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ORIGINAL PAPER

Ameliorative effects of Edaravone against Valproic Acid-Induced kidney damage

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Abstract

Valproic acid (VPA) is a well-known and increasingly documented antiepileptic drug that has been widely used in the treatment of epilepsy and/or epilepsy-related disorders. Prolonged clinical use of VPA has been reported to cause side effects such as nephrotoxicity. Edaravone (EDA) is a powerful free radical scavenger. The aim of the study was to investigate the protective effects of EDA against VPA-induced oxidative renal injury. Four experimental groups were formed by randomly assigning thirty-eight male Sprague Dawley rats. The first group, (Control Group, $n=8$), consisted of healthy rats. The second group, (Group II, $n=10$), comprised control rats given intraperitoneally EDA (30 mg/kg/day) for seven days. The third group (Group III, $n=10$) was administered intraperitoneally only VPA (500 mg/kg/day) for seven days. The last group (Group IV, $n=10$) was treated with VPA + EDA for seven days. On the 8th day, kidney tissues were immediately removed from rats. In kidney homogenates, reduced glutathione levels and Na/K^+-ATPase, paraoxonase1 and prolidase activities were remarkably decreased while catalase, superoxide dismutase, glutathione peroxidase, glutathione reductase, myeloperoxidase, and xanthine oxidase activities and lipid peroxidation, protein carbonyl, advanced oxidized protein products, and hydroxyproline contents were notably elevated in VPA given group. Consistently, administration of EDA decreased renal degenerative changes seen in the kidney tissue of VPA given rats. Treatment with EDA in the VPA group significantly resulted in the recovery of both biochemical and histopathological alterations. As a result, EDA is potentially beneficial to revert oxidative renal damage induced by VPA.

Keywords Edaravone · Kidney injury · Valproic acid · Oxidative stress · Antioxidant enzymes

Introduction

Valproic acid (2-propylpentanoic acid, VPA) is a branched short-chain aliphatic fatty acid, and it has anticonvulsant effects that is ubiquitously used to treat certain types of seizures, migraine, anxiety, bipolar, and posttraumatic stress disorders as well as other psychiatric conditions in children and adults (Wu et al. 2020). On the other hand, not only dose-dependent but also prolonged use of VPA has been reported to cause adverse effects on several organs such as liver (Celik et al. 2021; Oztopuz et al. 2020), brain (Turkyilmaz et al. 2021), lung (Bayrak et al. 2021; Oztay et al. 2020), lens (Dagsuyu et al. 2023), muscle (Ertik et al. 2023), pancreas (Deschenes et al. 2020), intestinal (Drokov et al. 2020), and testicular injuries (Çelik et al. 2022; Maneenin et al. 2018). In addition, it has a teratogenic effect (Lin et al. 2019). Up to date, numerous research articles have been published that VPA has unwanted effects on kidney tissues such as nephrotoxicity, general malfunction of the proximal

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renal tubular injury, known as Fanconi syndrome (El-Shenawy and Hamza 2016; Heidari et al. 2018; Maneenin et al. 2019). Furthermore, it has been shown that inflammation and apoptosis markers in the kidney tissues of mice increase as a result of exposure to VPA (Lee et al. 2023). Similarly, it has been revealed by Almikhlafi et al. (2023) that an acute overdose of VPA increases the formation of pro-inflammatory cytokines, thereby leading to both liver and kidney damage. Regarding the therapeutic use of VPA or its sodium salt, especially histological changes in kidney tissue have been investigated and it has been reported to cause degeneration in renal tubules, edema, and atrophy of the glomeruli (Al-Amoudi 2017). The data obtained by Gezginci-Oktayoglu et al. (2016) show that VPA gives rise to renal damage by promoting inflammation, oxidative stress (OS), and fibrosis.

Under normal cellular conditions, free radicals (reactive substances) called both reactive oxygen species (ROS) and reactive nitrogen species (RNS) have been continuously generated by cells. At low levels, they are involved in several cellular processes (e.g., important signaling pathways, apoptosis, cell differentiation, growth, and inflammation) (Ratliff et al. 2016). On the other hand, OS has been associated with an imbalance of these reactive substances' formation and detoxification, which brings about oxidative tissue injury. Moreover, OS is accepted as an important factor in the pathophysiology of the acute kidney injury which leads to a loss of renal function within a week (Gyurászová et al. 2020). On the other hand, accumulating evidence obviously indicates that the pathogenesis of VPA-induced (nephro) toxicity is the result of OS caused by the extravagant formation of ROS (Bayrak et al. 2021; Çelik et al. 2022; Gezginci-Oktayoglu et al. 2016; Kandemir et al. 2022).

Edaravone (EDA; 3-methyl-1-phenyl-2-pyrazoline-5-one or PMP and Radicava) is a clinically prescribed drug that has been used in the treatment of cerebral infarction and amyotrophic lateral sclerosis, since 2001 in Japan. After its approval, EDA was also used in South Korea in 2015, in the U.S. in 2017 and in Chinese in 2019 for the same therapeutic purpose (Cruz 2018; Bailly et al. 2020; Dash et al. 2018). Kikuchi et al. (2012) have reported that EDA can inhibit lipid peroxidation via its strong antioxidant effects and suppress OS induced by ROS. Thus, it can play a crucial role in multiple organ systems. Cumulative findings from published articles indicate that EDA can be useful to improve cancer treatment and protects against deleterious effects of chemotherapy as well as radiotherapy (Bailly 2019). Moreover, EDA has been shown to have vigorous effects on scavenging free radicals (e.g., hydroxyl and peroxynitrite radicals) (Watanabe et al. 2018) and antinecrotic, antiapoptotic, and anti-inflammatory activities (Atallah et al. 2023; Hassanein et al. 2023; Kikuchi et al. 2013; Zhao et al. 2020; Zeng et al. 2022). More so, at doses of 1 mg/kg, 5 mg/kg, 10 mg/kg, and 100 mg/kg EDA administration has recently been reported to have ameliorative effects against renal injury models in rats (Koike et al. 2021; Yang et al. 2023).

