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ORIGINAL PAPER

Effects of emodin and metformin on biochemical, histological and oxidative stress parameters in streptozotocin-induced diabetic rats*

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Abstract

Diabetes mellitus (DM) is a lifelong metabolic disease with a high mortality rate, which reduces the quality of life of many people worldwide. Metformin is a hypoglycemic drug usually used to treat type 2 diabetes. Emodin has various protective effects and is widely used in holistic medicine. The aim of this study was to investigate the effects of emodin, metformin and the emodin + metformin combination in liver and kidney tissues of rats with diabetes induced by streptozotocin (STZ). For this purpose, 5 groups: I – Group Control (CG), II – Group STZ control (SC), III – Group STZ + metformin 250 mg kg^{-1} (SM), IV – Group STZ + emodin 40 mg kg^{-1} (SE) and V – Group STZ + metformin 250 mg kg^{-1} + emodin 40 mg kg^{-1} (SME) were formed with 7 rats in each group. Liver, kidney and blood samples were taken at the end of the experiment. In the study, superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx), glutathione-S-transferase (GST), glutathione reductase (GR), reduced glutathione (GSH) and malondialdehyde (MDA) levels in liver and kidney tissues were analyzed. In addition, histopathology and immunohistochemical and serum toxicity biomarkers (AST, ALT, BUN and CREA) were

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analyzed. According to the experimental results, there was an increase in serum biomarker values in the SC group compared to the control group (p values for AST, ALT, CREA and BUN were p=0.005, p=0.001, p=0.006, p=0.001, respectively). Some antioxidant parameter values increased in SM, SE and SME groups compared to CG and SC groups (p=0.001 for GR in liver tissue and p=0.032 for GPX in kidney tissue). Histopathologic and immunohistochemical observations also supported biochemical parameter findings. In conclusion, oxidative stress parameters in liver and kidney tissue, serum samples, histopathologic and immunologic findings suggest that metformin, emodin and the metformin + emodin combination have positive effects on nephrotoxic and hepatotoxic side effects.

Keywords: emodin, hepatotoxicity, metformin, nephrotoxicity, streptozotocin

INTRODUCTION

Diabetes mellitus (DM), with incidence increasing rapidly in recent years, is one of the most common endocrine diseases characterized by hyperglycemia (Ghosh et al. 2011, Nisari 2019, Sahin et al. 2020). Oxidative stress is among the most important factors in the pathophysiology of this disease (Das et al. 2012). In diabetics, oxidative stress causes tissue damage, leading to excessive production of reactive oxygen species ROS (Halliwell, Gutteridge 2007, Ibuki et al. 2020). Enzymes such as superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx) can prevent damage caused by ROS and have a place in antioxidant defense systems produced during normal metabolism in the body (Ibuki et al. 2020). The liver and kidney are the organs most affected by diabetes. The liver plays an important role in the regulation of glucose metabolism by using glucose molecules as an energy source to maintain glucose levels in the body (Dubey et al. 2020). AST, ALT and ALP values are among the indicators of hepatocellular damage (Mathur et al. 2016, Dubey et al. 2020). In addition, diabetic nephropathy (DN), one of the complications of diabetes, is a clinical condition in which the structure and function of the kidney is impaired. For this condition, function tests such as BUN and creatinine provide information about kidney tissue (Dabla 2010, Dubey et al. 2020).

Metformin is a synthetic drug that improves type 2 diabetes by inhibiting glucose release from the liver (Tseng 2019, Balkan et al. 2020, Beheshti et al. 2022). Metformin was reported to have anti-inflammatory (Cameron et al. 2016) and antioxidant (Hou et al. 2010) properties (Beheshti et al. 2022). In recent years, the use of herbal medicines with strong antioxidant and anti-inflammatory properties has become increasingly common, especially to minimize hyperglycemia and other metabolic disorders associated with diabetes (Yazdi et al. 2020). One of these herbal medicines, emodin (6-methyl-1,3,8-trihydroxyanthraquinone), is a chemical compound belonging to the anthraquinone family (Mitra et al. 2022) which has therapeutic potential against diabetes-related complications (Han et al. 2019, Martorell et al. 2021). This polyphenol has a wide range of pharmacological effects, such

as antidiabetic, antiviral, antibacterial, immunosuppressive, neuroprotective, hepatoprotective, anti-inflammatory, and anti-atherosclerotic activities (Gupta et al. 2014, Dong et al. 2016, Martorell et al. 2021).

