v carboxymethyle cellulosse sodium, 10mL/k.

IDMN-Group-2: this group obtaineed a singlee-orael dose of IDMN (30mg/kg).

Group-3: IDMN +3HQE 30mg/kg: Rats in this group were given 3-hydrazinoquinoxaline-2-thiolorally atdose of 30mg/kg for3 days in a row. On 3rdday, they received IDMN of 30mg/kg orally, folllowed by the lasst dose of 3HQE one hourlater.

Group-4: IDMN +3HQE 60mg/kg Group: identical to Group-3, they received 3HQE atadose of 60mg/kg.

Group-5: IDMN +EZEGroup: They orallyadministered EZE (30mg/kg) forthree dayys in a row. 3rd day, they had IDMN (30mg/kg) orally, after the last dose of EZE one hou late.

Following four hours, IDMN-administration, they euthanized for more investigations[23]. Injectable anaesthesia was used for sedation including ketamine, and xylazine. Euthanasia involved the intraperitoneal administration of overdosse of ketaminee/xylazinee, it is followeed by a dislocatiion of the cervix. In this study, animals were monitored using criteria such as health status, behavior, pain, distress, and treatment response, alongside physiological parameters like body weight, temperature, and vital signs. Decisions on euthanasia adhered to institutional ethical guidelines and protocols, ensuring humane treatment and compliance with regulatory standards set by the Reseearch Ethiics Committee (Faculty of Pharmacey, KAU, Jedddah,SA,Refereence No. PH-1444-56).

Induction of Gastric Ulcers: Consistent with previous research, it was noted that theadministration of IDMN led to the formation of GU[24]. On the 2ndday of the experimment, the rats subjected t0 a 24 hours fasting periood with access to water. This fasting period helps ensure uniform baseline conditions among the experimental groups, minimizing variability in drug absorption and metabolism[25]. Subsequently, on the third day, all groups (excluding the control group) received an intragastric administration of indomethacin at 30 mg/kg, dissolved in a solution containing 0.5% carboxymethyl cellulose sodium (CMC-Na). Indomethacin stands out as the preferred option for constructing an experimental ulcer model due to its potent ulcerogenic properties in comparison to other NSAIDs. Its mechanism involves the suppression of prostaglandine-synthesiis through targeting both COX-1/COX-2 enzymes. The anti-inflammatory impact of IDMN is linked to the supression of the COX-2 enzyme, whereas its gastrointestinal side effects arise from COX-1 enzyme inhibition. Studies in the literature reveal that IDMN induces gastric damage by impeding the production of PGE-2, bicarbonate, and mucus derived from the COX-1 enzyme. Moreover, it stimulates gastric acid secretion, heightens oxidant parameters, and reduces antioxidant parameters[11,16,17,26].

Inflammatory biomarker assessment: Gastric tissue homogenates were analyzed for PGE2 activity utilising rat- PGE2-ELISA Kit(Cat No.# MBS262150,USA). Additionally, IL-6,TNF-α,IFN-γ, and RatIL-β1 in the supernatant were assessed using respective ELISA Kits (Cat No.# MBS269892, MBS2507393, MBS267050, MBS825017). Themucin-protein was measured using the Rat-MUCc1-ELISA kiit(Cat. No.# MBS1600651, St. Louis, MO, USA). All assays followed the manufacturers' protocols and utilized kits from Sigma-Aldrich,USA.

Statistical analysis: The datta provided in this research are represented as the meen standaird deviation (SD). To conduct multiple comparisons, aone-way-ANOVA was subjecteed, after Tukkey's-posst-hoc-test for in-depth analysis. Statistical-significancee was set at a probabiliity value (P) less than0.05. All statisticcal-analysess were executed using GraphPaad Instat softwar V8, and the graphics were produced via GraphPad Prism software V8 (GraphPad Software,USA).

