Plant Archives Volume 20 No. 1, 2020 pp. 2251-2257 **e-ISSN:2581-6063 (online), ISSN:0972-5210**

ANTIOXIDANT EFFICIENCY OF BERBERINE ON OXYTETRACYCLINE INDUCED RENAL TOXICITY IN RATS

Zena Shakir M. AL-Tamemi[1], Aryaf Mahmood Sabea[1], Gusson A.AL-Neamah[2], Jawad Kadhim Faris[1]*, Auday Sulaiman Khudhair[3] and Manal Muhi Murad[1]

[1*]Department of Physiology and Pharmacology, College of Veterinary Medicine, Al-Qasim Green University, Iraq.
[2]Department of Pathology, College of Veterinary Medicine, Al-Qasim Green University, Iraq.
[3]Department of Public health, College of Veterinary Medicine, Al-Qasim Green University, Iraq.

Abstract

Plants have been the foundation of many classical medicines throughout the world for since long time ago and continue to supply new remedies to humanity. This study was conducted to find out renoprotective activity of berberine (BBR) 50,100mg/kg body weight (b.w.) against oxytetracycline (OXT) 200 mg/kg induced renal toxicity in rats. Twenty eight, male rats were divided into four groups, group1: control, (1 ml/kg Saline orally) group 2: OXT (200 mg/kg), intraperitoneally (i.p.) for (7) consecutive days , group 3: OXT (200 mg/kg), i.p. plus BBR (50 mg//kg) orally for (7) consecutive days, group 4: OXT (200 mg/kg), i.p. plus BBR (100 mg//kg) orally for (7) consecutive days. At the end of the experiment, the blood, and kidney samples were taken. Kidneys was examined by Hematoxylin Eosin (HE) staining. Renal function test such as blood urea nitrogen (BUN), serum creatinine (SC), examination were determined. The levels of malondialdehyde (MDA), in serum were determined. Finally immunofluorescence were performed for measuring TNF-a in kidney. OXT induced renal damage was proved by a significant ($p \geq 0.01$) reduction in the body weight,and a significant ($p \leq 0.01$) increased serum BUN, SC , MAD, histopathological and immunohistochemical changes. BBR protective renal toxicity effect and oxidative damage caused by OXT significantly ($p \leq 0.01$) increasing in body weight and significantly ($p \geq 0.01$) decreasing BUN, SC , MAD and improving tissue morphology, in BBR (50 mg//kg) while, BBR (100 mg//kg) has more effects. These results confirm that BBR (100 mg//kg) antioxidant effects can protect OXT -induced renal toxicity in rats

Key words : Berberine , Oxytetracycline , Anti-oxidants , Renal toxicity.

Introduction

Nephrotoxicity is the adverse effect of substances on renal function (Perazella, 2009) These substances can involve molds and fungi, cancer therapeutics such as cisplatin, antibiotics, metals as mercury, arsenic, lead, and drugs of abuse as cocaine (Zhou *et al.*,2008). OXT is a type of antibiotic called a tetracycline. It is commonly used antibiotic for the treatment of Anthrax, Chlamydia, Cholera, Lyme disease, Typhus, Relapsing Fever, Tularaemia, Malaria, Plaque, Syphilis, Respiratory infection, Mycoplasma, Rickettsiae, Streptococcal infection and Acne. High doses of OXT is generally regarded as toxic, they produce a fairly large number of adverse effects, some of which can be life threatening (Saraswat *et al.*, 1997). OXT is a broad-spectrum antibiotic, with bacteriostatic activity against gram positive and gram negative bacteria, including some anaerobes in livestock, poultry and aquatic animals, that is widely used in controlling respiratory diseases and other genital and skin infections in human and animals. Large doses of OXT without medical supervision have deleterious effects on kidney and liver (Abdel Daim - Ghazy *et al.*, 2015; Dale - Mandelstam *et al.*, 2005). BBR is an isoquinoline alkaloid that is mainly isolated from coptidis rhizome (Derosa - Maffioli *et al.*,2014). This compound has a wide range of biological activities such as antioxidant, anti-inflammatory, antimicrobial, anticancer, anti-hypertensive, renoprotective and anti-hyperglycemic effects (Tang *et al.*, 2009; Javad-Mousavi *et al.*, 2016)). BBR has a protective effect against free radicals by

***Author for correspondence :** E-mail: jawadfaris1958@gmail.com

acting as a scavenger for reactive oxygen species (ROS) and reactive nitrogen species (RNS), (Tillhon, *et al.,* 2012). Moreover, BBR increases the activity of antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione (GSH) (Chen *et al.,* 2016). Furthermore, it has been shown that BBR decreases the oxidative stress parameters such as protein carbonyl (PC) content, MDA activity, nitric oxide (NO) level and myeloperoxidase (MPO) activity (Adil *et al.*,2016). It was demonstrated that BBR has hepatoprotective effect in liver ischemia/ reperfusion injury, doxorubicin and tetrachloride-induced acute hepatotoxicity (Lin *et al.,* 2017; Feng *et al.,* 2010).