Yazdi et al. (2020) investigated the effect of *Artemisia turanica* (AT) and metformin administration, while Figueiredo et al. (2020) investigated the effect of lycopene and metformin administration against oxidative stress in diabetic rats. In some studies on emodin, Wu et al. (2021) investigated the effect of low-dose emodin on hypercholesterolemia in a high-cholesterol rat model, while Wang et al. (2021) conducted research on the protective effects of emodin against cyclophosphamide-induced gonadotoxicity.

Although there are some studies about emodin and metformin separately in the literature, the number of studies using these substances together is very limited. Therefore, this study was carried out to determine the single/combined effects of emodin and metformin on liver and kidney dysfunction and oxidative stress in rats with diabetes induced by streptozotocin (STZ).

MATERIALS AND METHODS

Reagents

Streptozotocin, emodin, metformin, thiobarbituric acid (TBA), butylated hydroxytoluene (BHT), trichloroacetic acid (TCA), ethylenediaminetetraacetic acid (EDTA), reduced glutathione (GSH), metphosphoric acid, 5,5'-dithiobis (2 nitrobenzoic acid) (DTNB), Tris, 1-chloro-2,4-dinitrobenzene (CDNB), oxide glutathione (GSSG), nicotinamide adenine dinucleotide phosphate (NADP), potassium dihydrogen phosphate () and sodium chloride of technical grade used in this study were obtained from Sigma Chemical Company, St. Louis, MO, USA. Reagents for antioxidant enzyme analysis were purchased from Randox Laboratories Ltd.

Experimental animals

The experimental procedures performed in this study were carried out with the approval of Van Yuzuncu Yil University Animal Experiments Local Ethics Committee dated 26/12/2019 and numbered 12. The rats were obtained from Van Yuzuncu Yil University Experimental Animals Unit. *Wistar albino* rats weighing between 150-200 g were used in the experiments. Rats were fed *ad libitum* at room temp. (25±1°C) with 12 h light/12 h dark period.

Animals and experimental design

The experimental groups consisted of 5 groups with 7 rats in each group. Diabetic rats in this group underwent an experimentally-induced diabetic model with STZ. The groups are given in Table 1.

Table 1

Experimental groups

Groups		Explanation
Name	abbreviation	
Control	CG	ordinary rat food and tap water
STZ Control	SC	STZ (45 mg kg^{-1}) (intraperitoneal (ip)
STZ + metformin	SM	STZ (45 mg kg^{-1}) (ip) and 250 mg kg^{-1} metformin gastric gavage
STZ + emodin	SE	STZ (45 mg kg^{-1}) (ip) and 40mg/kg emodin gastric gavage
STZ + metformin + emodin	SME	STZ (45 mg kg^{-1}) (ip) and 250 mg kg^{-1} metformin and 40 mg/kg emodin gastric gavage

In the experimental groups, emodin 40 mg kg^{-1} (Zhan et al. 2000, Dang et al. 2008) and metformin 250 mg kg^{-1} (Figueiredo et al. 2020) were used daily. The rats in groups other than the control group were fasted overnight and diabetes was induced by intra-peritoneal injection with a single dose of STZ (45 mg kg^{-1}) (Chougala et al. 2012) dissolved in freshly prepared 0.1 M citrate buffer (pH=4.5) and the study lasted for 28 days. Fasting blood glucose values of the rats were measured 3 days after STZ injection in the diabetes-induced groups and rats with values above 300 mg dL^{-1} (Afify et al. 2018) were considered diabetic. At the end of the study, the rats were sacrificed after being anesthetized with 10% ketamine and liver, kidney and blood samples were taken.

Isolation of supernatants from liver and kidney tissue

For the determination of antioxidant enzymes, reduced glutathione level and malondialdehyde in tissues, the tissue extraction process was performed as follows; for extraction, a buffer containing 0.32 mol L^{-1} sucrose, 1 mmol L^{-1} EDTA, 10 nm L^{-1} TrisHCl (pH7.4) was prepared, 500 mg tissues were weighed on a digital balance (Chyo Jl-180) and 5 mL cold buffer was added. The tissues were thoroughly crushed with a glass baguette and homogenized in an ultrasonic homogenizer for 3-5 minutes. The homogenate was centrifuged at 9500 rpm for 30 min at +4°C in a refrigerated centrifuge (BHG Hermle). Clear supernatants obtained from liver and kidney tissues were prepared for analysis (Bati et al. 2020).

