Original Article

Effect of Atorvastatin on Indomethacin-Induced Gastric Ulceration in Rats: Role of Nitric Oxide and Gastric Motility

Dilgash A. Ali, Hani M. Almukhtar[1]

Health Sinjar General Hospital, Mosul, Iraq, [1]College of Pharmacy, University of Mosul, Mosul, Iraq

Abstract

Background: Non-steroidal anti-inflammatory drugs (NSAIDs)-induced gastric ulceration caused by inhibition of prostaglandins. Atorvastatin, a widely used hypolipidemic agent, has many beneficial pleiotropic effects like anti-inflammatory and antioxidant actions, it may also inhibit smooth muscle contraction. **Objectives:** The present study was to examine the gastric effect of atorvastatin in rats pretreated with indomethacin. **Materials and Methods:** Forty-five male albino rats were divided into seven groups. Group 1 (control): solvent, 10% hydroxy propyl cyclodextrin/10% of tween 80 (5 days), last day saline. Group 2: solvent (5 days), last day indomethacin. Group 3: simvastatin 20 mg/kg (5 days), last day saline. Group 4: atorvastatin 20 mg/kg (5 days), last day saline. Group 5: simvastatin 20 mg/kg (5 days), last day indomethacin. Group 6: atorvastatin 20 mg/kg (5 days), last day indomethacin. Group 7: atorvastatin 40 mg/kg (5 days), last day indomethacin. Serum nitric oxide (NO) was measured using a commercial kit. Serum malondialdehyde (MDA) was read by a spectrophotometer at 532 nm. Gastric muscle contractility was studied with an isolated tissue bath. **Results:** Indomethacin-induced gastric ulceration with increased MDA, decreased NO, and enhanced gastric motility. By contrast, atorvastatin dose-dependently reduced indomethacin ulceration enhanced NO, and decreased MDA level with inhibition of gastric motility. **Conclusion:** Atorvastatin inhibits indomethacin-induced gastric ulceration by enhancing NO level and secondary to inhibition of gastric motility.

Keywords: Atorvastatin, gastric motility, gastric ulceration, indomethacin, nitric oxide

Introduction

Gastric ulceration is considered to be the major side effect of NSAIDs through inhibition of prostaglandin synthesis.[1] Prostaglandin generation by the gastric layer aids protection against different ulcerogenic factors, they suppress acidity, stimulate mucous, and bicarbonate synthesis, in addition, prostaglandins enhance circulation in the mucosal layer and inhibit smooth muscle hypermotility.[2] Studies have shown that indomethacin could induce gastrointestinal tract (GIT) ulceration not only by inhibiting the protective mechanism but also by enhancing gastric motility.[3-5] Studies have suggested that indomethacin effect on motility is mediated peripherally by inhibition of PG synthesis, moreover it could centrally affect the cholinergic pathway, the latter is confirmed by the protective effects of atropine in experimental animals.[6] In addition to PG, another major protective substance in the gastric layer is nitric oxide (NO).[7] The presence of a Ca^{+2}-dependent NOS enzyme in the gastric layer suggests functioning of NO in the gastric epithelial cells, in fact, both PG and NO are both involved in the modulation of gastric acidity and maintenance of mucosal integrity. It is well known that NO exhibits protection of gastric mucosa by maintaining blood flow to the ulcer,[8] with additional benefit in wound repair due to angiogenesis and anti-inflammatory action.[9] Several experiments have shown that inhibition of NO synthesis by L-Nitro-arginine methyl ester (L-NAME) could exacerbate ulceration, in contrast, the administration of L-arginine, as NO precursor, has a protective effect against NSAIDs-induced ulcers.[10-12] Moreover, data have

Access this article online

Quick Response Code:

Website: https://journals.lww.com/mjby

DOI: 10.4103/MJBL.MJBL_1090_23

Address for correspondence: Dr. Hani M. Almukhtar, College of Pharmacy, University of Mosul, Mosul, Iraq. E-mail: hanialmukhtar@uomosul.edu.iq

Submission: 27-Jul-2023 **Accepted:** 05-Jan-2024 **Published:** 30-Apr-2026

How to cite this article: Ali DA, Almukhtar HM. Effect of atorvastatin on indomethacin-induced gastric ulceration in rats: Role of nitric oxide and gastric motility. Med J Babylon 2026;23:332-8.

shown that NO-releasing agents could also activate PG such as prostacyclin 2 and prostaglandin E (PGE2) via cyclooxygenase pathway.[13]

Atorvastatin is a commonly used hypolipidemic agent,[14] in addition to its therapeutic use for hypercholesterolemia, it has many beneficial pleiotropic effects like anti-inflammatory and antioxidant action.[15] It is also a smooth muscle relaxant.[16] It is well known that statins activate endothelial NO production.[17] The present study was designed to examine the gastroprotective effect of atorvastatin in rats pretreated with indomethacin.

Materials and Methods

Atorvastatin and simvastatin were a generous gift from Awa-Medica Pharmaceutical Company. Indomethacin used in this study is the commercially available induced capsule. Serum NO level was measured using commercial kits Elabscience. Krebs components were purchased from standard commercial supplies.