RESULTS

Influence of 3HQE on Stomach Morphology

The effectiveness of 3HQE in mitigating gastric damage induced by indomethacin-triggered peptic ulcers was assessed using stomach-mucosa of a rat model. Figure 1A illustrates the healthy state of the stomaachmucous-membrane among the controll-groupp, indicating no damage. In contrast, the indomethacin group exhibited bloody streaking wounds (Figure 1B). However, the IDMN+3HQE (30mg/kg)group displayed a diminished appearance of bleeding smears (Figure 1C). Following treatment with IDMN+3HQE (60mg/kg), rats displayed a diminish inulcer compareed to the indomethacin group, along with evidence of mild injuries (Figure 1D). As anticipated, esomeprazole treatment resulted in a notablle decrease in ulccer formation in comparisson with the grooup had only IDMN, effectively protecting thee layere of the gastriic mucoosa (Figure 1 E). This suggests that 3HQE is as effective as esomeprazole.

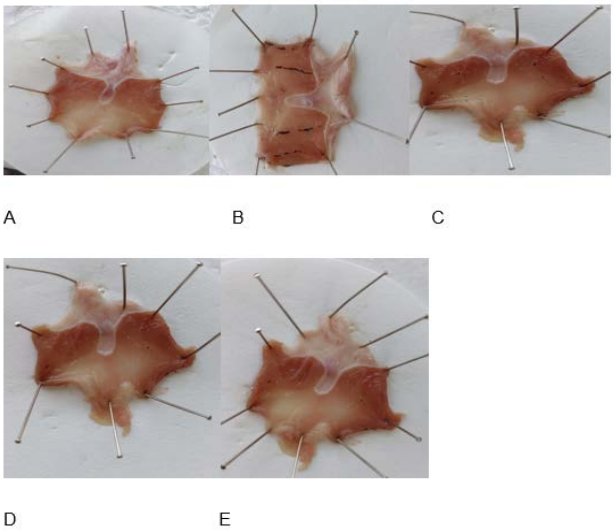

Figure 1. Photographs of the macroscopic appearance of rat stomachs. (1A): The stomacch-mucosa among the control-group displayed no lesions/rednesss. (1B) Rats treated with indomethacin displayed severe bleeding and mucous surface ulceration. Surface injuries were observed in (1C) indomethacin + 3-hydrazinoquinoxaline-2-thiol (30 mg/kg). (1D) indomethacin + 3-hydrazinoquinoxaline-2-thiol (60 mg/kg) showed minimal harm with a healthy mucosa. (1E) The injured mucosal-layer was retained to normal with indomethacin + esomeprazole(30mg/kg), displaying no visible redness or damage.

Impact of pre-treatment with 3HQE on inflammatory markers

In the rat-model ulcer, we evaluated the efficacy of 3HQE utilising molecular markers, including TNF-α,IL-6,IL-1β, IFN-gamma, andINOs. As depicted in Figure 2, the administration of indomethacin triggered a significant pro-inflammatory response, as evidenced by a marked increase in TNF-α,IL-6,IL-1β,IFN-gamma, and INOs concentrations in gastric tissues. This contrasted with the control-group that have not obtained IDMN. Conversely, pre-treatment with esomeprazole (30 mg/kg) or 3-hydrazinoquinoxaline-2-thiol atdoses (30 and 60mg/kg) demonstrated an anti-inflammatory effect, leading to a substantial reduction inTNF-α($p < 0.0001$),IL-6 ($p < 0.0001$),IL-1β($p < 0.0002$),IFN-gamma ($p < 0.0003$), andINOs ($p < 0.0003$), concentrations in comparison with the IDMN-treated-group. These results indicate that 3HQE exhibits effectiveness similar to esomeprazole in modulating molecular mediators in inflammatory processes (Figure 2).

