IOSR Journal Of Pharmacy
(e)-ISSN: 2250-3013, (p)-ISSN: 2319-4219
Volume 11, Issue 6 Series. I (June 2021), PP. 01-12
www.iosrphr.org

Protective Effects of Propranolol and Carvedilol on Experimentally Induced Ulcerative Colitis in Male Albino Rat

Nermeen Ramadan Ali Shaaban, Sohair Hanem Samir El-Menshawy, Amal Elsayed Salem, and Shireen Sami Mahmoud Othman
Clinical Pharmacology Department, Faculty of Medicine, Zagazig University, Egypt
Corresponding author: Nermeen Ramadan Ali Shaaban
Received 30 May 2021; Accepted 14 June 2021

ABSTRACT
Background: Ulcerative colitis (UC) is a chronic inflammatory disease of large intestine. Overproduction of free radicals, lowered antioxidant capacity, inflammation and abnormal apoptosis are involved in its pathogenesis. Propranolol and carvedilol, β-blockers with antioxidant and anti-inflammatory effects which may have a protective role in UC.
The study aimed to evaluate and compare the effects of propranolol and carvedilol on UC development and to distinguish which of them have greater beneficial effect on experimentally-induced UC in male albino rats.
Methods: Fifty male albino rats were randomly allocated to five groups with each group comprising eleven rats except control group composed of six rats. ***Group (1):*** control group, ***Group (2):*** ulcerative colitis group, ***Group (3):*** propranolol-pretreated (30 mg/kg/d), ***Group (4):*** carvedilol-pretreated (30 mg/kg/d) and ***Group (5):*** mesalazine-pretreated (300 mg/kg/d). Treated groups received drugs by oral gavage for seven days before and three days after induction of colitis. UC was induced ***in groups 2 to 5*** by intrarectal administration of 1 ml of 4% Acetic Acid (AA), while ***group (1)*** received 1 ml of normal saline solution administered intrarectally instead. To estimate the severity of AA-induced UC and the effect of propranolol and carvedilol, the following parameters were measured: colon weight/length (W/L) ratio, colon weight/body weight (CW/BW) ratio, colonic malondialdehyde (MDA) and colonic markers of inflammation [tumor necrosis factor (TNF-α) and nuclear factor kappa B (NF-κB)] and macroscopic and microscopic scorings.
Results: In UC group, colon W/L ratio, CW/BW ratio, macroscopic and microscopic scorings and colonic levels of MDA, TNF-α and NF-κB were significantly increased. Pretreatment with propranolol, carvedilol and mesalazine significantly reduced these parameters when compared to UC group. However, colon W/L ratio, CW/BW ratio, macroscopic scoring of mucosal damage and colonic MDA, TNF-α and, NF-κB levels in carvedilol-pretreated group were significantly lower than propranolol-pretreated group.
Conclusion: Both propranolol and carvedilol had a coloprotective effect against AA-induced UC depending on their ability to decreases inflammation and oxidative stress state in rat colon; but carvedilol had better effects than propranolol. Thus, carvedilol can be considered the β-blocker of benefit in patients with UC who have other co-existing diseases indicating the use of β-blockers.
KEYWORDS: Ulcerative colitis, Mesalazine, Carvedilol, Propranolol.

I. INTRODUCTION

Inflammatory bowel diseases (IBD) are most commonly represented by Crohn's disease (CD), which involves any segment of the gastrointestinal tract, and ulcerative colitis (UC), that occurs in the inner lining of the colon (large intestine) or rectum **(1)** with increasing risk of colorectal cancer resulting from pro-neoplastic effects of chronic intestinal inflammation **(2).** The incidence of IBD is increasing (150–250/100,000 population), especially in developed countries **(3).** The incidence in Egypt has increased in the past ten years **(4).**

The cytokines most associated with UC are tumor necrosis factor (TNF)-α, interleukin (IL)-6, and IL-1β **(5)**. The IL-1 family consists of three members: IL-1α, IL-1β, and IL-1 receptor antagonist (IL-1Ra). One of its members, IL-1β, is a major pro-inflammatory cytokine which is released by lipopolysaccharide (LPS)-stimulated immune cells **(6)**. Increased production of pro-inflammatory cytokines provokes mutual signal transduction pathways which drive the inflammatory cascade, mainly through activation of nuclear factor kappa B (NF-κB). TNF-α is elevated in the blood **(7)** stool samples **(8)** and mucosa **(9)** of patients with ulcerative colitis.

The most common comorbidities with UC included cardiovascular illness such as hypertension and heart failure, pulmonary disease. diabetes mellitus and metabolic syndrome **(10)**. Hypertension or aggravation of already existing hypertension represents a well-known side effect related to the administration of large doses of corticosteroids which often required for the flare-ups to be settled **(11).**

Beta (β)-blockers are widely indicated for the treatment of hypertension, myocardial infarction, congestive heart failure, cardiac arrhythmias and other conditions **(12).** Members of β-blockers act as classical receptor antagonists but can also stimulate signaling pathways in a G protein-independent, β-arrestin-dependent fashion **(13).** Carvedilol and propranolol are inverse agonists for Gs-dependent adenylyl cyclase (AC) activation but stimulate Extracellular Regulated-protein Kinase (ERK) pathway to varying degrees **(14),** which is one of the major signaling cassettes of the mitogen activated protein kinase (MAPK) signaling pathway**.** The ERK cascade activation will induce cellular processes that include mainly proliferation and differentiation **(15).**

We aimed in the current study evaluate and compare the effects of propranolol and carvedilol on UC development and to distinguish which of them have greater beneficial effect on experimentally-induced UC in male albino rats.