Animals

Forty-five albino male rats each aged 2.5–3 months weighing about 180–200 gm were used. All rats were housed under a controlled environment at a temperature of 22°C–25°C and humidity was maintained between 45% and 65%; they had free access to water and food except the day were received indomethacin the food was removed 24 h. Before administration to eliminate the confounding effect of food on indomethacin effect. All procedures and animal handling were performed in accordance with ARRIVE guidelines.[18] A preliminary study was performed to determine the minimum dose of indomethacin that causes apparent gastric lesions in rats after 6 h of indomethacin administration.

Experimental design and induction of gastric ulceration

The forty rats were divided into eight groups (five rats each) as follows:

Group A: orally administered 10% hydroxyl-propyl-cyclodextrin/10% of tween 80 (10 mL/kg) every day for 5 days. On the last day, they were administered 10% hydroxyl-propyl-cyclodextrin/10% of tween 80 (10 mL/kg), and after 1 h, they received saline (10 mL/kg). After 6 h, all the rats were sacrificed using decapitation method.

Group B: orally administered 10% hydroxyl-propyl-cyclodextrin/10% of tween 80 (10 mL/kg) every day for 5 days. On the last day they were administered 10% hydroxyl-propyl-cyclodextrin/10% of tween 80 (10 mL/kg), and after 1 h, they received indomethacin (8 mg/kg). After 6 h, all the rats were sacrificed using decapitation method.

Group C: orally administered simvastatin 20 mg/kg every day for 5 days. On the last day, they were administered simvastatin, and after 1 h, they received saline (10 mL/kg). After 6 h, all the rats were sacrificed using decapitation method.

Group D: orally administered simvastatin 20 mg/kg every day for 5 days. On the last day, they were administered simvastatin, and after 1 h, they received indomethacin (8 mg/kg). After 6 h, all the rats were sacrificed using decapitation method.

Group E: orally administered atorvastatin 20 mg/kg every day for 5 days. On the last day, they were administered atorvastatin 20 mg/kg, and after 1 h, they received saline (10 mL/kg). After 6 h, all the rats sacrificed using decapitation method.

Group F: orally administered atorvastatin 20 mg/kg every day for 5 days. On the last day, they were administered simvastatin, and after 1 h, they received indomethacin (8 mg/kg). After 6 h, all the rats were sacrificed using decapitation method.

Group G: orally administered atorvastatin 40 mg/kg every day for 5 days. On the last day, they were administered atorvastatin 40 mg/kg, and after 1 h, they received saline (10 mL/kg). After 6 h, all the rats were sacrificed using decapitation method.

Group H: orally administered atorvastatin 40 mg/kg every day for 5 days. On the last day, they were administered atorvastatin 40 mg/kg, and after 1 h, they received indomethacin (8 mg/kg). After 6 h, all the rats were sacrificed using decapitation method.

Ulcer index

The stomach was harvested directly after euthanizing rats. Then it was cut against the greater curvature; the mucosa was rinsed with normal saline to remove any contents or bleeding. The tissue was examined using TOMLOV digital microscope 10 digital microscope 50× UK to have a clearer vision for gross macroscopic scoring. The ulcer index was scored according to the report by Guth, *et al*.[19] as follows: (1) petechial lesion, (2) erosions <1 mm, (3) erosions between 1 and 2 mm, (4) erosions between n 2 and 4 mm, and (5) erosions greater than 4 mm. Then the scores summed to get the final score for each animal.

Histopathology

For histological assessment, gastric tissues were collected, cleaned with normal saline then placed into 10% paraformaldehyde solution in phosphate buffer saline overnight at 8°C, then gradually dehydrated by gradient ethanol and finally embedded in paraffin. The paraffin block was sectioned into 5-mm thick and stained with hematoxylin and eosin. A light microscope was used to determine the severity of gastric mucosal damage.

Serum NO

Blood samples were collected during sacrifices by beheading method. The blood let stand for approximately 30 min then centrifuged at 2000×*g* for 15 min. Serum NO level was measured using commercial kits Elabscience which is based on the Griess reaction assay.[20] The sample was measured using a spectrophotometer at 550 nm wavelength.

Serum MDA

Serum MDA was measured as a biomarker of lipid peroxidation as explained by Aguilar, *et al*.[21] This method depends on a thiobarbiturate reaction with MDA in an acidic medium to form a red-to-pink color thiobarbituric acid reactive substances. After boiling for an hour, cool it down to room temperature. Finally, the absorption was measured by a spectrophotometer at 532 nm wavelength.

Determination of gastric motility

Gastric muscle contractility was assessed by isolated tissue bath as previously described.[22] After stunning and decapitation. Fine dissection was carried out to get muscle segments. The stomach was cleaned and kept in Krebs solution composed of (mM): KCl 4.8, NaCl 118.5, $NaHCO_3$ 25, KH_2PO_4 1.2, $MgSO_4.7H_2O$ 1.2, $CaCl_2$ 1.9, glucose 10.1. The segments were mounted in a tissue bath at 37°C with continuous gas flow (O_2−95%/CO_2−5%). Each segment was connected by a thread to a transducer and precontracted to about 1 gm tension. The segments were then left for stabilization for about 30 min, the viability of the tissue was checked by adding KCl 70 mM for about 5 min for standardization, after washing with normal Krebs the tissue was left for about 30 min and then precontracted with cumulative addition of Ach to about 70% of KCl contraction, the acute response of atorvastatin was determined by the addition of atorvastatin 10 µM to one channel and dimethyl sulfoxide was added to the other as a control. Results were recorded with a special analyzing program (Labscribe, Oxford, UK).