Materials and Methods

This study was conducted at the period September 2019 in physiology and Pharmacology department of veterinary medicine of AL-Qassim green university.

Drugs

Berberine HCL (BBR) 100% Natural was purchased from Bulk supplement.com USA. Oxytetracycline (OXT) , 200 mg/ml, as ALamycin LA was purchased from Norbrook, IRELAND. Ketamine 10% inj. from Kepro-Holland. Xylazine, XYL- M2, VMD- Belgium.

Experimental rats

The number of laboratory animals used in the experiment are Twenty eight healthy male rats at aged (180) days and weighted (200-210) grams, obtained from the animal house of the College of Veterinary Medicine, AL-Qadisiya University, were kept for one (10) days as acclimatization period before the starting of the experiment, all rats were feed on concentrated food (pellets) and were given plain water, the animals room temperature was (19-23)°C, and the humidity was (45 - 55), that room was washing and sterilization once a week.

Experimental design

After a quarantine period of (10) days, twenty eight rats were randomly divided into four equal groups, each group consist of (7) rats, and they received the treatment as follows:

Group I: Control (1 ml/kg Saline orally) for (7) days.

Group II: OXT (200 mg/kg, i/p) for (7) days (Samah *et al.*,2018; Saraswat *et al.,* 1997).

Group III: OXT (200 mg/kg, i/p) + BBR (50 mg/kg orally by stomach tube) for (7) days. (Lotfi *et al.*,2018).

Group IV: OXT (200 mg/kg i/p) + BBR (100 mg/kg orally by stomach tube) for (7) days. (Saeed *et al.*,2018).

Body weight

All animals were weighed before and after treatment with using digital electronic balance.

Serum Preparation

At the end of experimental period, rats were fasted for (10) hrs, anaesthetized with ketamine (75 mg/kg) combined with xylazine (2.5 mg/kg). (Molina *et al.*,2015). Blood samples were collected by heart puncture in non-heparinized tubes, centrifuged at (4000) rpm for (10) minutes (Laessig *et al.*,1976). After separation the serum from the clot, using a sampler, the samples were used to measurement of BUN, SC and MDA level concentration.. The rats were sacrificed by cervical dislocation and the abdominal cavity was immediately opened, kidneys were removed and processed for histopathological and immunohistochemical studies.

Histopathological techniques

Sections were taken from kidneys tissues from different animals in each group immediately after sacrificed. These tissues were washed with the normal saline solution to remove blood, then fixed in 10% neutral formalin for (24) hrs, dehydrated in different concentration of alcohol, and processed for paraffin embedding. Sections of (5) µm thickness were cut using a rotary microtome. The sections were processed and passed through graded alcohol series stained with Haematoxylin and Eosin, cleared in xylene and examined microscopically according to (Bancroft *et al.,* 1996).

Immunohistochemical (IHC)

Staining was applied to assess the activity of tumor necrotic factor –alpha (TNF-α). IHC was performed on 5-ìm tissue sections. Briefly, 20 min were spent to incubate the tissue sections at 50 ° C, followed by its drying with a downward series of alcohol, and exposing with 1% hydrogen peroxide in distilled water for 5 minutes in a dark environment. To decrease the activity of endogenous peroxidase, after washing the antigens in phosphate-buffered saline (PBS) (pH=7.4), they were retrieved through autoclaving for 30 min in citrate buffer (C6 H5 Na3O7·2H2 O, pH=6). Thereafter, the sections were incubated with primary antibody (ab13847) for the whole night at temperature of 4°C. Optimum dilution was reported to be 1/300. In the next stage, the tissue sections were incubated in the goat polyclonal secondary antibody (HRP) (ab97051) for one hour at room temperature by adding 3, 32 - diaminobenzidine (DAB, Dako) to obtain an image of the antigen. Finally, hematoxylin (Sigma) was added to slightly counterstain tissue sections, and the sections were then dried in alcohol, cleansed in xylene (Sigma), and mounted for imagining. The primary antibody

was not used in the negative control slide's processing. Based on the data sheet, tissue of rat lung was applied as a positive control (Chen *et al.*,2016).

Statistical analysis

The statistical results of the data were analyzed according to Complete Randomized Design (C.R.D.) (AL- Rawi *et al.*,2000). The mean differences between the averages of the studied traits were determined at the probability level of (0.01) using the Duncan test (Duncan,1995). Statistical data were analyzed using the (SAS,2010).

Results

Body weight

No mortality were observed in the groups of rats that were given OXT either alone or in summation with BBR, with reduced appetite, decreased activity and progressive physical fatigue were observed in the rats from the OXT group. I/P injection of OXT produced significant ($P \geq 0.01$) decrease in the body weight compared to control. The animals were weighed before and after the experiments, whereas, treatment with BBR (100 mg/kg) produced significant ($P \leq 0.01$) improved than BRB (50 mg/kg) on the body weight compared to OXT control rats (Table 1).