With the presence of the information mentioned above, the main goal of our work was to investigate potential ameliorative effect of EDA against oxidative renal damage induced by VPA.

Materials and methods

Chemicals and reagents

The VPA (catalog number 8.14439; ≥ 99.0%) and EDA (catalog number 8.07080; ≥ 98.5%) were obtained from Merck (Darmstadt, Germany). Other chemicals were of analytical or HPLC grade and were used as received without any further purification.

Animal treatment

In this work, Male Sprague Dawley rats were used as subjects. The rats were 10–12 weeks old and clinically healthy. The animal study protocol followed in this research was approved by the Local Ethics Committee of the Istanbul University (approval number 2010/54). Four experimental groups were formed by randomly assigning thirty-eight rats. The first group, named control (Group I, $n = 8$), consisted of healthy rats. The second group, referred to as Group II ($n = 10$), comprised control rats given intraperitoneally EDA (30 mg/kg/day) for seven days (Hacihasanoglu and Yanardag 2015). The third group (Group III, $n = 10$) was administered intraperitoneally only VPA (500 mg/kg/day) for seven days (Tunali 2014). The last group (Group IV, $n = 10$) was treated with VPA + EDA for seven days (at the same dose and at the same time). At the end of the administration period, all the animals were sacrificed under anesthesia (ketamine hydrochloride and xylazine HCl were used), and kidney tissues were removed for biochemical and histopathological experiments. Kidney tissues taken from rats were frozen at -80 °C for further experiment after they were washed with cold saline.

On the day of the experiments, after blood and external vessels were carefully discarded, kidney tissue samples were homogenized in 10 volumes (1:10, weight/volume) of cold physiologic saline with a glass equipment (homogenizer). After centrifugation at 10.000 x g at 4 °C for 10 min, clear supernatants were obtained and used for biochemical assays.

Biochemical analysis

Supernatants were used for the biochemical assays i.e. reduced glutathione (GSH) and lipid peroxidation (LPO) levels, catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GPx), glutathione reductase (GR), myeloperoxidase (MPO), sodium/potassium adenosine triphosphatase (Na^+/K^+-ATPase), xanthine oxidase (XO), paraoxonase1 (PON1), and prolidase activities and protein carbonyl (PC), advanced oxidized protein products (AOPP), hydroxyproline (Hyp) and total protein levels determination.

Determination of reduced glutathione (GSH) levels

Beutler's method was used to determine the GSH levels of kidney tissues (Beutler 1975). Protein contents found in homogenate was precipitated with metaphosphoric acid and GSH was determined by reading the color of the complex formed with 5,5'-dithiobis-2-nitrobenzoic acid spectrophotometrically at 405 nm. Findings were presented in terms of nmol GSH/mg protein.

Determination of lipid peroxidation (LPO) levels

Evaluation of LPO levels in kidney tissues of experimental groups was done according to the methodology of Ledwozyw et al. (1986). The absorbance of the pink color produced at the conclusion of the reaction between the LPO product malondialdehyde (MDA) and thiobarbituric acid was evaluated spectrophotometrically at 532 nm in order to ascertain the extent of LPO. The results were presented as nmol MDA/mg protein.

Determination of catalase (CAT) activity

CAT activity was determined by following the reduction of H_2O_2 to H_2O and by monitoring decline in the absorbance at 240 nm (Aebi 1984). The results obtained were presented as U/mg protein.

Determination of superoxide dismutase (SOD) activity

The reaction protocol utilized a phosphate buffer with a pH of 7.8, o-dianisidine dihydrochloride, and riboflavin. After the addition of riboflavin under a fluorescent lamp, the absorbance values of the mixture were recorded at initial and after 8 min at 460 nm. The results were given as U/g protein by using the standard graph (Mylroie et al. 1986).

Determination of glutathione peroxidase (GPx) activity

In the determination of GPx activity, the reduction of H_2O_2 to H_2O was measured at 366 nm, after the addition of GSH, NADPH, and glutathione reductase to the reaction mixture during this measurement (Paglia and Valentine 1967) GPx activity was expressed as U/g protein.

Determination of glutathione reductase (GR) activity

The activity of the GR, which is catalyzed by oxidized glutathione to GSH, was performed by methodology of Beutler (1971). GR activity was determined by calculating the amount of NADH oxidized during the reduction of oxidized glutathione with GR. GR activity was given as U/g protein.

Determination of myeloperoxidase (MPO) activity

Activities of MPO in kidney homogenates were determined by the reaction of 4-aminoantipyrine, phenol (2%) and H_2O_2 with homogenate. The resulting color change was read at 510 nm in the spectrophotometer (Wei and Frenkel 1991). The results were defined as mU/g protein.

Determination of xanthine oxidase (XO) activity

This enzyme, XO, converts xanthine to uric acid. To evaluate its activity, phosphate buffer (pH 7.4), EDTA, xanthine, and sample were kept in the same environment and the initial absorbance reading was taken on the spectrophotometer. Second absorbance values were recorded spectrophotometrically after 10 min of incubation period at room temperature and at 286 nm (Corte and Stirpe 1968). XO activity was expressed as U/g protein.