Statistical analysis

Data were analyzed using a two-way analysis of variance (ANOVA) test and were presented as mean ± SE. When the ANOVA test turned significant *post hoc* test Bonferroni was performed. All experiments were analyzed statistically using GraphPad Prism 5 software (Boston, USA). $P < 0.5$ is considered statistically significant.

Ethical approval

All procedures and animal handling were performed in accordance with ARRIVE guidelines. The protocol of the study and animal handling procedure was reviewed and approved by the local ethics committee at the University of Mosul, Iraq, with reference number UM.VET.2022.067 on 2nd September, 2022.

Results

Histology

Indomethacin induced severe damage, disappearance of mucosa layer and part of submucosa [Figure 1]. Apparently, simvastatin 20 mg/kg and atorvastatin 40 mg/kg mitigate the ulcerogenic effect of indomethacin on stomach mucosa.

Ulcer index

The two-way ANOVA analysis of ulcer index data [Figures 2 and 3] showed a significant difference between rats treated with indomethacin and rats treated with saline ($P < 0.001$) and the effect of antihyperlipidemic drugs in general showed a significant effect ($P < 0.05$). Also, an interaction between antihyperlipidemic treatment group and indomethacin-induced ulcer interaction showed a significant effect ($P < 0.05$). *Post hoc* comparison showed a significant difference in ulcer index scoring between solvent and simvastatin 20 mg/kg ($P < 0.05$), and atorvastatin 40 mg/kg ($P < 0.01$) but not simvastatin 20 mg/kg ($P > 0.05$).

Serum malondialdehyde level

The two-way ANOVA analysis of serum MDA showed a significant difference between groups treated with indomethacin and groups received solvent only [F (1, 32) = 86.71, $P < 0.001$], the antihyperlipidemic drug treatment showed a significant difference in rats received solvent [F (3, 32) = 9.973; $P < 0.01$]. Finally, the interaction between both treatments, indomethacin and antihyperlipidemic drug, is significant [F (3, 32) = 7.562, $P < 0.01$]. The *post hoc* statistics showed a significant reduction in MDA in groups treated with simvastatin 20 mg/kg ($P < 0.001$), and atorvastatin 40 mg/kg ($P < 0.001$). Although, there was a trend that atorvastatin 20 mg/kg decreased MDA but it was insignificant ($P = 0.073$).

Serum Nitric oxide (NO) level

The two-way ANOVA analysis of serum NO level showed an insignificant difference between the groups treated with indomethacin and groups treated with solvent only [F (1, 32) = 0.0001661], while the antihyperlipidemic treatment showed a significant increase in NO level compared to rats received solvent only [F (3, 32) = 22.79; $P < 0.01$]. Finally, the interaction between both treatments (indomethacin and antihyperlipidemic drug) is insignificant [F (3, 32) = 0.4032]. The *post hoc* statistics showed a significant increase in serum NO level in rats treated with simvastatin 20 mg/kg [($P < 0.001$) and atorvastatin 40 mg/kg ($P < 0.001$)].

In vitro gastric motility

As shown in Figure 6A, atorvastatin elicited a time-dependent relaxation in rat stomach segments precontracted submaximally with pilocarpine. The effect reached a significant value after about 1 h incubation with