II. MATERIALS AND METHODS

A- Drugs and Chemicals:

Carvedilol powder and propranolol hydrochloride powder were purchased from Sigma-Aldrich, St Louis, Missouri, USA. Mesalazine powder (Shire Inc, USA), Acetic acid 4% solution (Sigma-Aldrich, St Louis, MO, USA), Diethyl ether solution (E.I.P.I.Co. A.R.E). ELISA Kits for Malondialdehyde (MDA) obtained from Biodiagnostic CO., USA. ELISA kits for TNF-α (Minneapolis, MN, USA) and NF-κB (Biodiagnostic CO., USA).

B- Animals:

Fifty male Wistar albino rats weighing 180-200 grams/rat were purchased from Zagazig Faculty of Veterinary Medicine- animal unit. They were housed under standard environmental conditions (28±2ºC) and 12 hours light/dark cycle. They were allowed free access to food and water ad-labitum. The animals were deprived from food but allowed free access to tap water for 12 hours prior to induction of UC. During fasting, rats were housed each in a separate cage with raised mesh bottom to prevent coprophagy (ingestion of hair and feces). All experimental protocols were approved by the ethical committee at Zagazig University.

C- Experimental Design

Fifty rats were randomly divided into the following 5 groups with each group comprising eleven rats except control group composed of six rats. ***Group (1):*** control group, ***Group (2):*** ulcerative colitis (UC) group **(16),** ***Group (3):*** propranolol-pretreated (30 mg/kg/d) **(17),** ***Group (4):*** carvedilol-pretreated (30 mg/kg/d) **(18)** and ***Group (5):*** mesalazine-pretreated (300 mg/kg/d) as reference standard **(19).** UC was induced on the 8th day ***in groups 2 to 5*** by intrarectal administration of 1 ml of 4% Acetic Acid (AA), while ***group (1)*** received 1 ml of normal saline solution administered intrarectally instead. Drugs are freshly prepared, dissolved in distilled water and administered by oral gavage to pretreated groups for 7 days before and 3 days after induction of colitis, while control group and UC group were received 10 mL/kg/day of distilled water orally for 7 days before and 3 days after intrarectal injection of normal saline solution and acetic acid, respectively. On the 11th day the animals were sacrificed and tissue samples were collected.

D- Induction of Ulcerative Colitis:

On the 8th day, animals were kept fasting for 12 hours (overnight) and UC was induced next morning in ***groups 2, 3, 4 and 5.*** Rats were anaesthetized using low-dose ether then they were kept in Trendlenberg position during the process. An 8 mm fetal Scalp Canula was advanced 8 cm from the anus and 1 ml of 4% acetic acid (AA) was given slowly intrarectally. Then, 2 ml of air was ejected to complete distribution of AA in colon, after that they were kept upside down position for thirty seconds to prevent leakage, as well as the rest of the solution was aspirated with rinsing the colon with 5 ml saline for 20 seconds **(20).** ***Group 1*** (normal control) received 1 ml of normal saline solution intrarectally instead.

E- Preparation of colonic tissue samples:

Animals' weights were measured just before sacrification using digital weight scale. Then all animals were sacrificed by cervical dislocation. Longitudinal abdominal incision was done, and the entire colon was dissected totally through proximal end, gently flushed with saline, placed on an ice-cold plate and blotted on filter paper to dry. Each colon was weighed and colon weight /body weight ratio was estimated. It was used as a parameter to assess the degree of edema and considered as an indicator of severity of colitis **(21).** Also, colon length was measured**.** Then the proximal 5 cm was scored macroscopically and maintained in formalin 10% for microscopic studies. The distal 5 cm was collected in liquid nitrogen and sent for laboratory investigation.

F- Biochemical assays

The Lipid peroxidation product, malondialdehyde (MDA), was estimated in the colonic tissue homogenates using MDA ELISA Kits (Biodiagnostic CO., USA) according to the manufacturer's instructions. Pro-inflammatory markers were assessed in in the colonic tissue homogenates using ELISA kits for tumor necrosis factor (TNF)-α (Minneapolis, MN, USA) and nuclear factor kappa B (NF-κB) (Biodiagnostic CO., USA) according to the manufacturer's instructions.

G- Macroscopic examination:

Morphologic injury was evaluated using the scoring system reported by **Bell et al. (22)** in the **Table 1.** Examination and scoring were done using magnifying lens by two different observers blinded to each other to eliminate our bias.

Table (1): Criteria for scoring "macroscopic mucosal damage" components of combined damage score:

No macroscopic changes	0
Mucosal redness	1
Mild mucosal edema, mild bleeding or tiny erosions	2
Moderate mucosal edema, mild bleeding ulcers	3
Large ulcers, extensive edema and tissue necrosis	4
Major sites of damage extending > 1 cm along length of colon	5
Major sites of damage extending > 2 cm along length of colon, with score increasing by 1 for each additional cm	6-10

H- Microscopic examination:

Tissue specimen prepared for histopathological examination were fixed in 10% formalin, put in paraffin and sliced into four-µm sections. Paraffin sections were deparaffinized with xylene, hydrated and stained with H & E for scoring mucosal damage. The grade of inflammation of the colon was evaluated using the scoring system in **table (2) (Fabia et al. (23).**

Table (2): Histological scoring system:

Histological changes	Score
Loss of mucosal architecture	0-3
Cellular infiltration	0-3
Muscle thickening	0-3
Crypt abscess formation	0-1
Goblet cell depletion	0-1

I- Statistical Analysis

The obtained results were tabulated as means ± SE. Comparison between different groups were made using one-way analysis of variances (one-way ANOVA) followed by Post-Hoc (least significant difference "LSD") tests as described by **Armitage and Berry (24)**. The differences were considered to be significant when $p < 0.05$. Statistical Package of Social Sciences (SPSS) computer software (version 16) was used to carry out the statistical analysis.

III. RESULTS

A- Effects on colon weight/length (W/L) ratio:

After induction of UC, the animals appeared to have diarrhea, soft stool, and rectal bleeding (in the first 2 days after colitis induction). Colon weight also markedly increased due to tissue edema. Colon W/L ratio was significantly ($p<0.05$) increased from 96.17±3.27 in control group to 174.2±4.09 mg/cm.