Determination of sodium/potassium-adenosine triphosphatase (Na^+/K^+-ATPase)

The methodology of Ridderstap and Bonting (1969) was used for the determination of Na^+/K^+-ATPase activity in renal tissue with the help of the determination of Mg^{2+}-ATPase in the presence of ouabain and ATP in acidic medium. The outcomes were expressed as nmol P_i/mg protein/h.

Determination of paraoxonase1 (PON1) activity

In kidney tissue homogenates, PON1 activities was performed according to method of Furlong et al. (1988). Briefly, the reaction medium containing assay buffer (Tris-HCl, pH 8.5, $CaCl_2$, and NaCl), tissue homogenate, and freshly

prepared paraoxon-ethyl as substrate. At 37°C, the reaction was initiated by the addition of the substrate solution, and the absorbance was recorded spectrophotometrically at 405 nm. The PON1 activity was expressed as U/g protein.

Determination of prolidase activity

This method is based on the color reaction of proline, found in tissue homogenate, with acidic ninhydrin solution (1:1 ratio of concentrated acetic acid) at pH 1.0 in boiling water for 1 h, thereafter the absorbance at 515 nm was measured with a spectrophotometer (Chinard 1952). The outcomes were expressed as U/mg protein.

Determination of protein carbonyl (PC) levels

The levels of PC were determined with 2,4-dinitrophenylhydrazone, which is formed by the reaction of carbonyl groups in proteins with 2,4-dinitrophenylhydrazine (DNPH) (Levine et al. 1990). Tissue homogenate and DNPH solution were added same tube, and the incubation was performed at room temperature. After trichloro acetic acid solution (20%) was added to each tube. The resultant precipitates were washed. Then, guanidine-HCl was put into each tube and the incubation was formed at 37°C for 30 min. Finally, the absorbance values were taken by using a spectrophotometer at 370 nm. The results were given in nmol PC /mg protein.

Determination of advanced oxidized protein products (AOPP) levels

The AOPP levels in kidney tissues were determined according to the method of Witko-Sarsat et al. (1996). The color of the product was measured at 340 nm in the presence of phosphate buffer (pH 7.40), KI, and concentrated acetic acid. The results were shown as pmol AOPP/mg protein.

Determination of hydroxyproline (Hyp) levels

In kidney tissue homogenates, Hyp levels were measured via collagen status, a major skin protein (Reddy and Enwemeka, 1996). The samples were hydrolyzed in the acidic medium and then neutralized. The neutralized samples were mixed with chloramine-T reagent. The chromophore was then developed with the addition of *p*-dimethylaminobenzaldehyde (Ehrlich's) reagent, and the absorbance of the reddish-purple complex was measured at 550 nm. The results obtained were expressed as µg Hyp/mg protein.

Determination of total protein contents

The amount of protein in the kidney tissues were quantified by the methodology of Lowry et al. (1951). This involved the measurement of the intensity of the blue-violet color, which is formed as a consequence of the reduction of proteins that have been reacted with copper ions in an alkaline medium with Folin reagent (phosphomolybdotungstic acid). This process was conducted spectrophotometrically at 500 nm.

Histopathological evaluation

Kidney tissues were fixed in Bouin's solution, dehydrated in increasing ethanol series, cleared in xylene, and embedded into paraffin blocks. The kidney tissue sections at 5 µm were taken by rotary microtome (Leica, Germany) and renal histology was evaluated by observation of the tissue sections stained with Masson's trichrome using Olympus (Tokyo, Japan) BX53 light microscope.

Statistical analysis

The data obtained from each analysis were entered into GraphPad Prism 6.0 (San Diego, USA) statistics software for statistical analysis. The Tukey's multiple comparison tests and one-way analysis of variance (ANOVA) were used to identify any significant differences ($p < 0.05$). In this research, all results were reported as the mean value along with the standard deviation (SD).

Results

Biochemical evaluations

Figure 1 illustrates GSH and LPO levels of kidney tissues. Levels of GSH in the VPA group were notably lower than that of control rats ($p < 0.05$). On the other hand, LPO levels remarkably rose in the VPA treated rats compared to control group ($p < 0.001$). Both GSH and LPO levels ($p < 0.0001$ and $p < 0.001$, respectively) considerably reverted after exposure to EDA in the VPA group (Fig. 1).

Figure 2 depicts activities of CAT, SOD, GPx, and GR. Activities of CAT remarkably elevated in EDA administered group in comparison with control group ($p < 0.01$). All enzyme activities notably heightened in VPA given group in comparison to control group, respectively ($p < 0.0001$ and $p < 0.001$). Contrarily, EDA treated with the VPA-administered animals caused a notable decrease in the CAT, SOD, GPx, and GR activities, respectively ($p < 0.0001$, $p < 0.001$, and $p < 0.01$) (Fig. 2).