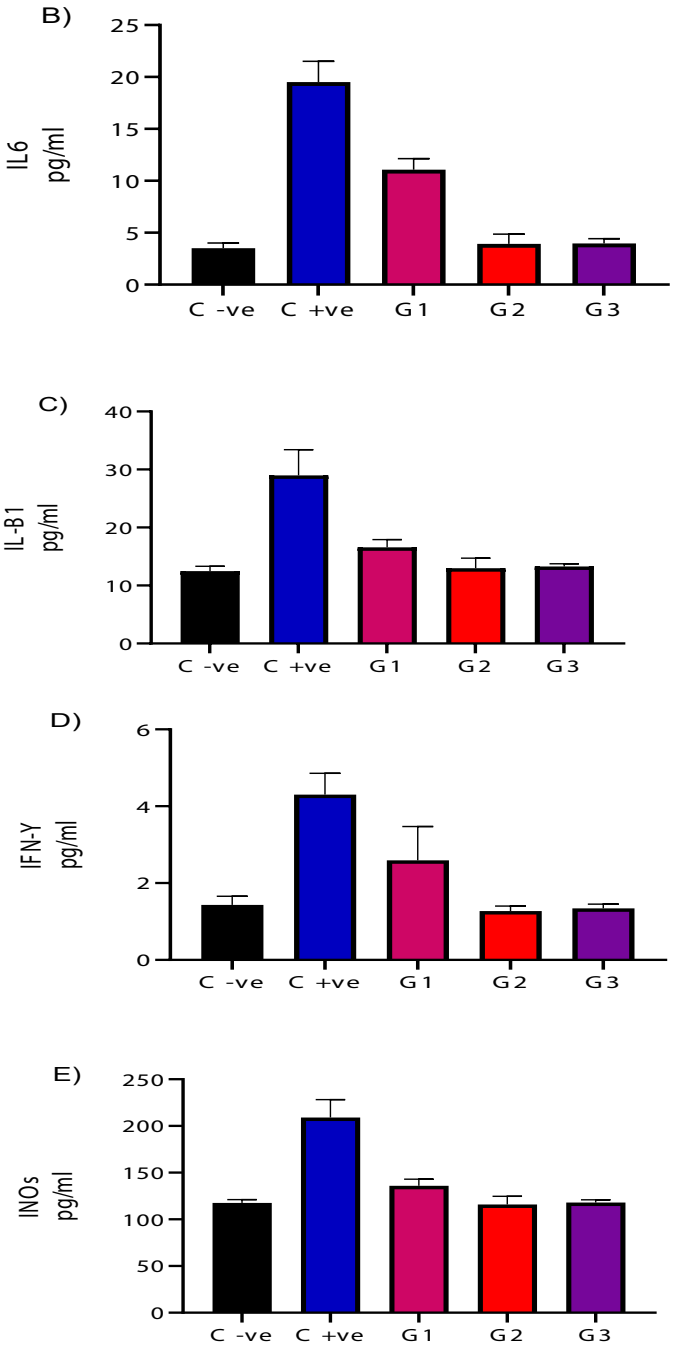

Figure 2. illustrates the impact of 3-hydrazinoquinoxaline-2-thiol pretreatment on A) TNF-α,B) IL-6,C) IL-1β,D) IFN-gamma,E) INOs concentrations in ratss with IDMN-inducced GU. The data, expressed asmean ± S.D. (n = 6), were considered statisstically-siignificant from the corresponding control and IDMN-groups at $P<0.05$, determined through one-way-analysis ofvariance (ANOVA) folllowed by Tukeey's post-hoc test.

Effects of pre-treatment with 3HQE on PGE2 and mucin concentrations

As shown in Figure 3, our findings depicted a reduction in mucin and prostaglandin levels after the adminisstration of indomethacin compareed tothe negative-control (untreated-group). Conversely, the group pre-treated with esomeprazole showed a substanntial($p < 0.0001$) increaise in PGE2 and mucin concentrations in contrast to the group subjected to indomethacin. Additionally, pre-treatment with 3-hydrazinoquinoxaline-2-thiol