Colon W/L ratios in propranolol-pretreated and carvedilol-pretreated groups were significantly ($p<0.05$) decreased to 148.8±4.22 and 134.5±2.84 mg/cm, respectively, as compared to UC group. Moreover, both groups were significantly ($p<0.05$) higher than mesalazine-pretreated group. However, the value of carvedilol-pretreated group was significantly ($p<0.05$) lower than that of propranolol-pretreated group **(Table 3)**.

B- Effects on colon weight/body weight (CW/BW) ratio:

Regarding colon Weight/ Body Weight (the ratio of the distal colon weight to animal body weight) was also markedly increased in UC due to tissue edema. CW/BW ratio value significantly ($p<0.05$) increased from 3.837±0.241 in control group to 8.980±0.420 mg/gm in UC group. CW/BW ratios in propranolol-pretreated and carvedilol-pretreated groups were significantly ($p<0.05$) decreased to 6.980±0.283 and 5.925±0.172 mg/gm, respectively, as compared to UC group. Moreover, both groups were significantly ($p<0.05$) higher than

mesalazine-pretreated group. However, the value of carvedilol-pretreated group was significantly ($p<0.05$) lower than that of propranolol-pretreated group **(Table 3)**.

C- Effects on colonic malondialdehyde (MDA) level:

Colonic MDA level in control group was 38.60±3.74 nmol/mg. In UC group, colonic MDA level significantly ($p<0.05$) increased from 38.60±3.74 in control group to 174.10±6.35 nmol/mg. Colonic MDA level in propranolol-pretreated and carvedilol-pretreated groups were significantly ($p<0.05$) decreased to 121.5±3.89 and 98.35±3.86 nmol/mg, respectively, as compared to UC group. Moreover, both groups were significantly ($p<0.05$) higher than mesalazine-pretreated group. However, the value of carvedilol-pretreated group was significantly ($p<0.05$) lower than that of propranolol-pretreated group **(Table 4)**.

D- Effects on colonic tumor necrosis factor-α (TNF-α) level:

In UC, colonic TNF-α level significantly ($p<0.05$) increased from 15.83±0.62 in control group to 93.43±3.51 pg/ml. Colonic TNF-α level in propranolol-pretreated and carvedilol-pretreated groups were significantly ($p<0.05$) decreased to 72.12±2.31 and 58.23±2.14 pg/ml, respectively, when compared to UC group. Moreover, both groups were significantly ($p<0.05$) higher than mesalazine-pretreated group. However, the value of carvedilol-pretreated group was significantly ($p<0.05$) lower than that of propranolol-pretreated group **(Table 4)**.

E- Effect on colonic nuclear factor kappa B (NF-κB) level:

Colonic NF-κB level in UC group significantly ($p<0.05$) increased from 103.30±4.01 in control group to 242.20±5.59 pg/mg. Colonic NF-κB level in propranolol-pretreated and carvedilol-pretreated groups were significantly ($p<0.05$) decreased to 198.10±7.59 and 168.90±4.46 pg/mg, respectively, when compared to UC group. Moreover, both groups were significantly ($p<0.05$) higher than mesalazine-pretreated group. However, the value of carvedilol-pretreated group was significantly ($p<0.05$) lower than that of propranolol-pretreated group **(Table 4)**.

F- Effect on macroscopic scoring for mucosal damage:

In control group, gross inspection of the entire colon under a magnifying glass (×3) showed normal smooth mucosa and the mean macroscopic score of mucosal damage was 0.167±0.167 **(Photo 1, Table 5).** In UC group, the colon showed edema, hyperemia of mucosa with continuous hemorrhagic large, ulcerated area, elevated edematous pseudopolyps and severe tissue necrosis and the mean macroscopic score of mucosal damage was significantly ($P<0.05$) increased from 0.167±0.167 in control group to 3.800±0.133 **(Photo 2, Table 5).** In propranolol-pretreated group, the colonic mucosa showed hemorrhagic ulcerated areas and elevated edematous pseudopolyps and the mean macroscopic score decreased significantly ($P<0.05$) from 3.800±0.133 in UC group to 3.000±0.211 **(Photo 3, Table 5).** Carvedilol-pretreated group showed mucosal ulcerated areas in between elevated edematous pseudopolyps and the mean macroscopic score decreased significantly ($P<0.05$) from 3.800±0.133 in UC group to 2.200±0.133. This value was also significantly ($p<0.05$) lower than that of propranolol-pretreated group 3.000±0.211 **(Photo 4, Table 5)**. The colonic mucosa of mesalazine-pretreated group showed showing mucosal edema with mild bleeding **(Photo 5, Table 5)**. The mean macroscopic score was 1.500±0.167 which was significantly ($p<0.05$) lower than that of UC, propranolol-pretreated and carvedilol-pretreated groups.

G- Effect on histopathological changes and microscopic scoring:

Light microscopic examination of colon tissues for microscopic examination of UC group showed markedly disturbed mucosal architecture, severe inflammation infiltrating mucosa and submucosa with thickening of muscle layer, crypt abscess formation and loss of goblet cells in mucosal glands. The mean microscopic score of mucosal damage in UC group was significantly ($P<0.05$) increased from 1.500±0.224 for control group to 8.100±0.277 **(Photo 1, Table 5)**. Propranolol-pretreated group microscopic examination showed disturbed mucosal architecture and moderate inflammation infiltrating mucosa, submucosa.