Biochemical analysis

In liver and kidney tissues, glutathione peroxidase (GPx) and superoxide dismutase (SOD) were measured in tissue according to the spectrophotometric kit protocol of Randox company. The activity of catalase (CAT), which catalyzes the hydrolysis of hydrogen peroxide to oxygen and water, was tested

by the Aebi (100) method (Aebi 1974). Glutathione reductase (GR) activity was tested by the Carlberg and Mannervik method (Carlberg, Mannervik 1985). Glutathione S-Transferase (GST) activity was determined by measuring the binding strength of glutathione to 1-chloro-2,4-dinitrobenzene (CDNB) spectrophotometrically at 340 nm wavelength (Mannervik, Guthenberg 1981). Reduced glutathione (GSH) was determined by measuring the yellow color formed by the reaction of sulfhydryl groups with DTNB (5,5'-2-dithiobis nitrobenzoic acid) at 412 nm against a blind sample (Beutler et al 1963). The level of malondialdehyde (MDA), a tissue lipid peroxidation product, was determined by spectrophotometric measurement at 532 nm of the violet color formed by TBA (Jain et al. 1989).

Serum AST, ALT, BUN, CREA and glucose detection

Serum biochemical parameters AST, ALT, BUN and creatinine (CREA) were measured in an autoanalyzer (Cobas 8000/Roche/Germany/serial No 1296-08) using Roche kits. Fasting blood glucose was detected by using a one touch glucometer (Accu-Chek sensor) of Roche Diagnostics, Germany, in the beginning and at the end of the experiment.

Histopathologic investigation

Tissue samples taken at the end of the evaluation were fixed in 10% formaldehyde solution for 48 h and embedded in paraffin blocks after routine tissue follow-up procedures. Sections with a thickness of 4 μm were taken from each block and preparations were made for histopathologic examination by staining with hematoxylin-eosin (HE) and examined by light microscopy (Olympus BX 51, Japan). The sections were evaluated as none (-), very mild (+), mild (++), moderate (++++) and severe (++++) according to histopathologic features (Kilic et al. 2019).

Immunohistochemical investigation

For immune peroxidase investigation, tissue sections placed on adhesive (poly-L-Lysin) slides were paraffinized and dehydrated. Endogenous peroxidase was then activated in 3% H_2O_2 for 10 min. The tissues were then boiled in 1% antigen retrieval solution (citrate buffer (pH^+6.1) 100X) and allowed to cool at room temperature. Sections were incubated with protein block for 5 min to prevent nonspecific background staining. Then, primary antibody (GFAP Cat No: ab68428, Reconstitution: 1/100, US) was added to the tissues and incubated according to the instructions for use. 3-3' Diaminobenzidine (DAB) chromogen was used as chromogen in the tissues. The stained sections were examined by light microscopy (Zeıss AXIO Germany). The examined preparations were evaluated as none (-), very mild (+), mild (++), moderate (++++) and severe (++++) according to their immunopositivity (Yildirim et al. 2019).

Statistical analysis

The Kruskal-Wallis test, one of the nonparametric tests, was used for statistical analysis of the differences between data obtained from the experimental groups, and the Mann Whitney *U* test was used for the comparison of paired groups. SPSS13.0 and R package program (R Core Team 2021) were used for these statistical analyses. The results are expressed as mean ± standard deviation (SD). $P<0.05$ was considered statistically significant.

RESULTS

Fasting blood glucose levels of the groups in Table 2 showed that there was an increase in SC, SM, SE and SME groups from the beginning of the experiment. Especially in SE and SME groups, there was a decrease after the first week and an increase in fasting blood glucose levels in the last week.

In the present study, according to Figure 1, there were statistically significant increases in ALT level in SC, SM and SE groups compared to the

Table 2

Mean blood glucose levels of rats in experimental groups (mean±SD)

Groups	CG	SC	SM	SE	SME
Beginning	96.1±10.2	101.2±35.7	105.4±70.4	103.1±25.7	103.5±65.4
Week I	95.8±8.1	350.3±90.5	380.9±82.9	398.7±69.1	402.7±56.9
Week II	106.4±7.7	425±49.8	421.1±146.6	380.6±122.2	345.6±124.6
Week III	105.3±17.5	465.4±88.9	410.6±152.0	361.3±153.5	259.1±132.3
Week IV	127.1±41.0	517±31.4	484.7±160.7	480.5±4.7	277.3±147.0