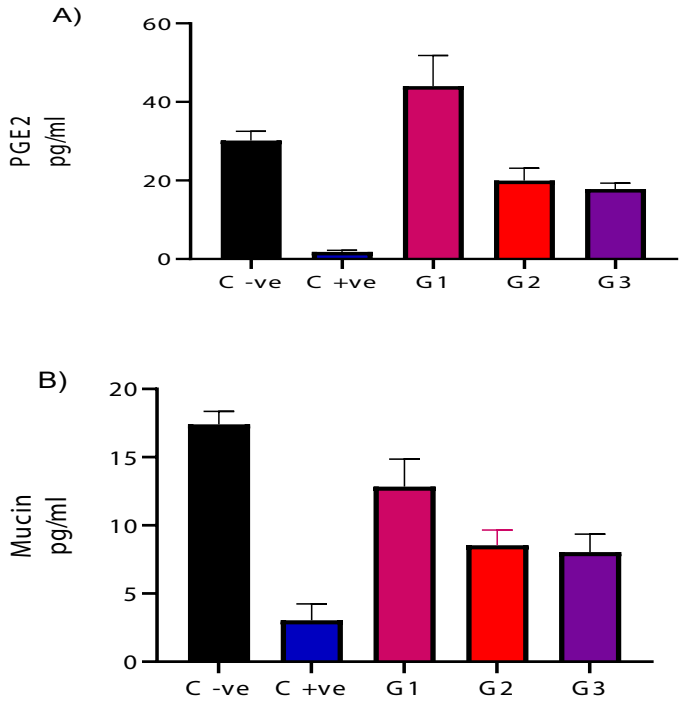

Figure 3. The influence of 3-hydrazinoquinoxaline-2-thiol pretreatment on A) PGE2 B) Mucin concentrations among rats with indomethacin-induced GU is depicted. The data, presennted as meaan ± S.D. (n = 6), demonstrated statistical signiificance from the corresponding control and IDMN groups at P<0.05. This signiificance was determiined through onne-way-analysis of variancce (ANOVA) followed byTukey's post-hocc-test.

at doses of 30 and 60 mg/kg demonstrated noteworthy increases in PGE2 ($p < 0.0001$) and mucin ($p < 0.0003$) concentrations in a manner dependent on the dosage when compared to the group exposed to indomethacin -exposed group. This suggests that 3-hydrazinoquinoxaline-2-thiol exhibited efficacy similar to esomeprazole in enhancing the concentration of both PGE2 and mucin.

DISCUSSION

GU is considered-benign mucosal lesion caused by excess stomach acid exposure, representing the most widespread gastrointestinal disorder. The gastrointestinal toxicity of NSAIDs, particularly the risk of complications, can be as high as 8% annually. This risk increases further for individuals with additional factors like a history of ulcer disease[27]. It has been proposed that the mechanism by which IDMN causes gastric damage involves the inhibition of the unleashing of defensive facttors like COX-1,PGE2,bicarbonaate, andmucus. Simultaneously, it leads to an elevation in corrosive elements such as acid and an increase in oxidant parameters, coupled with a reduction in antioxidant-parameters[11]. It isalso suspected that NSAID-meediated gastropatthy couldbe promoted by a reeduction inmucin level in the gastric mucosaa. Sustaining mucus formation may offer partial yet significant protection against reactive oxygen metabolites, preventing damage[5,28]. Our findings indicated a decrease in gastric mucus associated with stomach ulcers. Consequently, the mucosal membrane may be less equipped to safeguard against damage and hydrogen ion-back-diffusion, potentially hindering epithelial-healing. Comprehending these processes is vital for creating new drugs. Given the drawbacks of synthetic medications, exploring natural plant-based products, known for safety, efficacy, and affordability, is worthwhile for treating gastric ulcers[27]. Hence, this inspires us to discover a new drug with minimal side effects and affordable cost.

In this investigation, administering indomethacin orally to rats caused heightened gastric acidity/ulceration in comparison with control-group. These observations are consistent with earlier report highlighting indomethacin's tendency to raise gastric acidity, contributing to ulcer development[29]. For the first-time, this study thoroughly evaluated the impact of 3HQE on arat-model with inducced GU. Ourresults indicate that 3HQE effectively treated ulcer symptoms in rat groups, comparable to the effects of esomeprazole.