The mean microscopic score of mucosal damage significantly ($P<0.05$) decreased from 8.100±0.277 in UC group to 6.900±0.233 **(Photo 2, Table 5).** Carvedilol-pretreated group showed moderate disturbed mucosal architecture, moderate inflammation infiltrating mucosa, submucosa and muscle layer with muscle thickening **(Photo 3, Table 5).** The mean microscopic score of mucosal damage significantly ($P<0.05$) decreased to 5.500±0.224 when compared to UC group. This value was significantly ($p<0.05$) lower than that propranolol pretreated group**.** Mesalazine-pretreated group showed mild inflammation infiltrating mucosa with moderate muscle thickening. The mean microscopic score of mucosal damage significantly ($P<0.05$) decreased to 4.400±0.163 when compared to UC group. This value was significantly ($p<0.05$) lower than that of propranolol and carvedilol pretreated groups **(Photo 4, Table 5).**

Table (3): The effect (mean ± SE) of propranolol (30mg\kg), carvedilol (30mg\kg), mesalazine (300mg\kg) on colon weight/length (W/L) ratio and colon weight/body weight (CW/BW) ratio of experimentally-induced ulcerative colitis (n=6 in control group and 11 for each other groups):

Parameter / Groups	Colon W/L ratio *(mg/cm)*	CW/BW ratio *(mg/gm)*
Control group	96.17±3.27^{A}	3.837±0.241^{A}
Ulcerative colitis group	174.2±4.09^{B}	8.980±0.420^{B}
Propranolol-pretreated group	148.8±4.22^{C}	6.980±0.283^{C}
Carvedilol-pretreated group	134.5±2.84^{D}	5.925±0.172^{D}
Mesalazine-pretreated group	121.2±1.54^{E}	4.875±0.126^{A}

n: number of rats in each group
SE: Standard Error of Mean
Within the same column, values without common superscript capital letters are significantly different ($p<0.05$).

Table (4): The effect (mean ± SE) of propranolol (30mg\kg), carvedilol (30mg\kg), mesalazine (300mg\kg) on colonic MDA, TNF-α and NF-κB levels in experimentally-induced colitis (n=6 in control group and 11 for each other groups):

Parameters / Groups	MDA *(nmol/mg)*	TNF-α *(pg/ml)*	NF-κB *(pg/mg tissue)*
Control group	38.60±3.74^{A}	15.83±0.62^{A}	103.30±4.01^{A}
Ulcerative colitis group	174.10±6.35^{B}	93.43±3.51^{B}	242.20±5.59^{B}
Propranolol-pretreated group	121.5±3.89^{C}	72.12±2.31^{C}	198.10±7.59^{C}
Carvedilol-pretreated group	98.35±3.86^{D}	58.23±2.14^{D}	168.90±4.46^{D}
Mesalazine-pretreated group	78.53±3.87^{E}	43.45±1.78^{E}	145.80±3.28^{E}

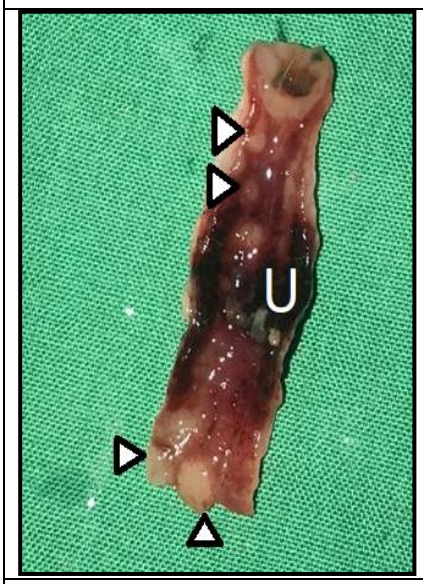
n: number of rats in each group
SE: Standard Error of Mean
Within the same column, values without common superscript capital letters are significantly different ($p<0.05$).

Table (5): The effect (mean ± SE) of propranolol (30mg\kg), carvedilol (30mg\kg), mesalazine (300mg\kg) on macroscopic and microscopic scores of mucosal damage of experimentally-induced ulcerative colitis (n=6 in control group and 11 for each other groups):

Parameters / Groups	Macroscopic score	Microscopic score
Control group	0.167±0.167^{A}	1.500±0.224^{A}
Ulcerative colitis group	3.800±0.133^{B}	8.100±0.277^{B}
Propranolol-pretreated group	3.000±0.211^{C}	6.900±0.233^{C}
Carvedilol-pretreated group	2.200±0.133^{D}	5.500±0.224^{D}
Mesalazine-pretreated group	1.500±0.167^{E}	4.400±0.163^{E}

n: number of rats in each group
SE: Standard Error of Mean
Within the same column, values without common superscript capital letters are significantly different ($p<0.05$).

Photo macrographs representing the pathological effects of propranolol (30mg\kg), carvedilol (30mg\kg), mesalazine (300mg\kg) on experimentally induced ulcerative colitis:

	**Group (1):** Control group: showing normal smooth mucosa with no macroscopic changes.
(Photo 1)	
	**Group (2):** Ulcerative colitis group: showing continuous hemorrhagic large, ulcerated area (U) and elevated edematous pseudopolyps of mucosa (arrowheads).
(Photo 2)	
	**Group (3):** Propranolol-pretreated group: showing continuous hemorrhagic large, ulcerated areas (U) and elevated edematous pseudopolyps of mucosa (arrowheads).
(Photo 3)	