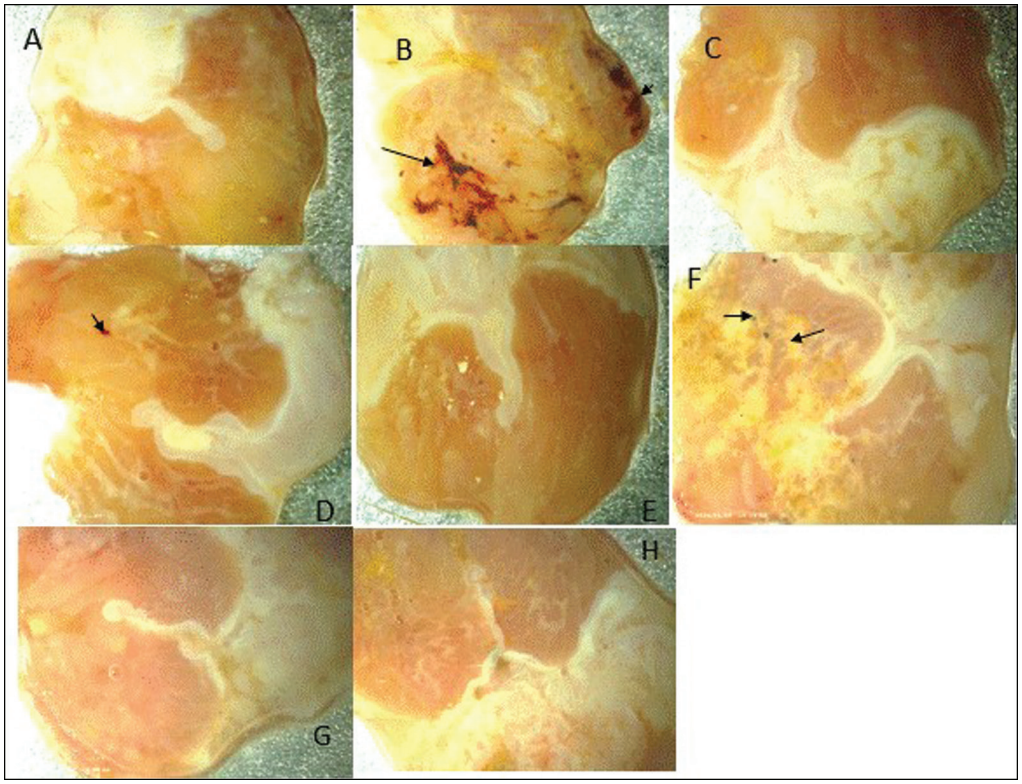

Figure 1: Macroscopic picture of the gastric lining: (A) rat stomach treated with saline and solvent only, (B) rat stomach treated with indomethacin (the black arrows show hemorrhagic lesion), (C) rat stomach pretreated with simvastatin 20 mg + saline, (D) rat stomach pretreated with simvastatin then indomethacin, (E) rat stomach pretreated with 20 mg/kg of atorvastatin then oral saline, (F) rat stomach pretreated with atorvastatin 20 mg + indomethacin (see black arrow), (G) rat stomach pretreated with atorvastatin then oral saline (H) rat stomach pretreated with atorvastatin 40 mg/kg then in the last day indomethacin (stomach mucosa looks intact). H and E stain, 100×

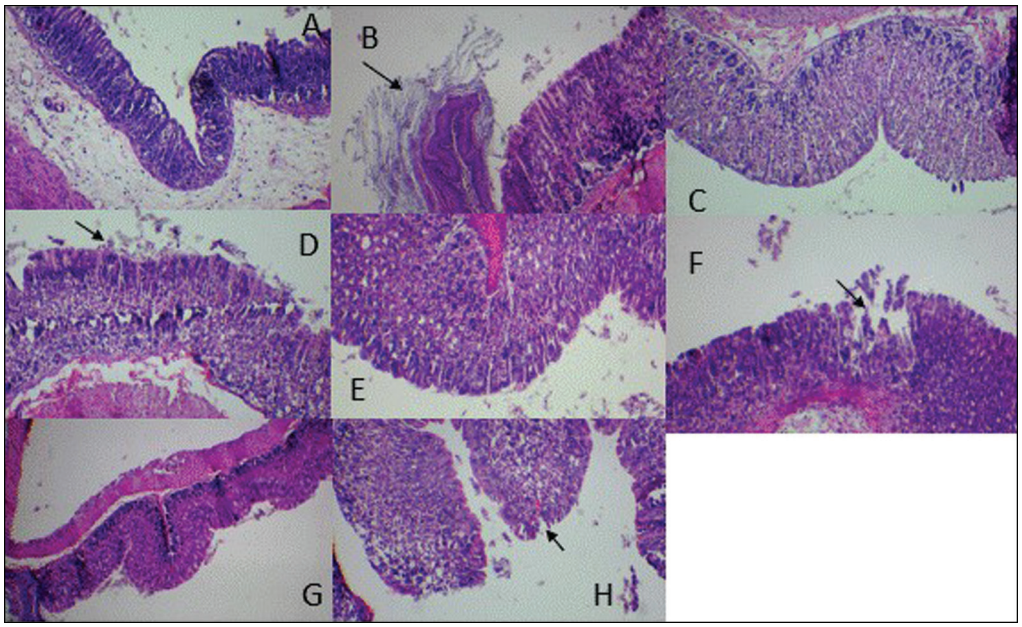

Figure 2: Stomach mucosa histology: (A) stomach mucosa is integral and intact in rats treated with solvent and saline only (B) showed a severe destruction of the entire mucosa layer and disruption of the normal gastric tissue morphology. (C) Intact stomach mucosa (not affected by simvastatin) (D) simvastatin pretreatment showed a gastroprotective effect against indomethacin-induce gastric ulceration (E) stomach mucosa have no alteration due to treatment with 20 mg/kg atorvastatin, (F) gastric mucosa damage is apparent due to indomethacin administration however, less severity when compared with rat gastric mucosa administered indomethacin only, (G) Intact stomach mucosa is intact (not affected by atorvastatin 40 mg/kg) (H) showed, rat stomach pretreated with atorvastatin 40 mg/kg then in the last day oral indomethacin (stomach mucosa looks intact). H and E stain, 100×

atorvastatin. In Figure 6B, indomethacin 1 μM incubation for about 20 min increased gastric smooth muscle tone, however, atorvastatin 10 μM inhibited gastric tone induced by indomethacin incubation.