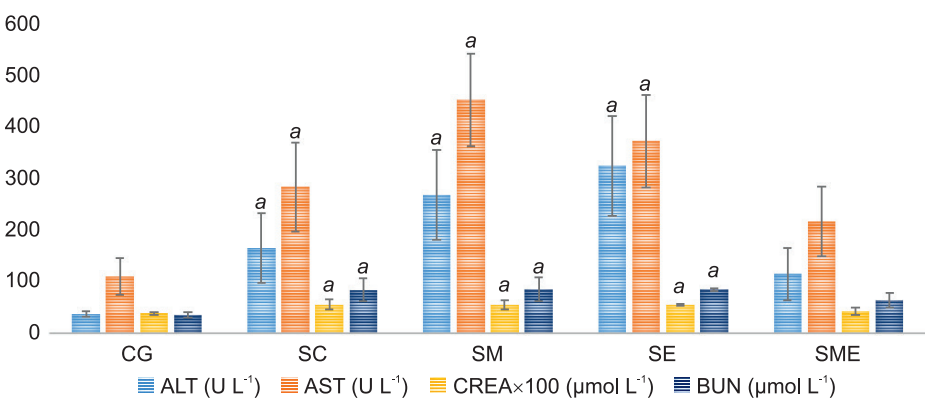

Fig. 1. ALT, AST, BUN and CREA levels among serum biochemical parameters:
a – the difference compared to the CG group is statistically significant ($p<0.05$),
b – the difference is statistically significant compared to SC group ($p<0.05$),

CG group. There were statistically significant increases in AST level in SC, SM and SE groups compared to the CG group. There were statistically significant increases in CREA level in SC, SM and SE groups compared to the CG group, and there were statistically significant increases in BUN level in SC, SM and SE groups compared to the CG group ($p<0.05$).

According to Figure 2, there was a statistically significant increase in GR activity level in the SE group compared to the CG group, and in the

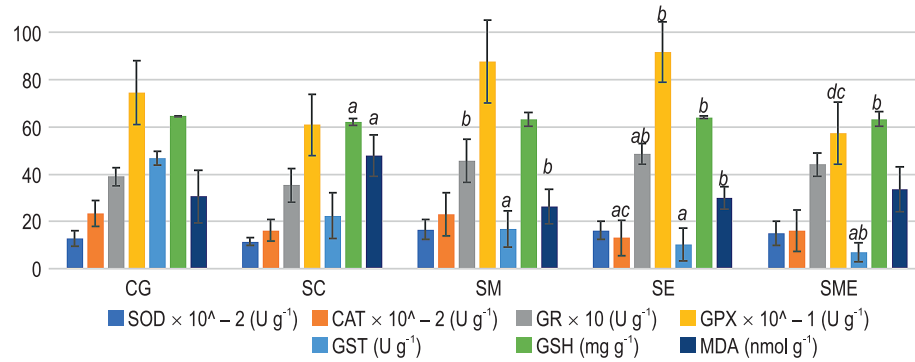

Fig. 2. Oxidant and antioxidant levels in liver tissue: *a* – the difference compared to the CG group is statistically significant ($p<0.05$), *b* – the difference is statistically significant compared to SC group ($p<0.05$), *c* – the difference is statistically significant compared to the SM group ($p<0.05$), *d* – the difference is statistically significant compared to SE group ($p<0.05$)

SM and SE groups compared to the SC group ($p<0.05$). In terms of GPx activity level, the SE group had a statistically significant increase compared to the SC group, while the SME group had a statistically significant decrease compared to SM and SE groups. In terms of GST activity level, SM, SE and SME groups had statistically significant decreases compared to CG group and the SME group had a statistically significant decrease compared to the SC group. In terms of GSH activity level, the SC group showed a statistically significant decrease compared to the CG group, while SE and SME groups showed statistically significant increases compared to SC group. In terms of MDA level, it was found that the SC group had a statistically significant increase compared to the CG group, while SM and SE groups had statistically significant decreases compared to the SC group ($p<0.05$).

According to Figure 3, there was a statistically significant increase in CAT activity level in the SE group compared to the CG group, and in the SE and SME groups compared to the SC group. In terms of GR activity level, the SM group showed a statistically significant increase compared to the SC group, while the SME group showed a statistically significant decrease compared to the SM group. As regards GPx activity level, the SE group had a statistically significant increase compared to both CG and SC groups, while the SME group had a statistically significant decrease compared to the SE group. With respect to MDA level, it was found that the SC group showed a statistically significant increase compared to the CG group ($p<0.05$).