Effect of BRB on OXT induced alterations in renal function parameters

Effect of OXT induced reduction in renal function in rats. A significant ($p \leq 0.01$) elevation in serum BUN, SC levels compared to the control group and significant ($p \leq 0.01$) elevation in MDA levels compared to control was observed after (7) days of treatment with OXT Whereas, treatment with BRB (50 mg/kg) prevented OXT induced significant ($p \geq 0.01$) reduction in serum BUN, SC levels and produced significant ($p \geq 0.01$) reduction on the MDA compared to OXT control rats. However, BRB (100 mg/kg) it has more effect than, BRB (50 mg/kg) on serum BUN, SC levels and MDA level compared to OXT control rats (Table 2).

Effect of BBR on OXT induced histopathological alteration in renal rat tissue

Light microscopic of kidney examination using H&E (400X) stain in control rats shown no clear lesion in (Fig.1a). Histopathological effects of OXT on kidney of treated rats are presented in rats treated with OXT for (7) days shown congestion of the capillary blood vessels within the glomerular tufts with dilation of bowman's space and deposition of round eosinophilic pretentious materials, also sloughing of the renal tubular epithelia with hydropic changes seen in (Fig.1b). Kidney rats treatment with OXT and BBR (50 mg/kg) shown histopathological section of kidney shown mild regeneration in some tubular epithelia in (Fig.1c). Kidney rats treatment with OXT and BBR (100 mg/kg) shown regeneration of tubular and glomerular epithelia in (Fig.1d).

Effect of BBR on OXT induced immunohistochemical alteration in renal rat tissue

Examination of tumer necrotic factor –alpha (TNF-α) by immunohistochemical staining (magnification×200) in kidney sections represented as (Fig. 2a) normal control group (Fig. 2b) OXT (200 mg/kg) group shown strong cytoplasmic staining within epithelial cells lining the renal tubules and glomerular tufts (Fig. 2c) OXT (200 mg/kg) plus (50mg/kg) BBR group shown moderate cytoplasmic staining within epithelial cells lining the renal tubules and glomerular tufts (Fig. 2d) OXT (200 mg/kg) plus (100

Table 1: Effect of Berberine on Oxytetrcycline-induced change of the body weight /gram of rats.

Oxytetracycline +Berberine 100 Mean ± SE	Oxytetracycline + Berberine 50 Mean ±SE	Oxytetracycline Mean ± SE	Control Mean ± SE	Traits
7	7	7	7	No. of rats
202.40 ± 0.02 A	202.41 ± 0.01 A	202.41 ± 0.02 A	202.45 ± 0.06 A	Weight at 1 day (g/animal) Ns
188.59 ± 2.57 B	178.49 ± 2.35 C	165.22 ± 1.84 D	212.66. ± 112 A	Weight at 8 day (g/animal) **

NS: Non significant. significant difference at 0.05. ** high significant difference at 0.01.

Table 2: Effect of Berberine on Oxytetracycline-induced change in kidney function of rats.

Oxytetracycline +Berberine 100 Mean ± SE	Oxytetracycline + Berberine 50 Mean ±SE	Oxytetracycline Mean ± SE	Control Mean ± SE	Traits
0.76 ± 0.01 C	0.87 ± 0.02 B	1.29 ± 0.01 A	0.62 ± 0.01 D	Serum creatinine (mg/dl) **
50.34 ± 1.07 C	63.40 ± 1.06 B	74.04 ± 1.33 A	41.59 ± 0.90 D	Blood urea nitrogen(mg/dl) **
0.79 ± 0.02 C	1.20 ± 0.01 B	2.45 ± 0.10 A	0.62 ± 0.01 D	Malonaldehyde(nMole/L) **

NS: Non significant. significant difference at 0.05. ** high significant difference at 0.01.