Discussion

Despite the advances in pharmaceutical preparation, NSAIDs-induced gastric ulceration is still a common clinical complication.[23] Certain medical agents can be added as add-on therapy to reduce the incidence of ulceration induced by NSAIDs.[24-26] The current data clearly proved that the oral ingestion of indomethacin caused severe gastric ulceration accompanied by an enhancement in the level of MDA with a reduction of NO level [Figures 4 and 5]. In fact, the ulcerogenic side effects of NSAIDs and specifically indomethacin are secondary to their ability to produce reactive oxygen intermediates which could in turn facilitate lipid peroxidation.[27] Data have shown that statin administration could minimize the ulcerogenic effect of NSAIDs in lab animals,[28] the underlying mechanism suggested is possibly secondary to enhanced NO and PGE2 synthesis. The current work is designed to identify the potential benefit of atorvastatin on the antioxidant status and NO concentration as a potential mechanism of atorvastatin gastroprotective effect, the study also examined the effect of atorvastatin on gastric muscle contraction. The results clearly showed that oral administration of atorvastatin for 1-week duration significantly ameliorated gastric ulceration with a reduction in ulcer index. Atorvastatin increases NO concentration, reduces MDA level, and protectes gastric mucosa against the ulcerogenic effect of indomethacin, in addition, it significantly inhibits gastric motility [Figure 6]. The latter may act as an additional gastroprotective mechanism of atorvastatin in experimental animals. It is well known that NSAIDs nonselectively inhibit cyclooxygenase enzyme with subsequent reduction in PG biosynthesis, the latter involved in the regulation of gastric motility and mucous production.[29] In fact, the effect of atorvastatin was dose-dependent since atorvastatin 40 mg/kg had more protective effect than atorvastatin (20 mg/kg/day). Statin group was proved to have antioxidant and reactive oxygen species scavenging effects.[30] This may be

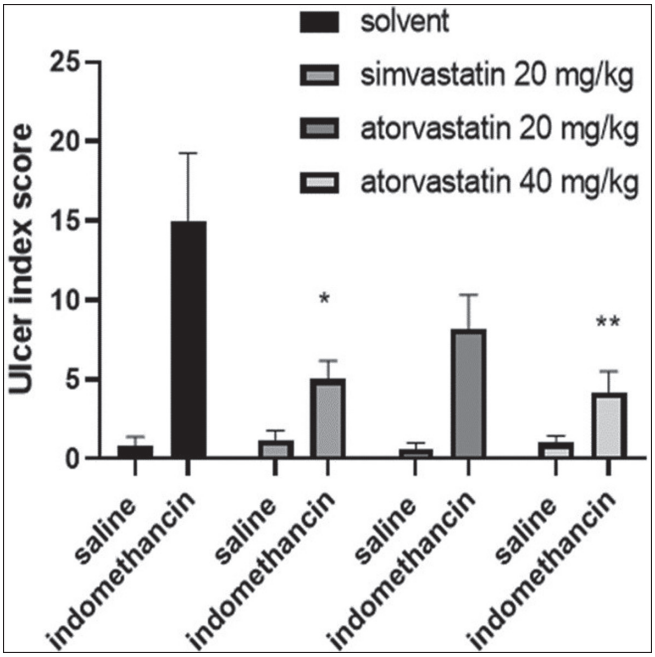

Figure 3: Ulcer index simvastatin 20 mg/kg and atorvastatin showed gastroprotective effect (*$P < 0.05$, **$P < 0.01$)

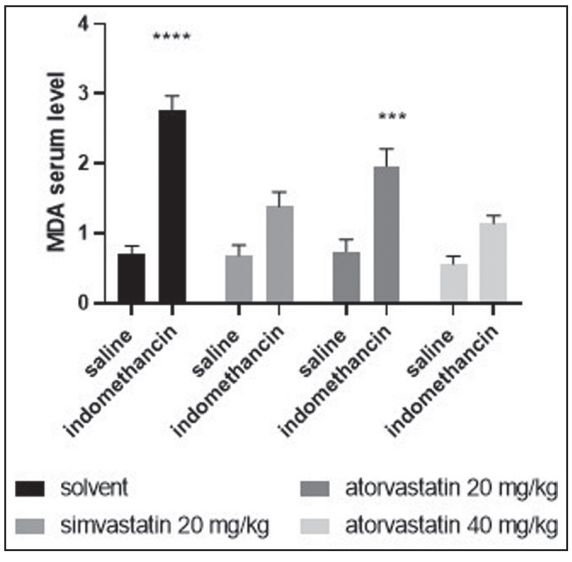

Figure 4: MDA serum level (μmol/L): indomethacin oral administration increases MDA serum level, and both simvastatin 20 mg/kg and atorvastatin 40 mg/kg attenuate indomethacin-induced elevation of MDA serum level. (****$P < 0.0001$, ***$P < 0.001$)