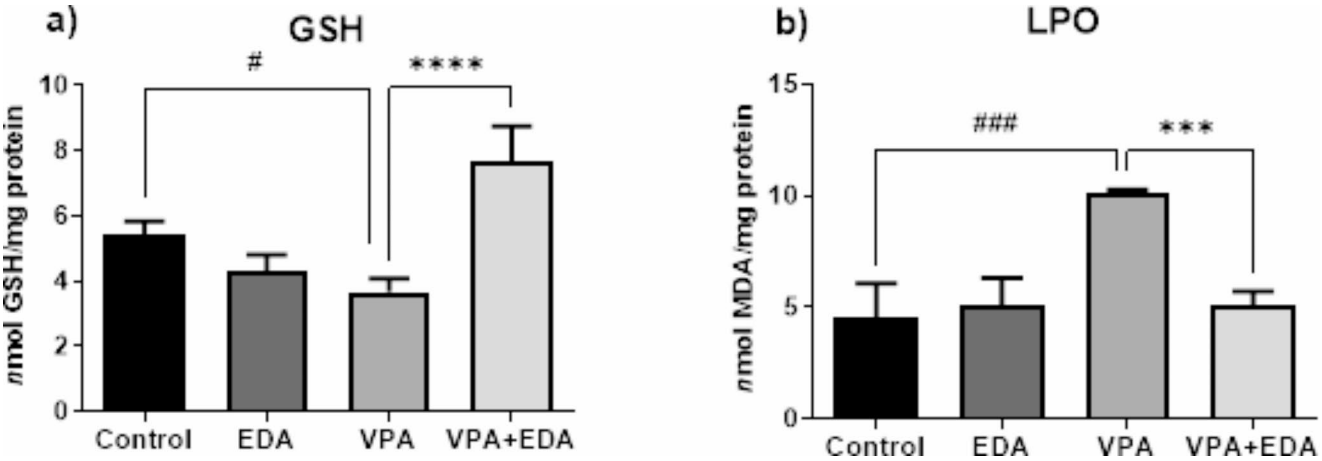

Fig. 1 Effect of edaravone on kidney tissue (**a**) GSH and (**b**) LPO levels of all groups of rats. All data are presented as mean ± SD. GSH: Glutathione reduced form; LPO: Lipid peroxidation; MDA: Malondialdehyde; EDA: Edaravone; VPA: Valproic acid; VPA + EDA: Valproic acid + Edaravone. $^{\#}p < 0.05$; $^{\#\#\#}p < 0.001$ vs. control; $^{****}p < 0.0001$; $^{***}p < 0.001$ vs. VPA

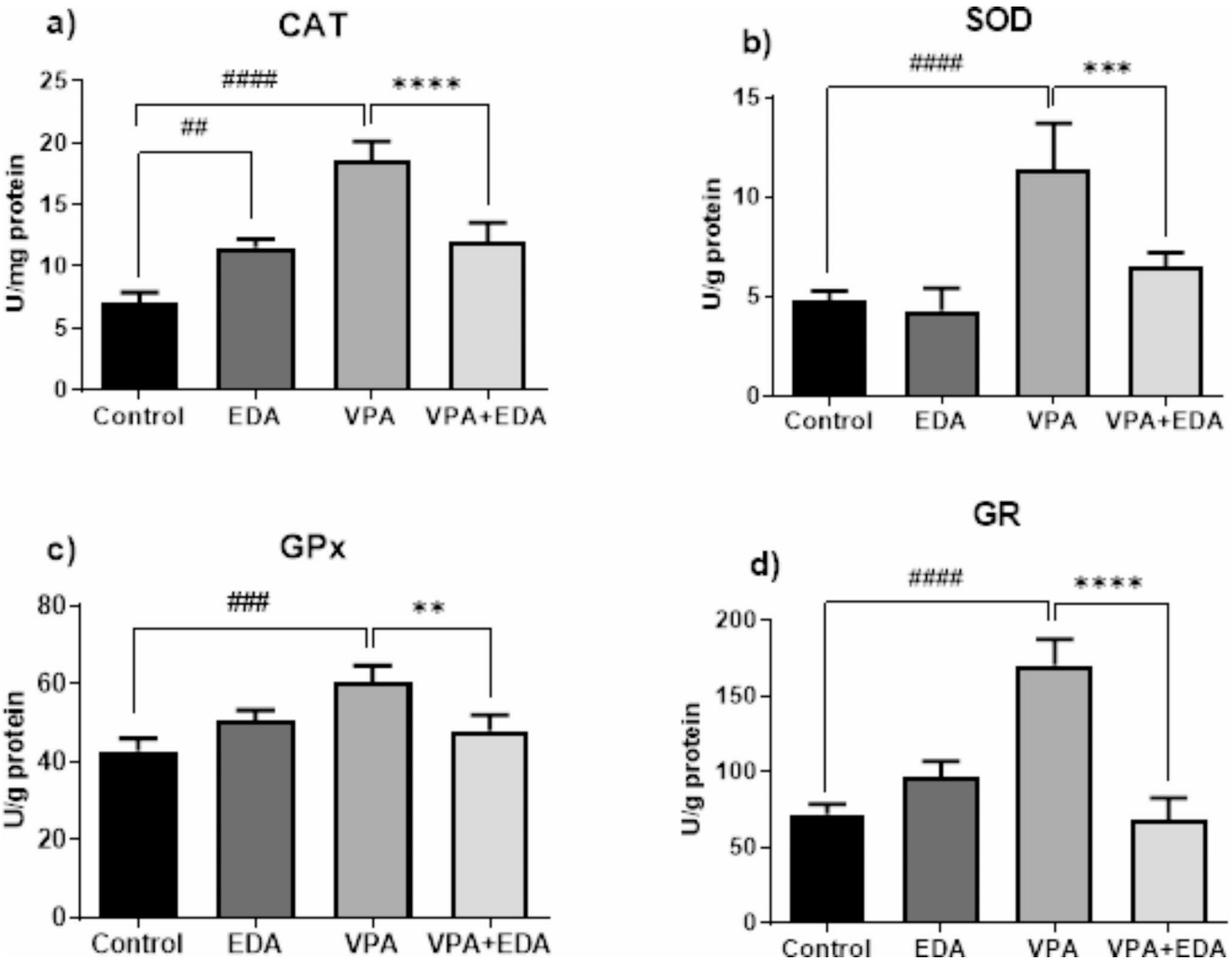

Fig. 2 Effect of edaravone on kidney tissue (**a**) CAT, (**b**) SOD, (**c**) GPx, and (**d**) GR activities of all groups of rats. All data are presented as mean ± SD. CAT: Catalase; EDA: Edaravone; GPx: Glutathione peroxidase; GR: Glutathione reductase; SOD: Superoxide dismutase; VPA: Valproic acid; VPA + EDA: Valproic acid + Edaravone. $^{\#\#}p < 0.01$; $^{\#\#\#\#}p < 0.0001$; $^{\#\#\#}p < 0.001$ vs. control; $^{****}p < 0.0001$; $^{***}p < 0.001$; $^{**}p < 0.01$ vs. VPA