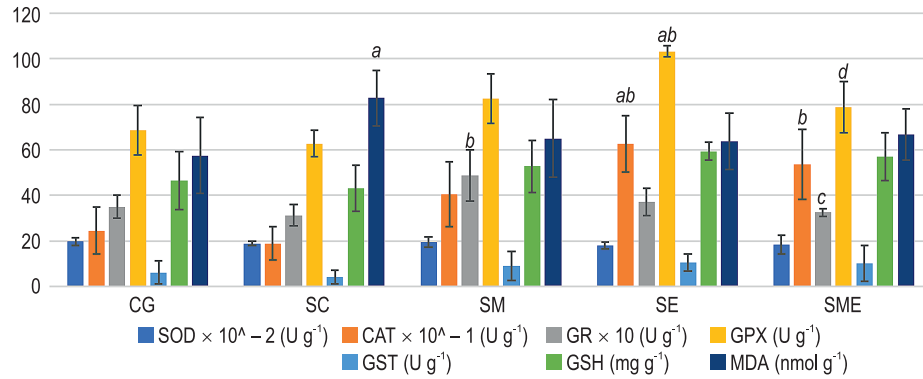

Fig. 3. Oxidant and antioxidant levels in kidney tissue: *a* – the difference compared to the CG group is statistically significant ($p<0.05$), *b* – the difference is statistically significant compared to SC group ($p<0.05$), *c* – the difference is statistically significant compared to the SM group ($p<0.05$), *d* – the difference is statistically significant compared to SE group ($p<0.05$)

Histopathology results

The results of the histopathology examination of liver and kidney tissues were as follows:

CG group: this group was observed to have normal histological appearance (Figures 4-5).

SC group: severe degeneration in hepatocytes, moderate coagulation necrosis, dilatation in sinusoids and severe hyperemia in vessels were observed (Figure 4). Dilatation in the kidneys, Bowman's capsule and tubular lumens, severe degeneration of tubule epithelium, moderate necrosis, and severe hyperemia in the vessels and glomeruli were detected (Figure 5).

SM group: mild degeneration of hepatocytes and moderate hyperemia of vessels were detected (Figure 4). Mild degeneration of kidneys and tubules and moderate hyperemia of vessels were observed (Figure 5). A statistically significant difference ($p<0.05$) was detected when compared to the diabetes group (SC).

SE group: moderate degeneration of hepatocytes, mild necrosis, and hyperemia in vessels were observed (Figure 4). Moderate degeneration of kidneys and tubules, and moderate hyperemia of vessels and glomeruli were observed (Figure 5). A statistically significant difference was found when compared to the diabetes group ($p<0.05$).

SME group: there was very mild degeneration in hepatocytes and mild hyperemia in vessels (Figure 4). Very mild degeneration of kidneys and tubules and mild hyperemia of vessels were observed (Figure 5) and a significant difference ($p<0.05$) was detected. Histopathological findings are summarized in Table 3.