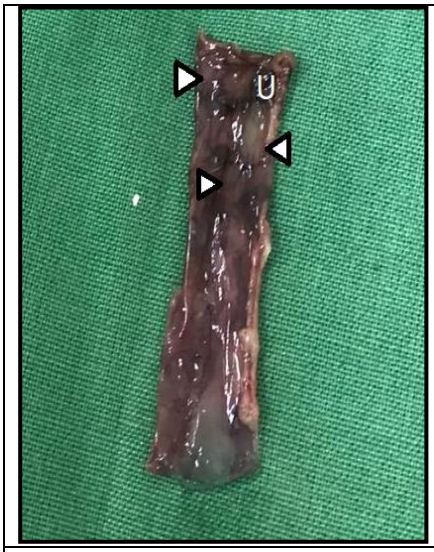
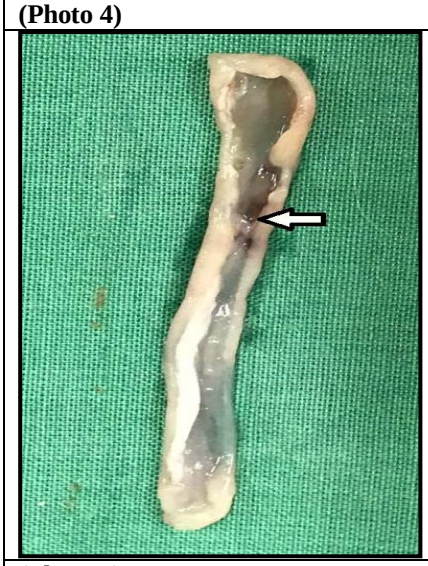
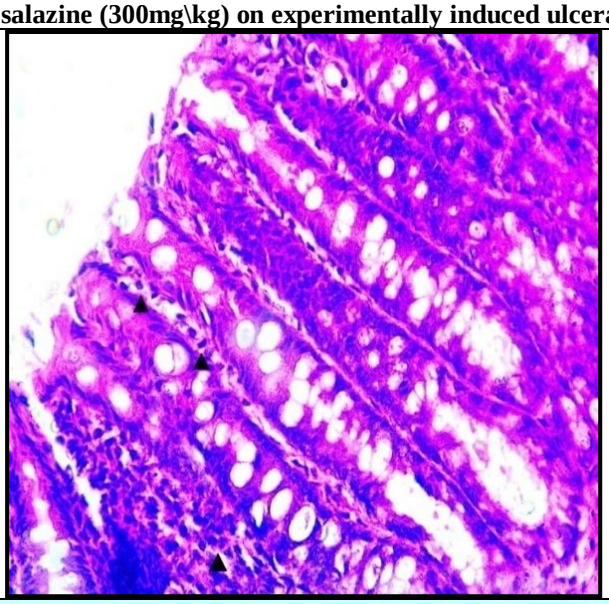
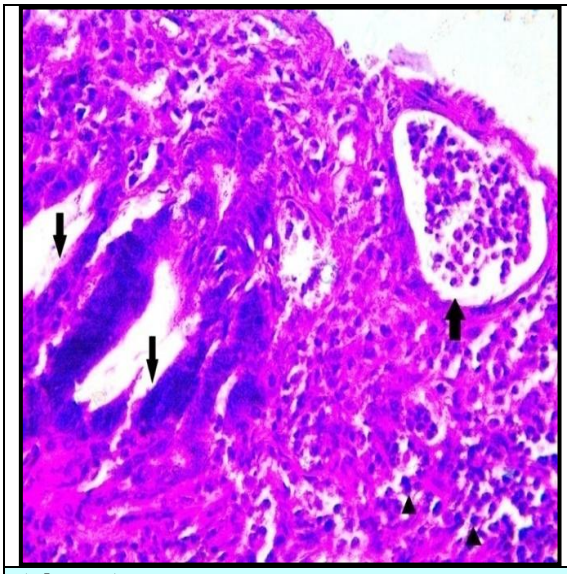
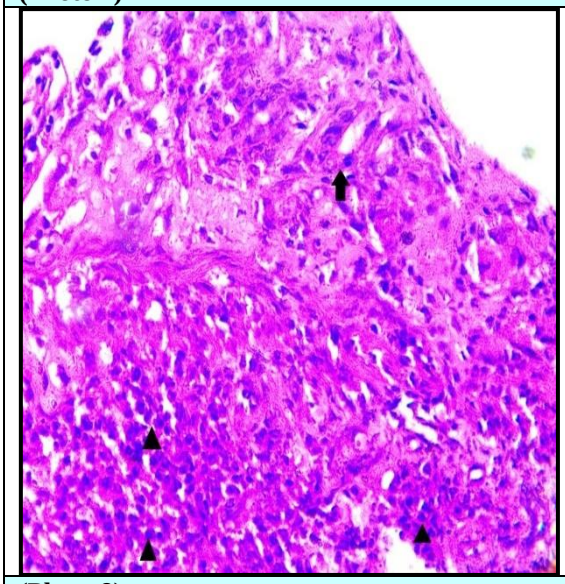
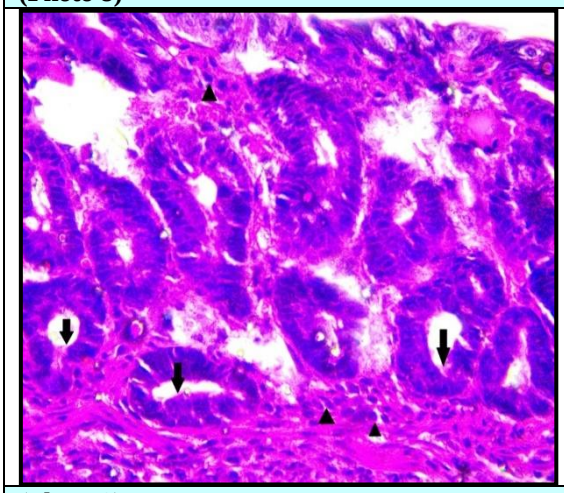
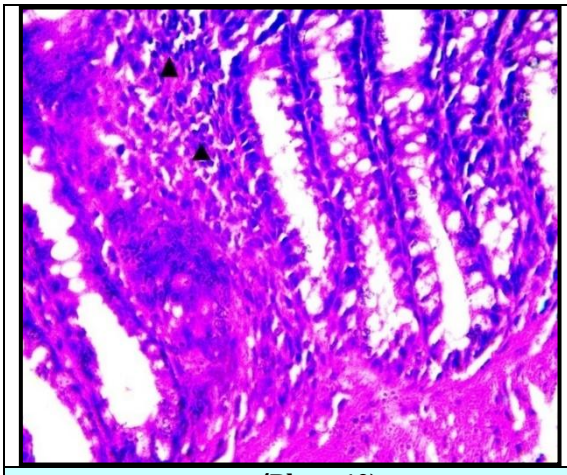
	**Group (4):** Carvedilol-pretreated group: showing hemorrhagic ulcers (U) in between elevated edematous pseudopolyps of mucosa (arrowheads).
(Photo 4)	
	**Group (5):** Mesalazine-pretreated group: showing small, tiny erosion (arrow).
(**Photo 5)**	