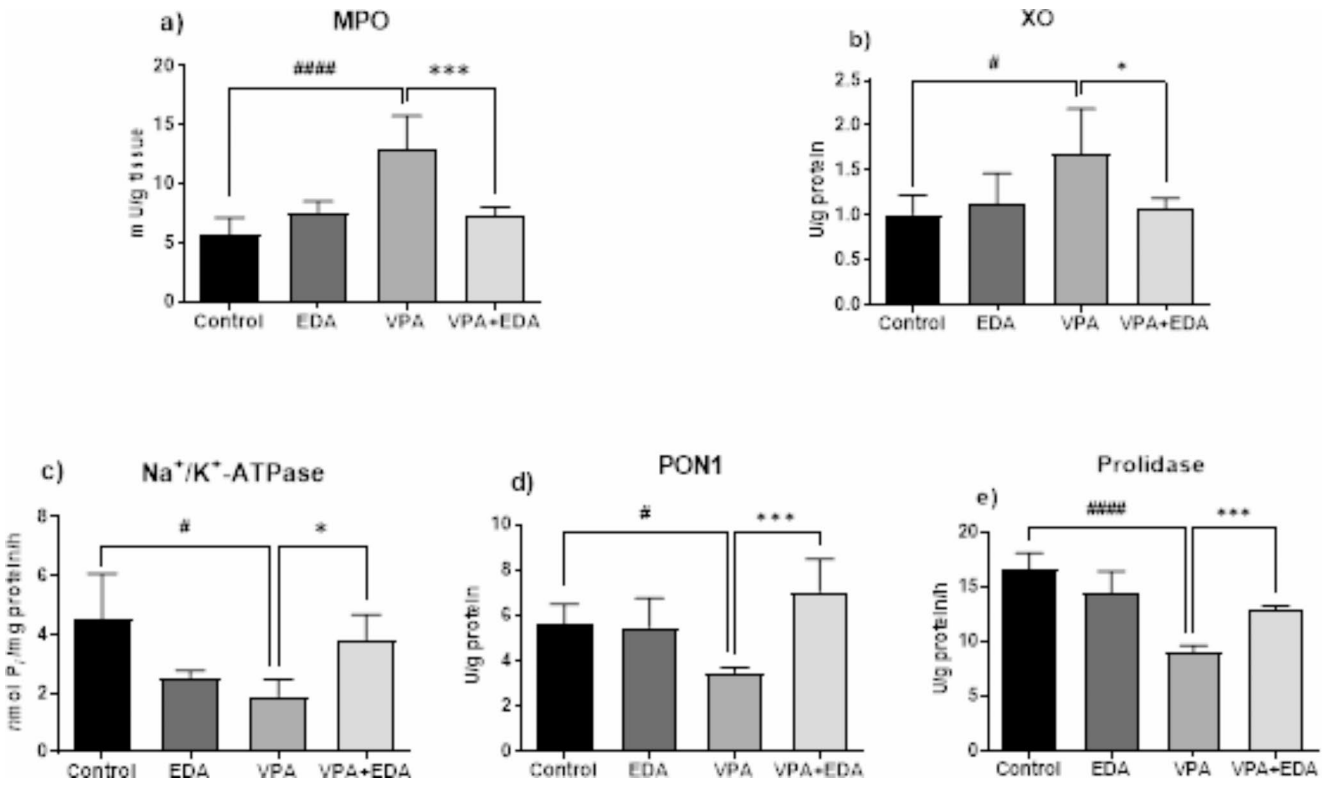

Fig. 3 Effect of edaravone on kidney tissue (**a**) MPO, (**b**) XO, (**c**) Na^+/K^+-ATPase, (d) PON1, and (e) prolidase activities of all groups of rats. All data are presented as mean ± SD. EDA: Edaravone; MPO: Myeloperoxidase; XO: Xanthine oxidase; Na^+/K^+-ATPase: Sodium/potassium adenosine triphosphatase; PON1: Paraoxonase1; VPA: Valproic acid; VPA + EDA: Valproic acid + Edaravone. $^{\#\#\#\#\#}p < 0.0001$; $^{\#}p < 0.05$ vs. control; $^{***}p < 0.001$; $^{*}p < 0.05$ vs. VPA