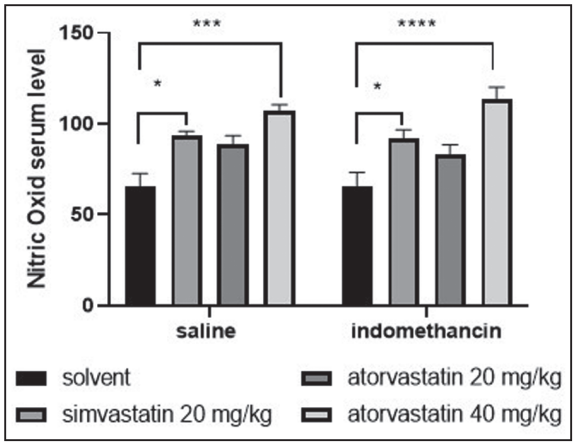

Figure 5: Serum NO level (μmol/L): indomethacin oral administration does not affect NO serum concentration, while both simvastatin 20 mg/kg and atorvastatin 40 mg/kg elevates NO serum level regardless of saline or indomethacin administration. (*$P < 0.05$, ***$P < 0.001$, ****$P < 0.0001$)

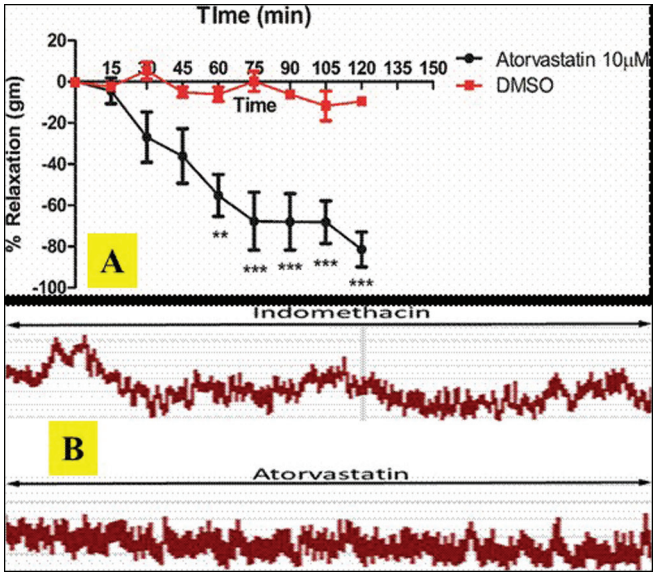

Figure 6 : (A) atorvastatin (10 μM) relaxant effect in rat stomach segments after pilocarpine contraction. Values are shown as relaxation percentages after pilocarpine-induced contraction and are mean ± SEM obtained from four different hearts. (B) Original organ bath trace for atorvastatin-induced rat stomach segments relaxation in the presence of indomethacin (1 μM)

one of the mechanisms by which statins protect gastric mucosa against indomethacin ulcerogenic effect.[28] The built-up of reactive oxygen species could remarkably alter the antioxidant mechanism causing oxidative damage with resultant stomach ulceration. Similarly, it has been shown that other statins have antioxidant effects, and they even enhance the activity of catalase enzyme.[31-34] Moreover, results have clearly confirmed the role of endogenous NO in maintaining gastric mucosal integrity.[35] NO regulates mucous secretion, hydrochloric acid secretion, PG synthesis and directly affects smooth muscle contractility. Interestingly, the NO released from nitrate preparation for example, nitroglycerin has gastroprotective effect against ulcers induced by indomethacin administration by enhancing blood flow to the affected area and reducing inflammation.[36] this study showed that treatment with atorvastatin significantly enhanced NO concentration in treated rats in comparison to control untreated rats, previous reports have similarly shown that statin treatment can significantly enhance NO level, and scavenge free radicals.[37] On the other hand, previous results have shown that the increased gastric motility due to indomethacin administration could increase the possibility of gastric ulceration, the underlying cause could be related to cyclooxygenase inhibition with resultant inhibition in PG synthesis. It is suggested that the increased gastric motility results in more mucosal folding and a reduction in blood supply.[4] In addition, the tight contraction of stomach muscle could block capillaries within the inner mucosa, with a resultant decrease in microcirculation.[3] Such effect can

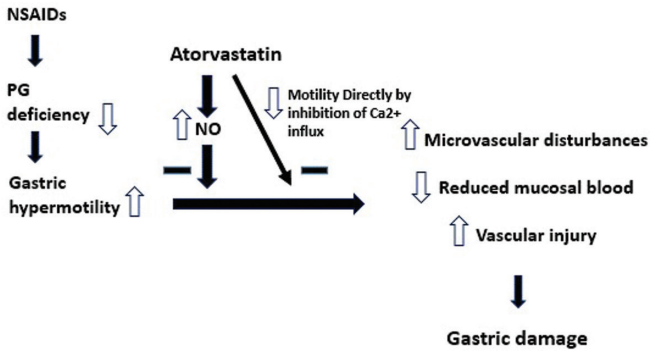

Figure 7: The proposed role of increasing gastric motility by NSAIDs in reduction of mucosal blood flow and the role of atorvastatin in reducing these functional changes with resultant prevention of gastric ulceration

be prevented by treatment with superoxide dismutase or the anticholinergic atropine. Thus, reactive oxygen species (ROS) generated by ischemia and reperfusion underlie the gastric lesion induced by indomethacin.[38] The undigested food could also physically irritate the gastric mucosa and thus more invasion by bacteria and other irritants.[39] The current results suggested that part of the gastroprotective effect of atorvastatin was mediated by inhibition of indomethacin-induced hypermotility. A previous study by Heeba, *et al*.[40] demonstrated that statins enhance PGE2 biosynthesis, the latter clearly plays a protective role in gastroprotection by stimulating mucous and bicarbonate secretion, it also maintains blood supply and enhances the resistance of the gastric epithelial cell to irritation. The activation of PG generation after treatment with atorvastatin could be induced, partly, by activation of NO synthesis.[41] Previous results have shown that statins with L-arginine administration enhanced the synthesis of prostaglandins in the stomach by contrast, NO synthase inhibition with L-NAME clearly inhibited such a mechanism [Figure 7].