In the assessment of the efficacy of 3HQE in an ulcer treatment model, clinical biomarkers including INOs,TNF-α,IL-1,IL-6, mucin, and PGE2 were employed. Esomeprazole also demonstrated an anti-inflammatory effect, the reeduction ofTNF-α leveels in gastric tisssue, which is in line with a former study indicating that esomeprazole administration decreased serumTNF-α in an eethanol-causing ulccer-modeel[30]. These alterations corresponded to heightened levels of measured inflammatory markers. It has been documented that TNF-α significantly contributes to the formation of IDMN-induced GU, primarily by triggering neutrophil infilttration. This process is associated with decreased PGE2 levels and elevated TNF-α levels in the gastric-mucosa[11]. For the inaugural time in our study, it has been displayed that after the adminiistration of 3HQE atadose of 60mg/kg to thegroup receiving indomethacin, rats exhhibited a drastic decrrease in the ulccer comppared to the IDMN-only group. Furthermore, there were signs of mild injuries, implying that its efficacy rivals that of esomeprazole. This novel finding underscores the potential of 3HQE as an effective agent in mitigating gastric ulcers. Historically, quinoxaline derivative drugs have been acknowledged for their anti-inflammatory properties[31]. This precedent suggests that 3-hydrazinoquinoxaline-2-thiol may manifest a dual activity, as an anti-gastric ulcer and anti-inflammatory agent. However, it is imperative to note that further investigations are warranted to delve deeper into this potential dual functionality. This may involve optimizing the doses required to achieve the desired dual effect, ensuring a comprehensive understanding of its therapeutic capabilities. These results align with previous findings, where pre-treatment with tetramethylpyrazine similarly elevated muciin-leve,ls anddecreased TNF-α andIL-6-levels. Additionally,tetramethylpyrazine-treated groups exhibited improved histopathological changes incomparison with the IDMN-induced GU-group[23]. On theotherhand, in a different study, piceatannol reduced IL-6 andTNF-α levels, while enhancing mucin and PGE2 content[13].

The study explores how gastric ulcers develop due to mucosal injury caused by inflammatory responses triggered by ethanol ingestion. Various keycytokines, includingTNF-α,IL-6,IL-10,PGE2,andIL-1β, secreted via macrophages while inflammation and gastric ulcer. The research assesses these cytokines in etthanol-caused rat models and examines the effectiveness of fustin, with the 100 mg/kg dose showing significance across all parameters[32]. It aligns with the previous study, our findings demonstrated alterations in inflammatory biomarkers after indomethacin treatment. Notably, the 60 mg/kg dose of 3HQE emerged as an effective agent in ameliorating all inflammatory mediators associated with gastric ulcers, as evidenced by the aforementioned parameters. Treatment with EZE elevated gastriic-PGE2levels, consistent with the study documenting EZE capability toenhance PGE2-levels amongrats with IDMN -caused ulcers[25]. Esomeprazole's anti-ulcerative efficacy via the alpha-2 adrenergic-receptor, closely linked to gastroprotective COX-1 andPGE2[33] was also observed. Furthermore, esomeprazole changed COX-2and PGE2, helping to heal ulcers by re-epithelialization[34].

The assessment of gastroprotective mediator PGE2 and mucin levels indicated a substantial decrease in the group administered indomethacin. Conversely, rats treated with either esomeprazole or 3HQE significantly improved gastric PGE2 and mucin levels. The diminished synthesis of PGE2 is implicated in NSAID-induced gastric ulcers, given its role in

safeguarding the gastriic-mucosa through enhanced mucus-secretiion, maintenance ofblood-flow, and reduction in acidic-secretioon[12,35]. Moreover, a recent rat study indicated a substantial increase in gastriic-muciin-content, mitigated neutrophiil-infiltratiion (asevidenced by decreased myyeloperoxidase-activiity), and a reduced increased serumnitric-oxide levels[35]. In summation, the dynamic involvement of pivotal inflammatory markers in the gastric ulceration process is discerned through the escalation of inflammatory markers and the concurrent diminution of protective molecules such as mucin and Prostaglandin E2 (PGE2) after the induction of ulcers by indomethacin. Our investigative study elucidates that 3-hydrazinoquinoxaline-2-thiol orchestrates the restoration of inflammatory biomarkers to baseline levels while concurrently reinstating protective molecules, notably PGE2, and mucin, in a manner akin to the observed effects of esomeprazole.