Photo micrographs representing the pathological effects of propranolol (30mg\kg), carvedilol (30mg\kg), mesalazine (300mg\kg) on experimentally induced ulcerative colitis:

	• **Group I:** Control group: showing normal mucosal architecture with mild inflammatory infiltrate (arrowheads) among the mucosal glands. (Hematoxylin and eosin X400).
(Photo 6)	

	**Group (2):** Ulcerative colitisgroup: showing markedly disturbed mucosal architecture with crypt abscess (upward arrow) formation, severe inflammation (arrowheads) infiltrating mucosa and loss of goblet cells (downward arrows) in mucosal glands. (Hematoxylin and eosin X400).
(Photo 7)	
	**Group (3):** Propranolol-pretreated group: showing markedly disturbed mucosal architecture and severe inflammation (arrowheads) infiltrating mucosa with remnants (upward arrow) of mucosal glands. (Hematoxylin and eosin X400).
(Photo 8)	
	**Group (4):** Carvedilol-pretreated group: showing mild disturbed mucosal architecture, severe inflammatory infiltrate (arrowheads) in the mucosa and submucosa with loss of goblet cells (downward arrows) in mucosal glands. (Hematoxylin and eosin X400).
(Photo 9)	

	**Group (5):** Mesalazine-pretreated group: showing nearly normal mucosal architecture, mild inflammation (arrowheads) infiltrating mucosa, (Hematoxylin and eosin X400).
(Photo 10)	

IV. DISCUSSION

Previous studies have demonstrated that the Acetic Acid (AA)-induced colitis in animals easily inducible model and closely resembles that of UC in humans in terms of pathogenesis, histopathological features and inflammatory mediator profile **(20).** The pathogenesis of acetic acid-induced colitis is complex, mucosal ischemia, increased vascular permeability, oxidative stress, excessive neutrophil infiltration, increased production of inflammatory mediators and cytokines have all been implicated **(25)**.

We selected male albino rats in the present study to avoid the protective effect of estrogen as **Kuralay et al. (26)** observed that there was discrepancy between researchers results which might be related to gender differential response to acetic acid. **Nie et al. (27)** also demonstrated that estrogen plays an important role in maintaining mucosal barrier function of the gastrointestinal tract, such as epithelial and physiologic barrier functions. They reported that relative absence of estrogen in postmenopausal women and men is associated with increased levels of ROS as well as with the risk of several diseases such as UC.

The current study was designed to investigate the possible protective effect of propranolol, carvedilol and mesalazine on acetic acid-induced UC in male albino rats. The following parameters were measured, colon weight/length (W/L) ratio, colon weight/body weight (CW/BW) ratio, macroscopic scoring, oxidative stress parameters in colonic tissue {malondialdehyde (MDA), markers of inflammation in colonic tissue {tumor necrosis factor α (TNF-α), and nuclear factor kappa B (NF-κB)}.

The results of the present study demonstrated that in the first two days after colitis induction with 1 ml of 4% acetic acid intrarectally, the rats appeared to have significant diarrhea, soft stool, and rectal bleeding. After sacrification, it was noticed that colon weight significantly increased in relation to body weight due to tissue edema and inflammatory changes. The rats in ulcerative colitis group showed significant increase colon weight/length (W/L) ratio and colon weight/body weight (CW/BW) ratio in relation to the control group.

These findings agree with **Ige et al. (28)** who found that instillation of acetic acid rectally cause increase in colon W/L ratio and CW/BW ratio when demonstrating the role of dietary maize formulations in the healing of experimental ulcerative colitis in male rats. **Kjellev et al. (29)** also added that the colonic W/L ratio is a highly useful and easily measurable macroscopic postmortem marker of colitis, closely reflecting both the epithelial hyperproliferation and the influx of inflammatory cells during the course of disease. They added that colon weight is increased in UC because of tissue edema and cellular infiltration. Also, colon length is reduced secondary to longitudinal muscle spasm and colon W/L ratio increased with AA-induced colitis.

In the present study, it was found that propranolol (30 mg/kg/d) significantly decreased colon W/L ratio, CW/BW ratio, macroscopic scoring as well as it significantly reduced colonic values of MDA, inflammatory cytokines (TNFα, and NF-κB).

These results in agreement with **Deng et al. (30)** who reported that propranolol effectively suppressed the expression of the proinflammatory cytokines and chemokines in colonic tissue which reflected on colon W/L ratio, CW/BW ratio and decreased mucosal redness, edema and congestion.

Rodrigues et al. (31) found that low doses of propranolol resulting in inhibition of inflammatory markers and decrease bone resorption in rats' model of experimental periodontal disease by inhibiting inflammation, reduced IL-1β, TNF-α and osteoclast differentiation without affecting heart functions or hemodynamic parameters.

As regard carvedilol pretreated group in the present study, it was found that carvedilol (30 mg/kg/d) significantly decreased colon W/L ratio, CW/BW ratio, macroscopic scoring as well as it significantly reduced colonic values of MDA, inflammatory cytokines (TNFα, and NF-κB).

These findings consistent with **Fatani et al. (23)** who reported that carvedilol (30 mg/kg/d) for 7 days markedly protected the colonic mucosa, reduced tissue edema and extravasation, attenuated cytokines production and prevented oxidative and inflammatory response in the colons of rats with AA-induced UC. **Le Leu et al. (32)** found that carvedilol can down-regulate the expression of many inflammatory mediators and cytokines in mice colitis model.