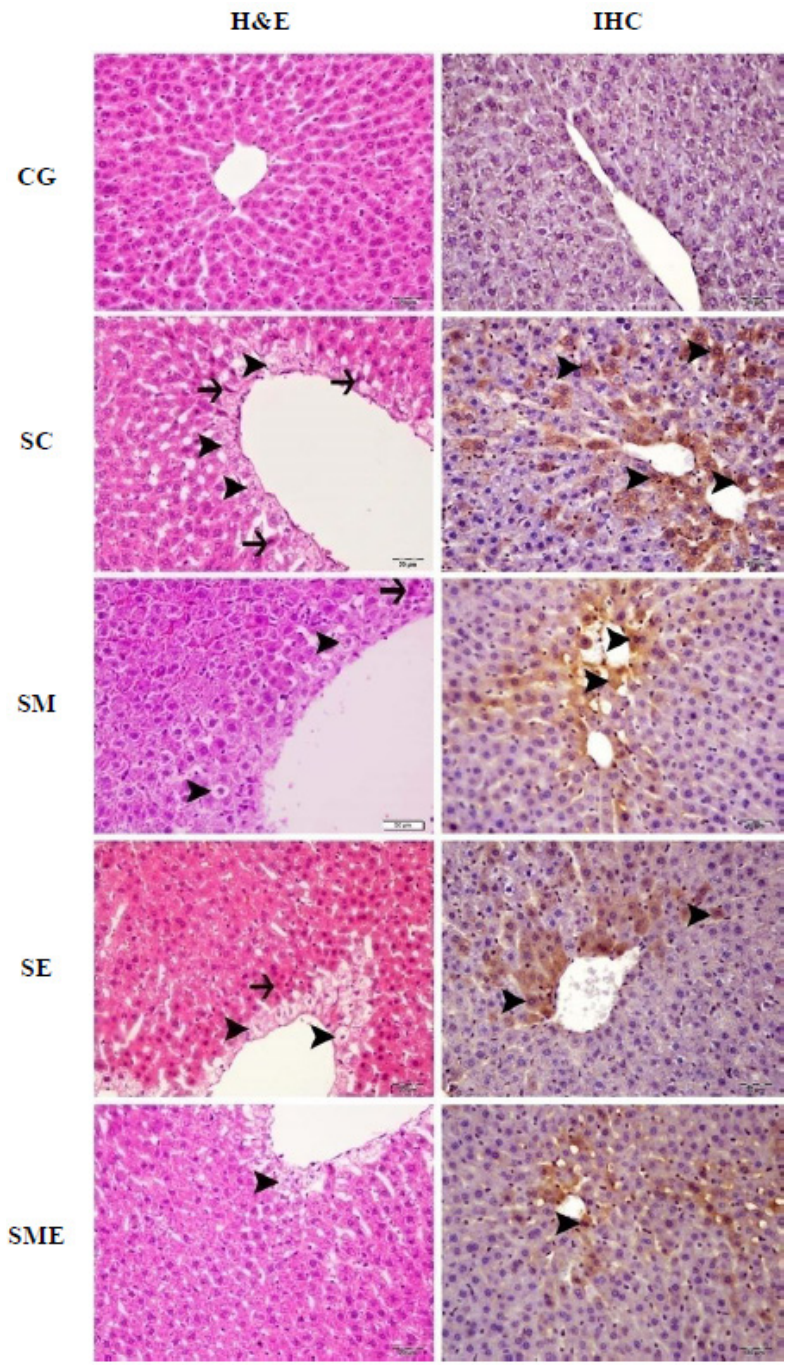

Fig. 4. Liver tissue, hepatocyte degeneration (arrowheads), necrosis (arrows), H&E, hepatocyte expression of 8 OHdG (arrowheads), IHC-P, Bar: 20 µm

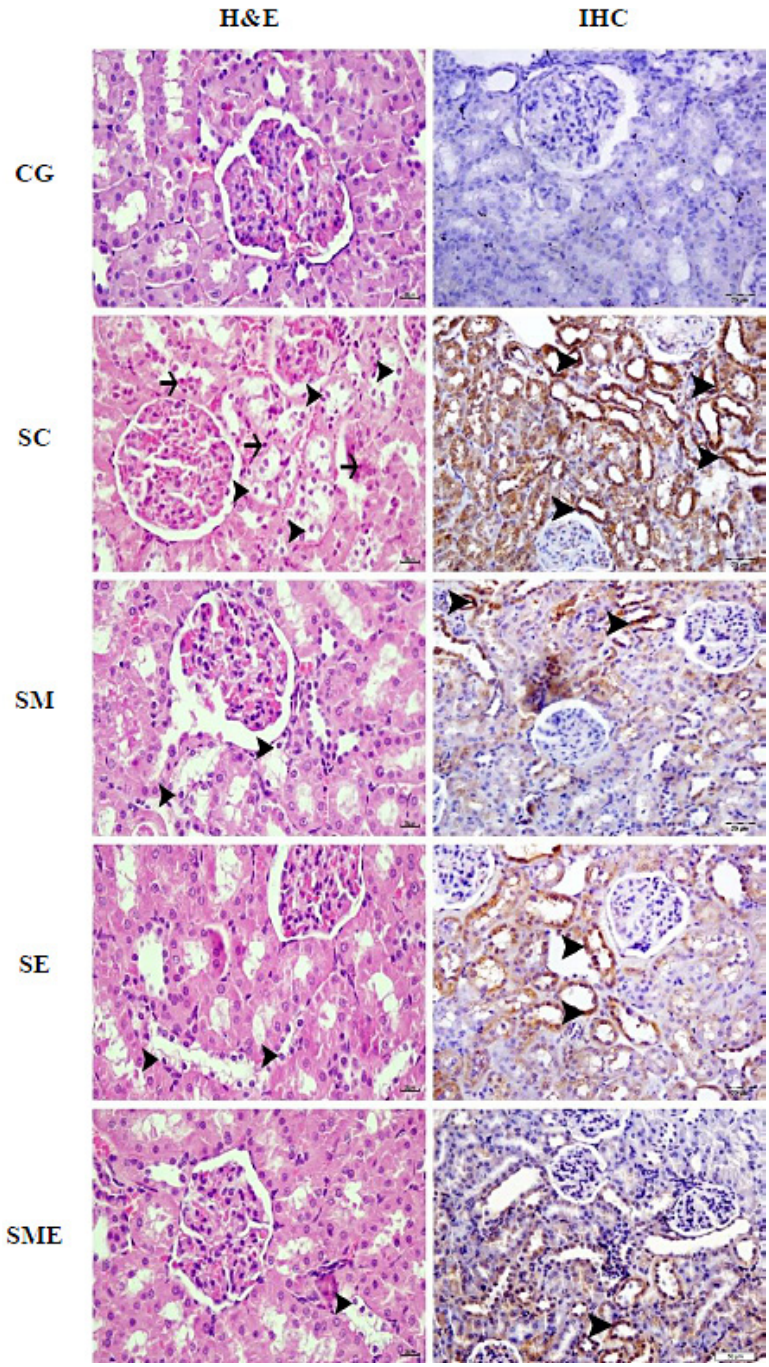

Fig. 5. Kidney tissue, tubulocytellular degeneration (arrowheads), necrosis (arrows), H&E, tubulocytellular endoplasmic 8 OHdG expression (arrowheads), IHC-P, Bar: 20 µm