In summary, atorvastatin gastroprotective mechanism involves both attenuation of aggressive factors like ROS and augmentation of defending mechanism like NO, the latter inhibits GIT motility which aids as a protective mechanism.

Financial support and sponsorship
Nil.

Conflicts of interest
There are no conflicts of interest.

References
1. Takeuchi K, Tanaka A, Kato S, Amagase K, Satoh H. Roles of COX inhibition in pathogenesis of NSAID-induced small intestinal damage. Clin Chim Acta 2010;411:459-66.
2. Halter F, Tarnawski AS, Schmassmann A, Peskar BM. Cyclooxygenase 2-implications on maintenance of gastric mucosal integrity and ulcer healing: Controversial issues and perspectives. Gut 2001;49:443-53.

3. Takeuchi K, Ueki S, Okabe S. Importance of gastric motility in the pathogenesis of indomethacin-induced gastric lesions in rats. Dig Dis Sci 1986;31:1114-22.
4. Takeuchi K, Miyazawa T, Tanaka A, Kato S, Kunikata T. Pathogenic importance of intestinal hypermotility in NSAID-induced small intestinal damage in rats. Digestion 2002;66:30-41.
5. Bozkurt A, Yuksel M, Haklar G, Kurtel H, Yegen BC, Alican I. Adenosine protects against indomethacin-induced gastric damage in rats. Dig Dis Sci 1998;43:1258-63.
6. Takeuchi K, Satoh H. NSAID-induced small intestinal damage--Roles of various pathogenic factors. Digestion 2015;91:218-32.
7. Liang TY, Deng RM, Li X, Xu X, Chen G. The role of nitric oxide in peptic ulcer: A narrative review. Med Gas Res 2021;11:42-5.
8. Kato S, Kitamura M, Korolkiewicz RP, Takeuchi K. Role of nitric oxide in regulation of gastric acid secretion in rats: Effects of NO donors and NO synthase inhibitor. Br J Pharmacol 1998;123:839-46.
9. Satoh H, Amagase K, Takeuchi K. Mucosal protective agents prevent exacerbation of NSAID-induced small intestinal lesions caused by antisecretory drugs in rats. J Pharmacol Exp Ther 2014;348:227-35.
10. Guha P, Dey A, Chatterjee A, Chattopadhyay S, Bandyopadhyay SK. Pro-ulcer effects of resveratrol in mice with indomethacin-induced gastric ulcers are reversed by L-arginine. Br J Pharmacol 2010;159:726-34.
11. Elliott SN, Wallace JL. Nitric oxide: A regulator of mucosal defense and injury. J Gastroenterol 1998;33:792-803.
12. Wallace JL, Miller MJ. Nitric oxide in mucosal defense: A little goes a long way. Gastroenterology 2000;119:512-20.
13. Salvemini D, Kim SF, Mollace V. Reciprocal regulation of the nitric oxide and cyclooxygenase pathway in pathophysiology: Relevance and clinical implications. Am J Physiol Regul Integr Comp Physiol 2013;304:R473-87.
14. Khawla AS, Edin S, Alta'ee A. Impact of lipid-lowering drugs on lipid profile in hyperlipidemia patients. Med J Babylon 2023;20:274.
15. Liao JK, Laufs U. Pleiotropic effects of statins. Annu Rev Pharmacol Toxicol 2005;45:89-118.
16. Kaleli-Durman D, Alp-Yildirim FI, Ozdemir O, Uydes-Dogan BS. Relaxant effect of atorvastatin on isolated rat gastric fundus strips: Implications for Ca(2+)-signalling mechanisms. Can J Physiol Pharmacol 2019;97:413-21.
17. Kamar SS, Latif NSA, Elrefai MFM, Amin SN. Gastroprotective effects of nebivolol and simvastatin against cold restraint stress-induced gastric ulcer in rats. Anat Cell Biol 2020;53:301-12.
18. Percie du Sert N, Hurst V, Ahluwalia A, Alam S, Avey MT, Baker M, *et al*. The ARRIVE guidelines 20: Updated guidelines for reporting animal research. PLoS Biol 2020;18:e3000410.
19. Guth PH, Aures D, Paulsen G. Topical aspirin plus HCl gastric lesions in the rat cytoprotective effect of prostaglandin, cimetidine, and probanthine. Gastroenterology 1979;76:88-93.
20. Sun J, Zhang X, Broderick M, Fein H. Measurement of nitric oxide production in biological systems by using Griess reaction assay. Sensors 2003;3:276-84.
21. Aguilar Diaz De Leon J, Borges CR. Evaluation of oxidative stress in biological samples using the thiobarbituric acid reactive substances assay. J Vis Exp 2020;159:1-22.