Araújo Júnior et al. (33) added that carvedilol at (1, 3, or 5 mg/kg) can reduce the stress oxidative, inflammatory response and fibrosis in ethanol-induced liver injury in a rat model through suppression of inflammatory cytokines. Furthermore, **Toyoda et al. (34)** who found that carvedilol has been shown to possess both ROS-scavenging and ROS-suppressive effects in patients with chronic heart failure. The antioxidant activity of carvedilol has been attributed to its carbazole moiety which is approximately ten-fold more potent as an antioxidant than vitamin E **(35).**

As regard comparing propranolol with carvedilol pretreated groups in the present study, carvedilol was better than propranolol as it significantly decreased colon W/L ratio, CW/BW ratio, macroscopic scoring as well as it significantly reduced colonic values of MDA, inflammatory cytokines (TNFα, and NF-κB) compared to propranolol group. The superior protective effect of carvedilol to propranolol may be due to the higher antioxidant activities and anti-inflammatory of carvedilol compared to propranolol and the difference between them in terms of their effects may be explained by their difference in receptor binding, antioxidant properties or potency for β-arrestin pathway.

These findings are in agreement with **Malig et al. (36)** who stated that the antioxidant effects of carvedilol originate from the unique carbazole moiety of carvedilol. Some of carvedilol metabolites are even more potent antioxidants and approximately 1,000-fold more potent than vitamin E **(37).** Moreover, **Esmaeeli et al. (38)** added that the affinity of carvedilol is approximately 4 times more than propranolol for β_1-adrenergic receptor (AR) and 2 times for β_2-AR.

V. CONCLUSION

Propranolol and carvedilol had a coloprotective effect against AA-induced colitis depending on their ability to decreases inflammation, oxidative stress state in rat colon. Carvedilol was better than propranolol. Thus, carvedilol can be considered the β-blocker of benefit in patient with UC especially in those with other coexisting diseases indicated β-blockers.

Conflict of Interest: No conflict of interest.

REFERENCES:

[1]. **Moura, F. A., de Andrade, K. Q., dos Santos, J. C. F., et al., (2015):** Antioxidant therapy for treatment of inflammatory bowel disease: does it work?. Redox Biology, 6, 617-639.

[2]. **Stidham, R. W., and Higgins, P. D. (2018):** Translational Research in Colorectal Cancer: Colorectal Cancer in Inflammatory Bowel Disease. Clinics in colon and rectal surgery, 31(3), 168.

[3]. **Stepaniuk, P., Bernstein, C. N., Targownik, L. E., et al., (2015):** Characterization of inflammatory bowel disease in elderly patients: a review of epidemiology, current practices and outcomes of current management strategies. Canadian Journal of Gastroenterology and Hepatology, 29(6), 327-333.

[4]. **Mostafa, E. F., Metwally, A., and Hussein, S. A. (2018):** Inflammatory Bowel Diseases Prevalence in Patients Underwent Colonoscopy in Zagazig University Hospitals. Afro-Egyptian Journal of Infectious and Endemic Diseases, 8(2), 81-87.

[5]. **Gupta, R. A., Motiwala, M. N., Mahajan, U. N., et al., (2018):** Protective effect of Sesbania grandiflora on acetic acid induced ulcerative colitis in mice by inhibition of TNF-α and IL-6. Journal of ethnopharmacology, 219, 222-232.

[6]. **Lee, H. S., Ryu, D. S., Lee, G. S., et al., (2012):** Anti-inflammatory effects of dichloromethane fraction from Orostachys japonicus in RAW 264.7 cells: suppression of NF-κB activation and MAPK signaling. Journal of ethnopharmacology, 140(2), 271-276.

[7]. **Murch, S. H., Lamkin, V. A., Savage, M. O., et al., (1991):** Serum concentrations of tumour necrosis factor alpha in childhood chronic inflammatory bowel disease. *Gut*, *32*(8), 913-917.

[8]. **Braegger, C. P., Nicholls, S., Murch, S. H., et al., (1992):** Tumour necrosis factor alpha in stool as a marker of intestinal inflammation. The Lancet, 339(8785), 89-91.

[9]. **Masuda, H., Iwai, S., Tanaka, T., et al., (1995):** Expression of IL-8, TNF-alpha and IFN-gamma m-RNA in ulcerative colitis, particularly in patients with inactive phase. Journal of clinical and laboratory immunology, 46(3), 111.

[10]. **Michalak, A., Mosińska, P., and Fichna, J. (2016):** Common links between metabolic syndrome and inflammatory bowel disease: current overview and future perspectives. Pharmacological Reports, 68(4), 837-846.

[11]. **Triantafillidis, J. K., Merikas, E., and Georgopoulos, F. (2011):** Current and emerging drugs for the treatment of inflammatory bowel disease. Drug design, development and therapy, 5, 185.

[12]. **Farzam, K., and Jan, A. (2020):** Beta blockers. StatPearls [Internet] StatPearls Publishing; Treasure Island (FL): Nov 21, 2019. Beta Blockers.

[13]. **Kim, I. M., Tilley, D. G., Chen, J., et al., (2008):** β-Blockers alprenolol and carvedilol stimulate β-arrestin-mediated EGFR transactivation. Proceedings of the National Academy of Sciences, 105(38), 14555-14560.

[14]. **Wisler, J. W., DeWire, S. M., Whalen, E. J., et al., (2007):** A unique mechanism of β-blocker action: carvedilol stimulates β-arrestin signaling. Proceedings of the National Academy of Sciences, 104(42), 16657-16662.

[15]. **Keshet, Y., and Seger, R. (2010):** The MAP kinase signaling cascades: a system of hundreds of components regulates a diverse array of physiological functions. In MAP kinase signaling protocols (pp. 3-38). Humana Press, Totowa, NJ.

[16]. **Zeytunlu, M., Korkut, M., Akgün, E., et al., (2004):** The comparative effects of calcium channel blockers in an experimental colitis model in rats. *Turk J Gastroenterol*, *15*(4), 243-249.