Table 3
Scoring of histopathological and immunohistochemical findings in liver and kidney tissues

Tissues	Findings	CG	SG	SM	EM	SME
LIVER	Degeneration in hepatocytes	-	++++	++	++	+
	Necrosis in hepatocytes	-	+++	+	+	-
	Hyperemia in the veins	-	++++	+++	+++	++
	8 OHdG expression	-	++++	++	++	+
KIDNEY	Degeneration of tubules epithelium	-	++++	++	++	+
	Necrosis in tubulespithelials	-	+++	+	+	-
	Hyperemia in the veins	-	++++	++	+++	++
	8 OHdG expression	-	++++	++	++	+

Immunohistochemistry results

The results of the immunohistochemical examination of liver and kidney tissues were as follows:

CG group: this group had negative 8 OHdG (8-hydroxy 2 deoxyguanosine) expression (Figures 4-5).

SC group: severe cytoplasmic 8 OHdG expression was observed in hepatocytes (Figure 4). In the kidneys, severe 8 OHdG expression was detected in tubule epithelium (Figure 5).

SM group: mild cytoplasmic 8OHdG expression was detected in hepatocytes (Figure 4). Mild expression of 8 OHdG was detected in kidneys and tubular epithelium (Figure 5). A statistically significant difference ($p<0.05$) was detected when compared to the diabetes group (SC).

SE group: mild cytoplasmic 8 OHdG expression was detected in hepatocytes (Figure 4). Mild expression of 8 OHdG was observed in the kidneys and tubule epithelium (Figure 5). A statistically significant difference ($p<0.05$) was detected when compared with the diabetes group.

SME group: very mild cytoplasmic 8 OHdG expression was detected in hepatocytes (Figure 4). In kidneys, very slight cytoplasmic 8 OHdG expression in tubule epithelium (Figure 5). A statistically significant difference ($p<0.05$) was detected when compared with the diabetes group. Histopathological findings are summarized in Table 3.

DISCUSSION

This study was carried out to determine the single/interactive effects of emodin and metformin on histopathological and oxidative stress parameters in liver and kidney tissues in rats with induced experimental diabetes. The results obtained were discussed with current data.

When the fasting blood glucose levels in our study were analyzed, the blood glucose levels in the SM, SE and SME groups decreased in the 3rd week, but did not decrease to the control group levels. Yıldız et al. (2018) reported that, similarly to our study, gilaburu plant reduced the increased blood glucose value in rats with experimental diabetes, but did not reduce it to normal values.

In some studies on diabetes, it was stated that AST and ALT levels increased in the diabetic group compared to the control group, but serum enzyme levels decreased in the group given plant extracts (Jiyil et al. 2019, Dubey et al. 2020). Our current study is parallel to these studies and it was observed that AST and ALT levels increased in diabetic groups compared to the control group, but in the SME group, the values were close to the control group. Increased creatinine level in the blood is an indicator of kidney damage and impaired glomerular filtration rate is an indicator of glomerular function. Protein and creatinine are metabolically broken down and are among the waste products excreted by the kidney. In kidney damage, these substances accumulate in the body along with other substances, leading to increased creatinine and BUN levels (Dubey et al. 2020). Jivil et al. (2019) stated that creatinine and BUN levels increased in diabetic groups compared to the control group, but decreased compared to the diabetes group in the groups given plant extract. In parallel with our study, it was observed that creatinine and BUN levels increased in the diabetes groups compared to the control group, especially in the SME group, and were closer to the control group. In STZ-induced diabetic rats, it can be said that the combination of metformin + emodin has more favorable effects than single administrations of these substances.

Oxidative stress can be prevented by an enzymatic system responsible for eliminating the increase in reactive oxygen species, such as superoxide radical, hydroxyl radical and hydrogen peroxide, that are formed during cellular metabolism. The enzymatic system converts hydrogen peroxide into water and oxygen using CAT and GPx (Perestrelo et al. 2022). In a study by Seedevi et al. (2020), the MDA level decreased in the group treated with crude polysaccharide and rhamnose-enriched polysaccharide derived from *G. lithophila*, and SOD and GPx activities were close to the controls. In another study, it was reported that there was an increase in MDA levels in liver and kidney tissue in STZ-induced diabetic rats, but a decrease in SOD, CAT, GR and GPx activities (Li 2007). The present study is similar to the study by Li (2007) and it was observed that the MDA level increased

in the STZ-induced diabetes group compared to the controls, while antioxidant parameter activities decreased. This suggests that oxidative stress has an important role in diabetes complications and oxidative stress caused by the hyperglycemic state leads to decreases in antioxidant and pro-oxidant (MDA) parameters (AlFaris et al. 2020, Ajayi et al. 2022).