22. Almukhtar H, Garle MJ, Smith PA, Roberts RE. Effect of simvastatin on vascular tone in porcine coronary artery: Potential role of the mitochondria. Toxicol Appl Pharmacol 2016;305:176-85.
23. Hawkins C, Hanks GW. The gastroduodenal toxicity of nonsteroidal anti-inflammatory drugs: A review of the literature. J Pain Symptom Manage 2000;20:140-51.
24. Elliott SN, McKnight W, Cirino G, Wallace JL. A nitric oxide-releasing nonsteroidal anti-inflammatory drug accelerates gastric ulcer healing in rats. Gastroenterology 1995;109:524-30.
25. Hawkey CJ, Karrasch JA, Szczepanski L, Walker DG, Barkun A, Swannell AJ, *et al*. Omeprazole compared with misoprostol for ulcers associated with nonsteroidal antiinflammatory drugs omeprazole versus misoprostol for NSAID-induced ulcer management (OMNIUM) study group. N Engl J Med 1998;338:727-34.
26. Husain A, Al-Shimmery S, Hamed M, Al-Alwany O, Chabuck ZA, Chabuck G, *et al*. Assessment of tumor necrosis factor-α, interleukin-17, and vitamin D3 levels on a group of gastrointestinal tumor patients in Babylon Provence, Iraq. Med J Babylon 2023;20:362-7.
27. Bhattacharyya A, Chattopadhyay R, Mitra S, Crowe SE. Oxidative stress: An essential factor in the pathogenesis of gastrointestinal mucosal diseases. Physiol Rev 2014;94:329-54.
28. Abdeen A, Aboubakr M, Elgazzar D, Abdo M, Abdelkader A, Ibrahim S, *et al*. Rosuvastatin attenuates piroxicam-mediated gastric ulceration and hepato-renal toxicity in rats. Biomed Pharmacother 2019;110:895-905.
29. Kunikata T, Tanaka A, Miyazawa T, Kato S, Takeuchi K. 16,16-Dimethyl prostaglandin E2 inhibits indomethacin-induced small intestinal lesions through EP3 and EP4 receptors. Dig Dis Sci 2002;47:894-904.
30. Shishehbor MH, Brennan ML, Aviles RJ, Fu X, Penn MS, Sprecher DL, *et al*. Statins promote potent systemic antioxidant effects through specific inflammatory pathways. Circulation 2003;108:426-31.
31. Zinellu A, Mangoni AA. A systematic review and meta-analysis of the effect of statins on glutathione peroxidase, superoxide dismutase, and catalase. Antioxidants (Basel) 2021;10:1-16.
32. Ota H, Eto M, Kano MR, Kahyo T, Setou M, Ogawa S, *et al*. Induction of endothelial nitric oxide synthase, SIRT1, and catalase by statins inhibits endothelial senescence through the Akt pathway. Arterioscler Thromb Vasc Biol 2010;30:2205-11.
33. Su Y, Xu Y, Sun YM, Li J, Liu XM, Li YB, *et al*. Comparison of the effects of simvastatin versus atorvastatin on oxidative stress in patients with type 2 diabetes mellitus. J Cardiovasc Pharmacol 2010;55:21-5.
34. Li J, Sun YM, Wang LF, Li ZQ, Pan W, Cao HY. Comparison of effects of simvastatin versus atorvastatin on oxidative stress in patients with coronary heart disease. Clin Cardiol 2010;33:222-7.
35. Petersson J, Phillipson M, Jansson EA, Patzak A, Lundberg JO, Holm L. Dietary nitrate increases gastric mucosal blood flow and mucosal defense. Am J Physiol Gastrointest Liver Physiol 2007;292:G718-24.
36. Calatayud S, Sanz MJ, Canet A, Bello R, de Rojas FD, Esplugues JV. Mechanisms of gastroprotection by transdermal nitroglycerin in the rat. Br J Pharmacol 1999;127:1111-8.
37. Nakata S, Tsutsui M, Shimokawa H, Yamashita T, Tanimoto A, Tasaki H, *et al*. Statin treatment upregulates vascular neuronal nitric oxide synthase through Akt/NF-kappaB pathway. Arterioscler Thromb Vasc Biol 2007;27:92-8.
38. Granger DN, Kvietys PR. Reperfusion injury and reactive oxygen species: The evolution of a concept. Redox Biol 2015;6:524-51.
39. Satoh H. Role of dietary fiber in formation and prevention of small intestinal ulcers induced by nonsteroidal anti-inflammatory drug. Curr Pharm Des 2010;16:1209-13.
40. Heeba GH, Hassan MK, Amin RS. Gastroprotective effect of simvastatin against indomethacin-induced gastric ulcer in rats: Role of nitric oxide and prostaglandins. Eur J Pharmacol 2009;607:188-93.
41. Birnbaum Y, Ye Y, Rosanio S, Tavackoli S, Hu ZY, Schwarz ER, *et al*. Prostaglandins mediate the cardioprotective effects of atorvastatin against ischemia-reperfusion injury. Cardiovasc Res 2005;65:345-55.