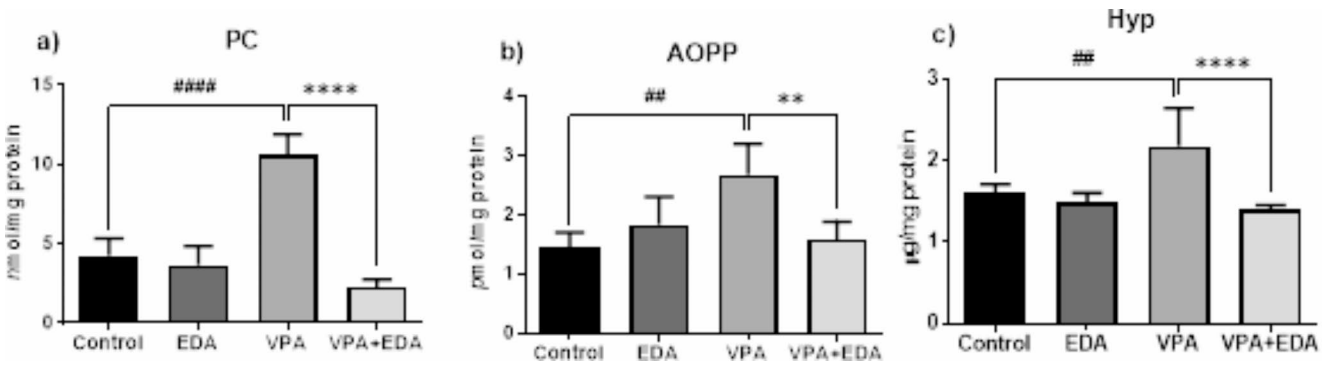

Fig. 4 Effect of edaravone on kidney tissue (**a**) PC, (**b**) AOPP, and (**c**) Hyp levels of all groups of rats. All data are presented as mean ± SD. AOPP: Advanced oxidized protein products; EDA: Edaravone; Hyp: Hydroxyproline; PC: Protein carbonyl; VPA: Valproic acid; VPA + EDA: Valproic acid + Edaravone. $^{\#\#\#\#}p < 0.0001$; $^{\#\#}p < 0.01$ vs. control; $^{****}p < 0.0001$; $^{**}p < 0.01$ vs. VPA

MPO, Na^+/K^+-ATPase, PON1, XO, and prolidase activities of all experimental groups are depicted in Fig. 3. We observed a notable elevation of both MPO ($p < 0.0001$) and XO activities ($p < 0.05$) in the VPA-administered animals in comparison with the control group. Treatment with EDA to VPA group resulted in a considerable diminishment in these enzyme activities, respectively ($p < 0.001$ and $p < 0.05$) (Fig. 3). On the contrary, there was a considerable decrease in Na^+/K^+-ATPase ($p < 0.05$), PON1 ($p < 0.05$), and prolidase ($p < 0.0001$) activities in the VPA-administered animals compared to the control group. These enzyme activities sharply elevated when EDA was administered to the VPA group, respectively ($p < 0.05$ and $p < 0.001$) (Fig. 3).

Figure 4 shows the PC, AOPP, and Hyp contents of kidney tissues. As seen in Fig. 4, we detected that PC, AOPP, and Hyp contents were found to be a remarkable increase in VPA treated group in comparison with the control group ($p < 0.0001$ and $p < 0.01$). On the other hand, administration

of EDA to VPA given group notably reverted these alterations, respectively ($p < 0.0001$ and $p < 0.01$) (Fig. 4).

Histopathological evaluations

Figure 5 illustrates VPA-induced histopathological damage. The kidney tissues in the control and EDA-given groups were normal. VPA caused significant histopathological damage in the kidney tissues such as necrotic areas and disruption of epithelium in proximal and distal tubules, desquamated nuclei in tubule lumens. EDA treatment decreased the degenerative changes seen in the kidney tissues of VPA given group (Fig. 5).

Discussion

VPA, an antiepileptic drug, has been used in the treatment of psychiatric conditions such as epilepsy and bipolar disorder for over 45 years (Richens and Ahmad 1975). Even though VPA has been revealed to be well tolerated and primarily detoxified by the liver, to date, many research articles have been published that clinical use of VPA can cause adverse/side effects on both human and experimental animal models on several tissues and organs such as skin (Tunali-Akbay et al. 2017), and kidney (Maneenin et al. 2019). In kidneys, the use of sodium salt of VPA (as an antiepileptic drug) can result in excessive production of ROS accompanied with OS, giving rise to structural changes in proximal renal tubules and thereby bringing about in loss of their ability to reabsorb amino acids, electrolytes, urea, and glucose