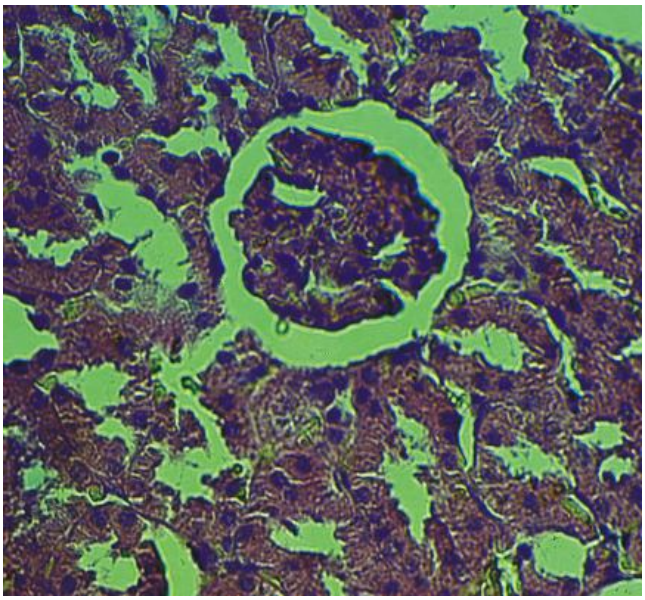

Fig. 1a: H&Ex400

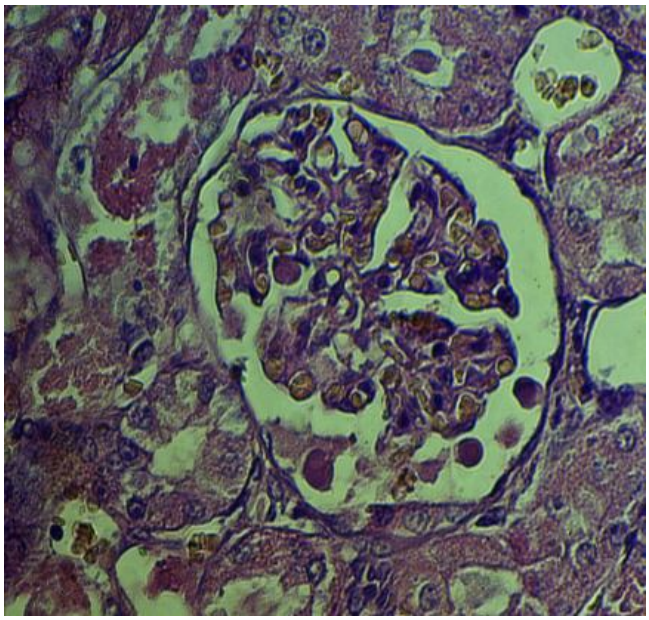

Fig. 1b: H&Ex400

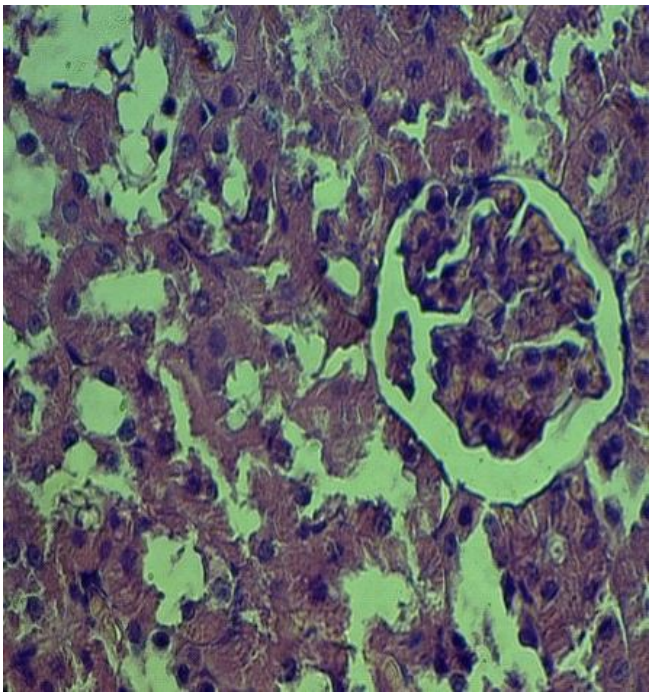

Fig.1c: H&Ex400

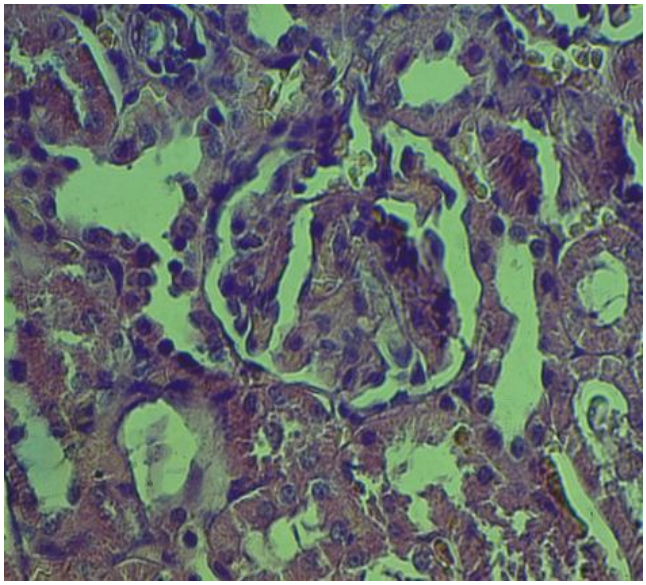

Fig. 1d: H&Ex400

mg/kg) BBR group shown mild cytoplasmic staining within epithelial cells lining the renal tubules and glomerular tufts.