Inflammation in the gastrointestinal tract can lead to persistent tissue damage over time. Changes inthelevels ofproinflammatorycytokines likeTNF-α,IFN-γ,IL-1,andIL-6 are believed in playiing a cruucial functiion in regulating this inflammmatory-response. Some NSAIDs, including COX-2 inhibitors and ibuprofen, are known to pose gastrointestinal risks, albeit to varying extents. Blocking COX-1 reduces the secretion ofprotectiveprostaglandins among gastric mucosa, increasing the risk of mucosal injury. Additionally, inhibiting COX-2 may also contribute to mucosal damage. Inthisstudy, we demonstraated that pretreatment with 3HQEresulted in a decrease inTNF-αand IL-6, as well as IFN-γ and IL-1 levels. Furthermore, it led to an increase in mucin and PGE2 levels. These findings provide insight into how 3-hydrazinoquinoxaline-2-thiol may alleviate GU in a rat modeel.

Theprecise mechanism underlying the gastroprotective effects of 3-hydrazinoquinoxaline-2-thiol remains unclear. However, our investigative study sheds light on its potential mechanisms by demonstrating that 3-hydrazinoquinoxaline-2-thiol contributes to the normalization of inflammatory biomarkers while simultaneously enhancing the levels of protective molecules. Through our experimentation with different doses of 3HQE, we observed noticeable reductions in inflammatory biomarkers such as TNF-α,IL-6,INF-γ,and IL-β1 comparred tothe group administered only indomethacin. Additionally, there was a notable increase in gastroprotective mediator levels, including PGE2 and mucin, in comparison with the IDMN-only group. These outcomes indicate that the observed changes in inflammatory biomarkers and gastroprotective mediators may be attributed to the activity of3HQE. Nevertheless, further investigations are warranted to comprehensively eluciidate themechanisms underlyying the therapeutic activity of 3HQE in the treatment of peptic ulcers.

Studieshave showedthat prior adminisstration of esomeprazole leads to anincrease in PGE2 levels and inhibits the release of gastric-acid, pepsin, and gastrin. Furthermore, esomeprazole displays anti-oxidant properties by decreasing malondialdehyde-levels, boosting theexpression ofanti-oxidant agents such glutathione andsuperoxide dismutaase, andreducing thecompensatory-transcriptionalelevation ofSOD1-gene.

Additionally, EZE reduces levels ofmyeloperoxidase,TNF-α, and IL-1β, thus exhibiting anti-inflammatorey efficacy. Moreover, it has been observed that EZE diminishes the elevated phossphorylation-levels of nuclearfactor-kappa B (NF-κB) p65 andp38 MAPK and inhibits the nuclear translocation of NF-κB p65[25]. As a result, we postulate that 3-hydrazinoquinoxaline-2-thiol exerts a gastroprotective function against ulcers by modulating inflammatory biomarkers.

In the future, it is imperative to thoroughly investigate the safety profile of 3-hydrazinoquinoxaline-2-thiol to assess its viability for clinical application. This investigation should encompass an evaluation of potential toxicity and adverse effects associated with the compound. By conducting comprehensive safety studies, we can gain valuable insights into the compound's potential risks and benefits, thereby informing its suitability for use in clinical settings. This proactive approach to assessing safety profiles is essential for ensuring patient well-being and advancing the development of safe and effective therapeutic interventions.

CONCLUSION

In our study, we have extensively investigated and documented the activity of 3HQE in alleviating gastric damage induced by indomethacin in our rat model. Through meticulous experimentation and analysis, we have meticulously examined the effect of 3HQE on different aspects of gastric health and pathology in response to IDMN adminisstration. Our findingsprovide robust evvidence supporting the effectiveness of 3HQE as a potential therapeutic-agent for mitigating gastric damage in experimental models. The novel application of this drug has demonstrated its efficacy in diminishing proinflammatory markers, includingTNF-α,IL-6,IL-1β,IFN-γ, andINOs. Conversely, it exhibits an elevating effect on the levels of PGE2 and mucin, showcasing its potential as non-expensive phytochemical alternate for the therapy of chemically induced GU. This discovery prompts the need for further investigations to delve into the gastroprotective effects of 3-hydrazinoquinoxaline-2-thiol, not only in additional experimental models but also potentially in clinical applications.

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