[17]. **Wang, W., Xia, M. X., Chen, J., et al., (2016):** Gene expression characteristics and regulation mechanisms of superoxide dismutase and its physiological roles in plants under stress. Biochemistry (Moscow), 81(5), 465-480.

[18]. **Fatani, A. J., Al-Hosaini, K. A., Ahmed, M. M., et al., (2015):** Carvedilol attenuates inflammatory biomarkers and oxidative stress in a rat model of ulcerative colitis. Drug Development Research, 76(4), 204-214.

[19]. **Faramarzpour, A., Tehrani, A. A., Tamaddonfard, E., et al., (2019):** The effects of crocin, mesalazine and their combination in the acetic acid-induced colitis in rats. In Veterinary Research Forum (Vol. 10, No. 3, p. 227). Faculty of Veterinary Medicine, Urmia University, Urmia, Iran.

[20]. **Randhawa, P. K., Singh, K., Singh, N., et al., (2014):** A review on chemical-induced inflammatory bowel disease models in rodents. The Korean journal of physiology and pharmacology: official journal of the Korean Physiological Society and the Korean Society of Pharmacology, 18(4), 279.

[21]. **Labib, D. A. A., Shaker, O. G., and Elfarouk, L. O. (2016):** Protective effects of nebivolol on acetic acid-induced ulcerative colitis in rats. Kasr Al Ainy Medical Journal, 22(3), 99.

[22]. **Bell, C. J., Gall, D. G., and Wallace, J. L. (1995):** Disruption of colonic electrolyte transport in experimental colitis. *American Journal of Physiology-Gastrointestinal and Liver Physiology*, *268*(4), G622-G630.

[23]. **Fabia, R., Willen, R., Ar'Rajab, A., et al., (1992):** Acetic acid-induced colitis in the rat: a reproducible experimental model for acute ulcerative colitis. European surgical research, 24(4), 211-225.

[24]. **Armitage, P., Berry, G., and Matthews, J. N. S. (2008):** Statistical methods in medical research. John Wiley and Sons.

[25]. **Chamanara, M., Abdollahi, A., Rezayat, S. M., et al., (2019):** Thymol reduces acetic acid-induced inflammatory response through inhibition of NF-kB signaling pathway in rat colon tissue. Inflammopharmacology, 27(6), 1275-1283.

[26]. **Kuralay, F., Yildiz, C., Ozutemiz, O., et al., (2003):** Effects of trimetazidine on acetic acid-induced colitis in female Swiss rats. Journal of Toxicology and Environmental Health Part A, 66(2), 169-179.

[27]. **Nie, X., Xie, R., and Tuo, B. (2018):** Effects of estrogen on the gastrointestinal tract. Digestive diseases and sciences, 63(3), 583-596.

[28]. **Ige, S. F., Adeniyi, M. J., Olayinka, A. T., et al., (2020):** Role of dietary maize formulations in the healing of experimental acetic acid induced ulcerative colitis in male rats. Chinese Journal of Physiology, 63(4), 156.

[29]. **Kjellev, S., Lundsgaard, D., Poulsen, S. S., et al., (2006):** Reconstitution of Scid mice with CD4+ CD25− T cells leads to rapid colitis: an improved model for pharmacologic testing. International immunopharmacology, 6(8), 1341-1354.

[30]. **Deng, Q., Chen, H., Liu, Y., et al., (2016):** Psychological stress promotes neutrophil infiltration in colon tissue through adrenergic signaling in DSS-induced colitis model. Brain, behavior, and immunity, 57, 243-254.

[31]. **Rodrigues, W. F., Madeira, M. F. M., Da Silva, T. A., et al., (2012):** Low dose of propranolol down-modulates bone resorption by inhibiting inflammation and osteoclast differentiation. British journal of pharmacology, 165(7), 2140-2151.

[32]. **Le Leu, R. K., Young, G. P., Hu, Y., et al., (2013):** Dietary red meat aggravates dextran sulfate sodium-induced colitis in mice whereas resistant starch attenuates inflammation. Digestive diseases and sciences, 58(12), 3475-3482.

[33]. **Araújo Júnior, R. F. D., Garcia, V. B., Leitão, R. F. D. C., et al., (2016):** Carvedilol improves inflammatory response, oxidative stress and fibrosis in the alcohol-induced liver injury in rats by regulating Kuppfer cells and hepatic stellate cells. PLoS One, 11(2), e0148868.

[34]. **Toyoda, S., Haruyama, A., Inami, S., et al., (2020):** Effects of carvedilol vs bisoprolol on inflammation and oxidative stress in patients with chronic heart failure. Journal of Cardiology, 75(2), 140-147.

[35]. **Alanazi, A., Fadda, L., Alhusaini, A., et al., (2020):** Antioxidant, antiapoptotic, and antifibrotic effects of the combination of liposomal resveratrol and carvedilol against doxorubicin-induced cardiomyopathy in rats. Journal of biochemical and molecular toxicology, 34(7), e22492.

[36]. **Malig, T. C., Ashkin, M. R., Burman, A. L., et al., (2017):** Comparison of free-radical inhibiting antioxidant properties of carvedilol and its phenolic metabolites. MedChemComm, 8(3), 606-615.

[37]. **de Oliveira, A. L. C., dos Santos-Silva, A. M., da Silva-Júnior, A. A., et al. (2020):** Cholesterol-functionalized carvedilol-loaded PLGA nanoparticles: anti-inflammatory, antioxidant, and antitumor effects. Journal of Nanoparticle Research, 22, 1-14.

[38]. **Esmaeeli, A., Keshavarz, Z., Dehdar, F., et al., (2020):** The effects of carvedilol, metoprolol and propranolol on cisplatin-induced kidney injury. Drug and Chemical Toxicology, 1-7.

Nermeen Ramadan Ali Shaaban, et. al. "Protective Effects of Propranolol and Carvedilol on Experimentally Induced Ulcerative Colitis in Male Albino Rat." *IOSR Journal of Pharmacy (IOSRPHR)*, 11(06), 2021, pp. 01-12.