Discussion

The kidney is an important organ for the excretion of xenobiotics and their metabolites and is especially susceptible to damage because of a larger perfusion and an increased concentration of excreted compounds that occur in renal tubular cell. (Mohamed *et al.,* 2003). An earlier study reported that the high doses of OXT could produce renal failure.(Tarara *et al.,* 1976). It is well documented that OXT inhibits the incorporation of amino acid into protein, causing an increase in urea level. It also increases the urinary sodium excretion and may induce vomiting and diarrhea, thus further compromising renal perfusion.(Davies, 1991). OXT, widely used to cure many of the human and animal diseases. In developing countries, OXT is often used for extended periods at overdose, without supervision, which might lead to unexpected toxicological problems, some with apparent clinical symptoms, gastrointestinal upset as diarrhea, while others are, hidden symptoms such as immunosuppressant, for ordinary breeders this leads to severe economic losses (Southwood, 2006). In this study, the effect of BBR on

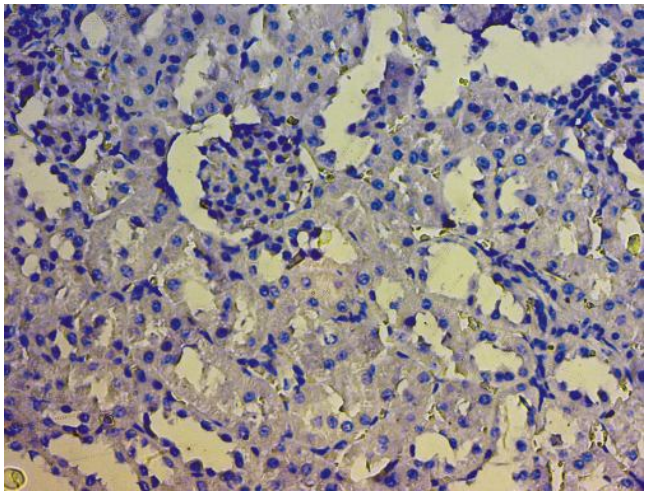

Fig. 2a: Immunohistochamical x200

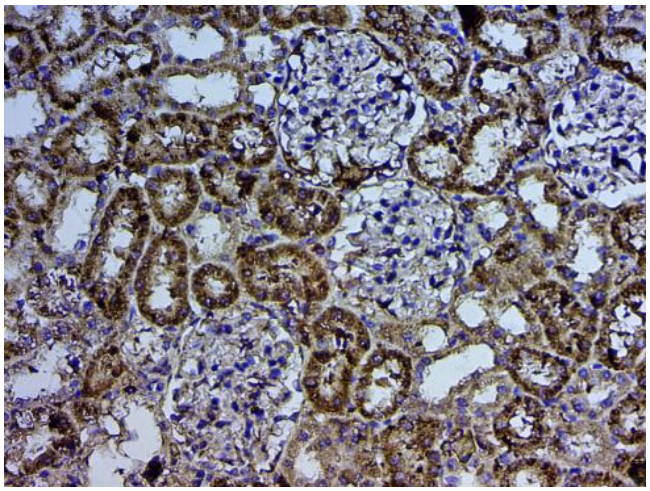

Fig. 2b: Immunohistochamical x200

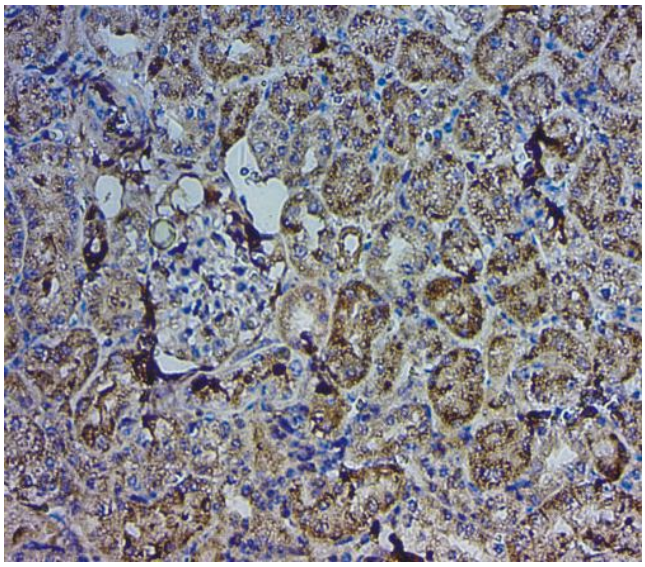

Fig. 2c: Immunohistochamical x200

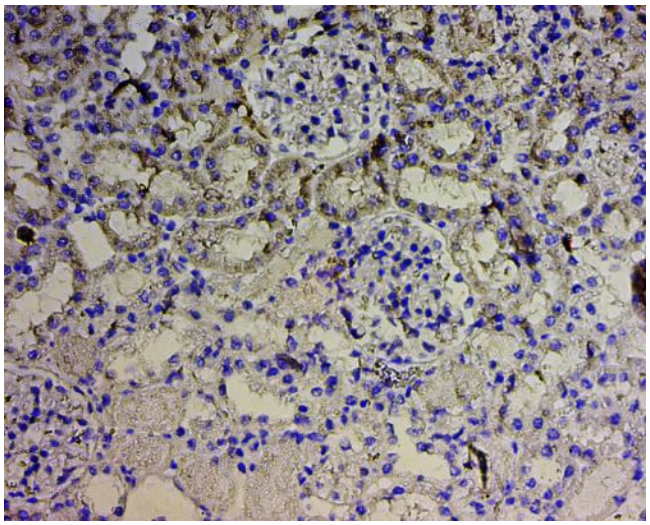

Fig. 2d: Immunohistochamical x200

OXT- induced nephrotoxicity in rats was inspected. Results of this study indicated that, i/p injection of (200 mg/kg) of OXT for (7) days, causes significant nephrotoxicity as appear by produced significant ($p \leq 0.01$) reduction in the body weight, when compared to control. Our results are in acceptance with previous findings of (Samah *et al.*,2018). According to (Erdem *et al.*, 2000), increased catabolism and anorexia are responsible for decreased food intake and causes body weight loss further, following loss of the tubular cells, involved in renal water reabsorption leads to dehydration and decreases body weight (El-Zawahry *et al.*,2007). In the present study, the administration of OXT for (7) days produced a significant ($p \leq 0.01$) increase of serum BUN and SC levels. These results are in acceptance with those gained by other investigators (Samah *et al.*, 2018; Gnanasoundari - Leelavinothan *et al.*, 2006; Abdel Daim - Ghazy *et al.*, 2015). In the present study, increased serum BUN and SC in OXT-treated rats reflect the renal damage (Jayanthi - Subash *et al.*, 2010). In the present study, renal injuries caused by OXT might be refer to the oxidative stress resulting from excessive free radical production (Naseer - Alam *et al.*, 1987). We found that OXT treatment caused increased MDA levels, these results are in acceptance with those obtained by other investigators **(**Abdel Daim - Ghazy *et al.*, 2015; Naseer - Alam *et al.