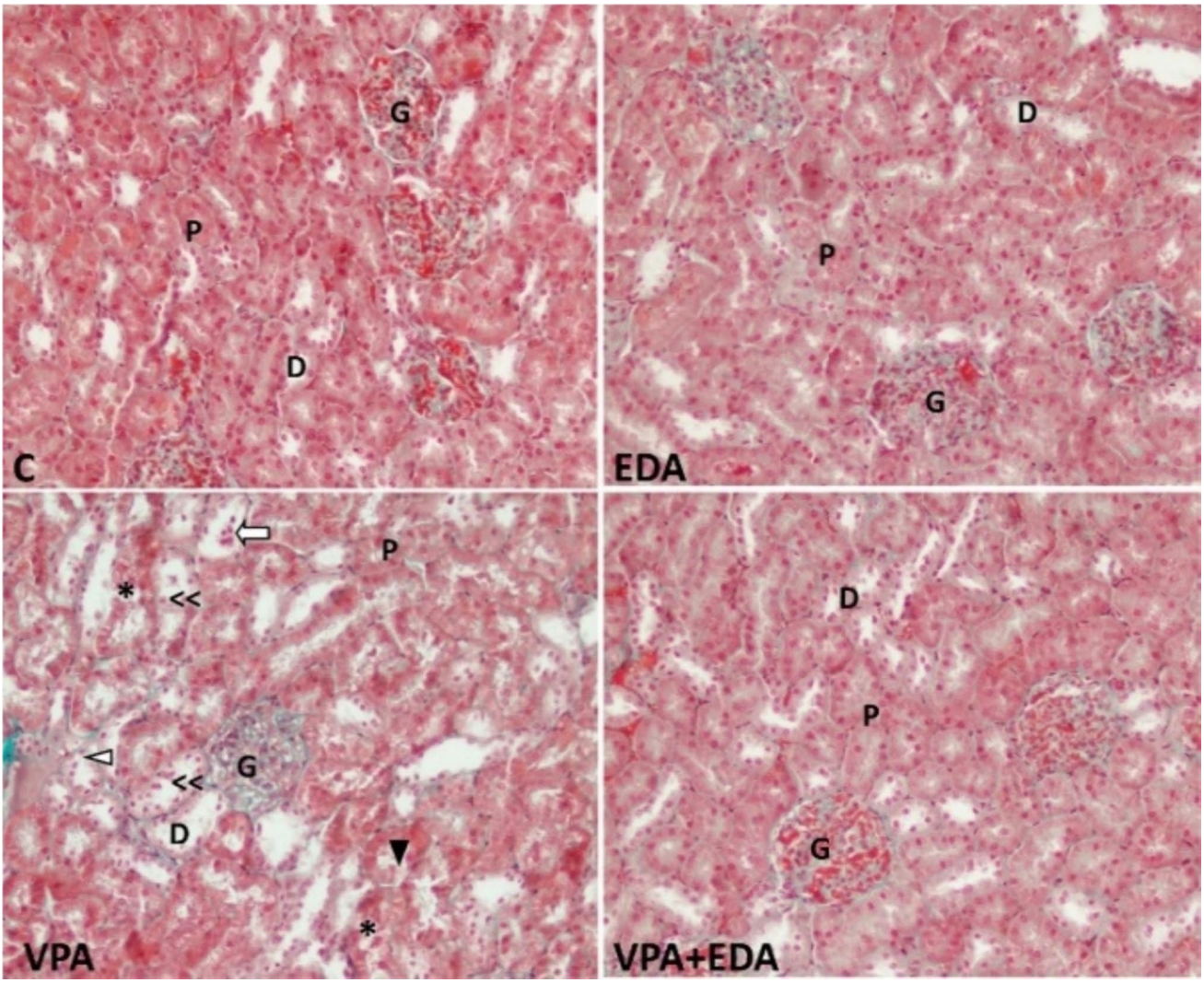

Fig. 5 Histopathological changes in kidney tissues. C: Control; EDA: Control group given Edaravone; VPA: Valproic acid group; VPA+EDA: Valproic acid+Edaravone given group. G: Glomerulus; P: Proximal tubules; D: Distal tubules, *: Necrotic areas in proximal tubules, «: Necrotic areas in distal tubules, black arrowhead: Epithelial disruption proximal tubules, whitearrowhead: Epithelial disruption in distal tubules, ⇨: Desquamated nuclei in tubule lumens (Magnification x200)

(Knights and Finlay 2014; Azırak et al. 2022). On the other hand, EDA is as robust an antioxidant as natural antioxidants (e.g., GSH) (Kamogawa and Sueishi 2014). Because of EDA's powerful antioxidant role, we aimed to investigate whether EDA has ameliorative effects on VPA-induced oxidative renal damage in rats.

Important biochemical markers such as GSH and LPO were evaluated as an index of oxidative renal injury. In the present work, remarkable declining levels of GSH were seen whereas notable rising levels of LPO were detected in the VPA given group in comparison to normal rats. In a study on the influence of caffeic acid against sodium valproate (SVP)–induced nephrotoxicity in rats conducted by Gad (2018), it was found similar alterations in both GSH and LPO levels. El-Shenawy and Hamza (2016) reported that administration of both low and high dose SVP caused nephrotoxicity and they found a remarkable elevation of LPO levels. In another article about liver toxicity induced by VPA has been shown that GSH levels were significantly lower whereas LPO levels were notably higher than that of the control group (Abdelkader et al. 2020). Another study done by Jutrić et al. (2023) reported similar findings in mouse kidneys with a 3.33-fold lower dose of valproic acid salt. According to our results, we detected a considerable recovery of both GSH and LPO levels as a result of EDA given to the VPA group. EDA treatment to kidney tissue showed an ameliorative effect since EDA has a potent free radical scavenger (Kikuchi et al. 2012).

In addition to non-enzymatic antioxidants in cells, there are important first-line antioxidant enzymes such as CAT, SOD, GPx, and GR which protect the cells from the deleterious effects of free radicals. If effective measures for the prevention and management of oxidant formation are not put in place by antioxidants in the cell, a decrease and/or increase in the activities of these antioxidant enzymes occurs (Chaudhary et al. 2015). In the present study, it was seen that all aforementioned enzyme activities tended to increase markedly in VPA-induced renal OS. However, there are inconsistent results regarding the change of these enzyme activities in the literature. There are published studies inconsistent with our results (Bayrak et al. 2021; El-Shenawy and Hamza 2016; Gezginci-Oktayoglu et al. 2016) and in the same line with our findings (Chaudhary et al. 2015; Çelik et al. 2022; Tunali 2014). On the other hand, a study by Kandemir et al. (2022), was conducted on the effects of SVP in rat liver and kidney tissues and reported that SVP induced OS in both tissues by inhibiting SOD, CAT, and GPx activities. Another research with opposite outcomes by Jutrić et al. (2023) also revealed that the administration of 150 mg of valproate per kg of body weight a day in the kidneys of mice gave rise to a drop SOD and CAT activities. The possible reason for their contradictive findings may be related to the difference between duration time and application route of SVP. In the current study, a considerable drop in the activities of these enzymes was seen when EDA was implemented in VPA given rats. This may be associated with the elevated GSH and declined LPO levels in kidney tissues concomitantly with the EDA administration, which could explain a notable recovery of the first-line defense enzymes.