It was reported that the levels of biomarkers related to oxidative damage in kidney and liver tissues of diabetic rats treated with metformin decreased (Karise et al. 2017, Corremans et al. 2018). Similar to the present study, in one of the studies in which the combination of metformin was used with different substances, Yazdi et al. (2020) investigated the effect of *Artemisia turanica* (AT) and metformin administration in diabetic rats against oxidative stress in tissues. It was stated that AT application alone or in combination with metformin had a more positive effect than metformin alone in increasing the SOD and CAT activity of the plant extract. Similarly, in another study, lycopene and metformin were administered alone or in combination. In this study, unlike lycopene, metformin alone did not increase SOD activity in liver and kidney tissues, but increased CAT activity. It was stated that the improvement in CAT activity may be sufficient to reduce oxidative stress in the tissues of diabetic rats treated with metformin (Figueiredo et al. 2020). Alhaider et al. (2011) reported that metformin may protect against nephropathy in STZ-induced diabetic rats.

Beheshti et al. (2022) reported that chronic administration of low doses of metformin decreased MDA levels in liver tissue, increased SOD and CAT activities, and protected against lipopolysaccharide (LPS)-induced toxicity. In addition, Wu et al. (2021) reported a decrease in SOD and CAT activities and an increase in MDA levels in liver tissue of rats with a high-cholesterol diet (HCD). However, it was reported that MDA levels decreased in the groups treated with different doses of emodin compared to the HCD group. Low-dose emodin (10 mg kg^{-1}) significantly increased hepatic SOD activity, but treatment with higher doses of emodin (30 and 100 mg kg^{-1}) inhibited hepatic SOD and CAT activity to a great extent. The present study is consistent with the literature and a significant increase in antioxidant parameter activities was observed with emodin, metformin and the emodin + metformin combination compared to the diabetes group. We believe that this may indicate a hypoglycemic effect. Emodin's ability to scavenge free radicals, reduce MDA levels in liver and serum, and improve the activities of enzymatic antioxidants such as SOD (in liver and serum) and CAT (in liver and serum) may be associated with its antioxidant activity (Wang et al. 2021, Wu et al. 2021, Zhao et al. 2021). In this case, antioxidant enzymes especially were reported to be an important factor in cellular defense against oxidative damage in the liver and kidney tissues of rats with STZ-induced experimental diabetes (AlFaris et al. 2020). In addition, thanks to the antioxidant effect of emodin and the hypoglycemic property of metformin, positive effects were observed in biochemical parameter values with the combination of these two

agents.

In a study by Balamash et al. (2018), histopathological examination of liver tissue of diabetic control group identified pathological changes indicating degenerative changes, such as the presence of shrunken hepatocytes with numerous dark spots and small dark nuclei (apoptosis). However, histopathological results from rats showed that metformin, olive oil and the olive oil + metformin combination restored the normal structural organization of the liver tissue in diabetic rats. The present study is in accordance with the previous study; it was determined that especially the emodin + metformin combination caused degeneration, necrosis, vascular hyperemia and 8 OHdG expression in hepatocytes close to the control group.

In studies on kidney tissue, it was reported that glomerular sclerosis and severe destruction were found in the kidneys of diabetic rats with histopathological evaluation (Gopal et al. 2013). In addition, there was atrophy of glomerular capillaries and deformity of renal corpuscles in the kidney tissue of the diabetic group (Balamash et al. 2018). In the present study, when the values for the emodin + metformin combination, one of the treatment groups, were examined in kidney tissue, degeneration of tubule epithelium, necrosis in tubule epithelium, hyperemia in vessels and 8 OHdG expression were close to the control group. According to these findings, we suggest that the emodin + metformin combination may be useful as a preventive measure to reduce the risk of developing diabetes.

CONCLUSIONS

In this study, it can be concluded that both separate and combined administration of emodin and metformin had positive effects on liver and kidney tissue in rats with experimental diabetes induced. For this reason, we consider that emodin may be useful in the treatment of diabetes and related complications in addition to metformin. However, studies using different doses and durations are required to find an effective therapeutic agent for the prevention and treatment of DM-related complications.

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