*, 1987; Yonar *et al.*, 2012). The formed free radicals seemed to initiate lipid peroxidation in OXT treated rats suggesting that, the increase in lipid peroxidative index as evaluated in term of plasma MDA might be associated with cellular damage (El Sayed *et al.*,2014). Histopathological changes, induced by, i/p injection of (200 mg/kg) of OXT for (7) days, which add with clearly elevated levels of renal biochemical markers BUN, SC and MDA activities. Similar findings were noticed by (Abdel Daim - Ghazy *et al.*, 2015; Samah *et al., 2018*; Tarara *et al., 2*1976). The results of the present study

suggest that, the accumulation of free radicals, and that increased oxidative stress is a basis for cellular damage. The current study showed that OXT increases the renal TNFα and the relative to all rat body weight and kidney enzymes. According to the results obtained from our research, we realized that the renal tissue inflammation, the serum level of the pro-inflammatory cytokine TNFα, which is one of the most important cytokines released during kidney damage. Treatment with BBR for (7) days reduced the OXT injured kidney produces, a significant increased in body weight and significant decrease in serum BUN, SC , levels and significant decreased in MDA levels. Histopathological changes induced by OXT were modulation the kidney damages, through regeneration of tubular and glomerular epithelia. In immunohistochemical shown the ability of BBR to suppress the pro-inflammatory cytokine TNFα, which is one of the most important cytokines released during kidney damages. Our results demonstrate the ameliorative effect BBR (100 mg/kg) on OXT for (7) days induced kidney toxicity in the rats. This can be explained on the anti-inflammatory (Zhou - Mineshita *et al.,* 2000**)**, analgesic, antinociceptive and antipyretic activity (Esra *et al.,* 2002), anti microbial (Iwasa *et al.,* 1998). Also effective antioxidant and free radical scavenger that prevents ROS formation and exerts protective effects on cardiac, hepatic and renal functions (Lee *et al.,* 2010). As for BBR (50 mg/kg), it had less effect than (100 mg/kg). Based on the advance findings, it can be conclude that, OXT had adverse effects on the kidney. BBR (100 mg/kg) administration showed a marked renoprotective activity. The protective effects of BBR (100 mg/kg) may be due to the its anti-inflammatory effects or antioxidant effects or antimicrobial effects, individually or synergistically. .

Conclusion

This study explained that BBR declines OXT induced nephrotoxicity. The effect of BBR against OXT induced nephrotoxicity could be mediated through its anti-inflammatory, antimicrobial, antioxidant,, and anti-apoptosis action.. However, BBR treatment was able to alleviate renal damage associated with OXT treatment and this is assign to its the antioxidant activity and its ability to prevent inflammation.

References

Abdel-Daim, M.M. and E.W. Ghazy (2015). Effects of *Nigella sativa* oil and ascorbic acid against oxytetracycline-induced hepato-renal toxicity in rabbits. *Iran J. Basic Med., Sci.*, **18:** 221-227.

AL- Rawi, K.M. and M.K. Abdul-Aziz (2000). Design and Analysis of Agriculture Experiments. Dar AL-Kutob press for printing and publishing, Mosul University.

Bancroft, D., A. Stevens and R. Turmer (1996). Theory and practice of histological technique, 4th ed., Churchill Living Stone, Edinburgh, London Melbourne, 47- 67.

Chen, W., S. Wei, Y. Yu, H. Xue, F. Yao and M. Zhang (2016). Pretreatment of rats with increased bioavailable berberine attenuates cerebral ischemia-reperfusion injury via down regulation of adenosine-52 monophosphate kinase activity. *Eur. J. Pharmacol*., **779:** 80-90.

Dale, M.M. and J. Mandelstam (2005). Antiobiotics. Microsoft Encarta Online Encyclopedia.

Davies, D.M. (1991). Textbook of Adverse Drug Reactions. 4th ed. Oxford: Oxford Medical Publications.

Duncan, C.B.(1995). Multiple range and multiple (F) test. *Biometrics*, **11:** 1-12.

El Sayed, A.S., A.M. Badawi and M.T. Asmaa (2014). The Protective effect of Olive Leaf and Pomegranate Peel Extracts On Oxidative Stress And Liver Injury Induced By Oxytetracycline In Albino Rats. *J. Drug Res. Egypt*, **35(1)**.

El-Zawahry, B.H. and E.M. Abu El Kheir (2007). The Protective Effect of Curcumin Against Gentamicin-Induced Renal Dysfunction and Oxidative Stress in Male Albino Rats. *The Egyptian J. Hosp. Med*., **29:** 546-56.

Erdem, A., N. Gundogan, U. Alp and A. Kara (2000). The protective effect of taurin against GM induced acute tubular necrosis in rats. *Nephrol Dial Transplant*, **15:** 1175-82.

Esra, K., K. Müberra, Y.K. Erdem and C.B. Hüsnü (2002). A comparative study on the anti- inflammatory, antinociceptive and antipyretic effects of isoquinoline alkaloids from the roots of Turkish Berberis species. *Life Sci.*, **72(6):** 645-657.

Iwasa, K., D.U. Lee, S.I. Kang and W. Wiegrebe (1998). Antimicrobial activity of 8-alkyl- and 8-phenyl-substituted berberines and their 12-bromoderivatives. *J. Nat. Proc*., **61:** 1150-1153

Derosa, G. and P. Maffioli (2014). Alkaloids in the nature: pharmacological applications in clinical practice of berberine and mate tea. *Curr. Top. Med. Chem*., **14(2):** 200–206.

Gnanasoundari and P. Leelavinothan (2006). Impact of Naringenin on Oxytetracycline-Mediated Oxidative Damage in Kidney of Rats Antioxidant Effect of Naringenin Muthurangam. *Renal Failure*, **28:** 599–605.

Jayanthi, R. and P. Subash (2010). Antioxidant effect of caffeic Acid on oxytetracycline induced lipid peroxidation in albino rats. *Indian J. Clin. Biochem*., **25:** 371-375.

Laessig, R.H., J.O. Westgard and R.N. Carey (1976). Assessment of aserum separator device for obtaining serum specimens for clinical analysis. *Clin.Chem*., **22:** 235-239.

Lee, I.A., Y.J. Hyun and D.H. Kim (2010). Berberine ameliorates TNBS induced colitis by inhibiting lipid peroxidation, enterobacterial growth and NF êB activation. *Eur. J.*

Pharmacol., **648:** 162-170.

Lotfi Aski, M., M.H. Rezvani, M. Khaksari, Z. Hafizi, Z. Pirmoradi, S. Niknazar and F. Zare Mehrjerdi (2018). Neuroprotective effect of berberine chloride on cognitive impairment and hippocampal damage in experimental model of vascular dementia. *Iran J. Basic Med. Sci*., **21:** 53-58.

Tang, J., Y. Feng, S. Tsao, N. Wang, R. Curtain and Y. Wang (2009). Berberine and Coptidis rhizoma as novel antineoplastic agents: a review of traditional use and biomedical investigations. *J. Ethnopharmacol*., **126(1):** 5–17.

Adil, M., A.D. Kandhare, G. Dalvi, P. Ghosh, S. Venkata, K.S. Raygude and S.L. Bodhankar (2016). Ameliorative effect of berberine against gentamicin-induced nephrotoxicity in rats via attenuation of oxidative stress, inflammation, apoptosis and mitochondrial dysfunction. *Ren. Fail.*, **38 (6):** 996–1006.

Mohamed,M., M.D. Abdel Latif, A. Sahar and M.D. Elglammal (2003). Sodium fluoride ion and renal function after prolonged sevoflurane or isoflurane anesthesia. *Eng. J. Anesth*., **19:** 79–83.

Molina, A.M., M.R. Moyano, J.M. Serrano-Rodriguez, N. Ayala, A.J. Lora and J.M. Serrano-Caballero (2015). Analyses of anaesthesia with ketamine combined with different sedatives in rats. *Veterinarni Medicina*, **60(7):** 368–375.

Tillhon, M., L.M. Guaman Ortiz, P. Lombardi and A.I. Scovassi (2012). Berberine: new perspectives for old remedies. *Biochem. Pharmacol*., **84(10):** 1260–1267.

Naseer, F. and M. Alam (1987). The protective effect of ascorbic acid onoxytetracycline induced nephrotoxicity and hepatotoxicity. *J. Pak. Med. Assoc.*, **37:** 73-75.

Perazella, M. (2009). Renal vulnerability to drug toxicity. *Clin. J. Am. Soc. Nephrol.*, **4:** 1275–1283.

Tarara, R.P., L.G. Hansen and J. Simon (1976). Effects of repeated administration of oxytetracycline on kidney, liver, and liver mixed-function oxidases in the rat. *Toxicology and Applied Pharmacology*, **35(2):** 321- 33.

Saeed Mehrzadia, L., C.L. Iman Fatemib, M. Esmaeilizadehd, H. Ghaznavie, G. Had Kalantarf and M. Goudarzif (2018). Hepatoprotective effect of berberine against methotrexate methotrexate induced liver toxicity in rats. *Biomedicine & Pharmacotherapy*, **97:** 233–239.

Javad-Mousavi, S.A., A.A. Hemmati, S. Mehrzadi, A. Hosseinzadeh, G. Houshmand, M.R.R. Nooshabadi, M. Mehrabani and M. Goudarzi (2016). Protective effect of Berberis vulgaris fruit extract against Paraquat-inducedpulmonary fibrosis in rats. *Biomed. Pharmacother*., **81:** 329–3

Samah, S.O. da, R.S. Waheeb and Z. Kh. El-Maddawy (2018). Potential efficacy of Coenzyme Q10 against oxytetracycline-induced hepatorenal and reproductive toxicity in male rats. *Journal of Applied Pharmaceutical Science*, **8(01):** 098-107 .

Saraswat, B., P.K.S. Viseu, G.K. Patnaik and B.N. Dhawan (1997). Protective effects of picroliv active constituent of picroorhiza kurrooa against oxytetracycline induced hepatic damage. *Indian J. Exp. Biol*., **35:** 1302–5

SAS, (2010). Statistical Analysis System. SAS institute inc. Virgin 7.12 Tsozo, North Carolina state University of Cary, NC, USA.

Southwood, L.L. (2006). Principles of antimicrobial therapy: what should we be using? *Vet. Clin. North Am. Equine Pract*., **22:** 279-296.

Tarara, R.P., L.G. Hansen and J. Simon (1976). Effects of repeated administration of oxytetracycline on kidney, liver and liver mixed function oxidases in the rat. *Toxicol. Appl. Pharmacol.*, **35:** 321–331.

Chen, X., Y. Zhang, Z. Zhu, H. Liu, H. Guo, C. Xiong, K. Xie, X. Zhang abd S. Su (2016). Protective effect of berberine on doxorubicininduced acute hepatorenal toxicity in rats. *Mol. Med. Rep*., **13(5):** 3953–3960.

Feng, Y., K.Y. Siu, X. Ye, N. Wang, M.F. Yuen, C.H. Leung, Y. Tong and S. Kobayashi (2010). Hepatoprotective effects of berberine on carbon tetrachloride-induced acute hepatotoxicity in rats. *Chin. Med.*, **5:** 33–41.

Lin, Y., M. Sheng, Y. Weng, R. Xu, N. Lu, H. Du and W. Yu (2017). Berberine protects against ischemia/reperfusion injury after orthotopic liver transplantation via activating Sirt1/FoxO3alpha induced autophagy. *Biochem. Biophys. Res. Commun*., **483(2):** 885–891.

Yonar, M.E. (2012). The effect of lycopene on oxytetracycline-induced oxidative stress and immunosuppression in rainbow trout (Oncorhynchus mykiss, W.). *Fish Shellfish Immunol*., **32(6):** 994-1001.

Zhou, H. and S. Mineshita (2000). The Effect of Berberine Chloride on Experimental Colitis in Rats In Vivo and In Vitro. *J. Pharm. Exp. Ther.*, **294:** 822-829.

Zhou, Y., V.S. Vaidya, R.P. Brown, J. Zhang, B.A. Rosenzweig, K.L. Thompson, T.J. Miller, J.V. Bonventre and P.L. Goering (2008). Comparison of kidney injury molecule-1 and other nephrotoxicity biomarkers in urine and kidney following acute exposure to gentamicin, mercury, and chromium. *Toxicol. Sci.*, **101:** 159–170.