MPO, a lysosomal enzyme and OS marker, is present in granulocytic and monocytic cells and it has also peroxidase activity. MPO catalyzes the conversion of H_2O_2 and chloride anion to form hypochlorous acid and other ROS (Ojo et al. 2022). MPO activity in kidney homogenate was evaluated as a marker of VPA-induced renal damage. XO is an enzyme that has both dehydrogenase and oxidoreductase activities, which is responsible for the formation of reactive substances (e.g., superoxide anion and H_2O_2) (Nishino et al. 2008). According to the current study, we observed a noteworthy elevation of both MPO and XO activities in the VPA group when compared with the control group. Previous researches on VPA-induced liver and kidney toxicity were in parallel with our results (Gezginci-Oktayoglu et al. 2016; Sokmen et al. 2012). Moreover, VPA-induced OS led to a considerable rise in MPO activities in rat's small intestine (Oktay et al. 2015) and Oztopuz et al. (2020) showed that SVP-induced hepatotoxicity in rats caused an insignificant increase in MPO activity. In our study, there was a significant decline in both MPO and XO activities when EDA given to the VPA group. Similar findings were reported for liver (Hacihasanoglu Cakmak and Yanardag 2015), gingiva (Oktay et al. 2016), and pancreas (Oktay et al. 2017). Na^+/K^+-ATPase, an integral membrane enzyme, acts as an ion pump in all eukaryotic cells and that is necessary for the regulation of both Na^+ and K^+ ion homeostasis against their electrochemical gradients (Azalim et al. 2020). It has been reported that a rise in both ROS and RNS gives rise to a decrease in Na^+/K^+-ATPase activity as a result of oxidative modification of this enzyme such as S-glutathionylation of Na^+/K^+-ATPase subunits (Liu et al. 2018; Petrushanko et al. 2012). In the present study, decreased renal Na^+/K^+-ATPase activity was observed in VPA injected rats. Gezginci-Oktayoglu et al. (2016) also showed that Na^+/K^+-ATPase activity in the kidney notably declined in VPA given group. The possible reason for the decrease in renal Na^+/K^+-ATPase activity after 500 mg/kg/day VPA injection may be linked to an increase in LPO levels and concomitantly disruption of membrane integrity as a consequence of oxidative damage in the kidney caused by VPA. On implementing EDA, the Na^+/K^+-ATPase activity in renal tissue increased significantly. PON1 is an enzyme that is widely used as a biomarker based on its protective effect against organophosphate toxicity. Besides, it has been demonstrated that a decrease in PON1 activity is related to a rise in OS

because of its antioxidant properties (Ceron et al. 2014; Permpongpaiboon et al. 2011). In our study, PON1 activity in VPA given group was significantly found to be lower than control group. It has been reported by Sokmen et al. (2012) that VPA given rats caused a remarkable decrease in PON1 activity in liver tissues. In another research on SVP and Levetiracetam treatment in children, serum PON1 activity also decreased (Türe and Yazar 2020). Tavafi et al. (2020) reported that PON1 activity declined as a result of OS induced by renal ischemia/reperfusion injury. According to our study, kidney PON1 activity in VPA given group was attenuated by EDA administration. Our findings were in parallel with the findings of Hacihasanoglu Cakmak and Yanardag (2015). Prolidase, a metallo-dependent hydrolase, is the only intracellular enzyme that is essential for cleaving the iminodipeptides containing a proline or a Hyp residue at the carboxyl terminus (Wilk et al. 2021). Furthermore, several research groups reveal that change in prolidase activity in blood or tissue is used as an index of biomarker for OS (Kapucu et al. 2021; Pirinççi et al. 2016). In the current work, there was a notable diminishment in kidney prolidase activity in the VPA injected group compared to control rats. The decrease in prolidase activity in kidney tissue may be owing to leakage from cells. This could result from the disruption of cell membrane integrity caused by VPA-mediated OS in renal tissue, hence accompanied by an increase in LPO levels concomitantly with a decrease in renal Na^+/K^+-ATPase activity. On the other hand, it has been indicated that OS-related nephropathy, as well as renal cell carcinoma, resulted in the elevation of serum prolidase activity (Pirinççi et al. 2016; Verma et al. 2014). In the present study, EDA treatment caused a rise in prolidase activities in VPA given rats by providing restoration of the disrupted renal cell membrane, thus decreasing OS.

It is exactly known that whenever ROS generated as a result of an imbalance of redox status in cell, oxidative modification of protein molecules as well as enzymes may occur in physiologic and pathophysiologic processes (Levine et al. 1994). When we measured PC and AOPP levels to determine the extent of kidney tissue damage caused by VPA, it was observed a significant increase in both PC and AOPP levels after VPA exposure. The administration of EDA prevented these alterations. A notable decrease in these marker's levels may be due to the potent antioxidant action of EDA (Kamogawa and Sueishi 2014). More so, SVP treatment (5, 10, and 20 mg) to kidney postnuclear homogenate has been revealed to cause a significant elevation of PC levels in terms of a dose-dependent manner under in vitro conditions (Chaudhary et al. 2015). Hyp is the chief component of fibrous protein in the extracellular matrix such as collagen and high levels of Hyp are considered an important indicator of fibrosis in tissues (Sharma et al. 2021). Measurement of the Hyp contents is used to assign the effect of OS-induced pathophysiological processes on the tissue (Cissel et al. 2017). For this reason, we determined Hyp levels as an index of tissue fibrosis in the kidney (Heidari et al. 2019). In the current study, there was a significant elevation in renal Hyp levels in VPA given group in comparison to the control group. Our results were in agreement with Abdelkader et al. (2020). When rats were treated with EDA, the renal Hyp levels remarkably declined. These data suggest that EDA may have shown a protective effect against kidney fibrosis by reducing the level of Hyp that has increased due to OS.

The beneficial effects of EDA were demonstrated on the impaired oxidant/antioxidant status of the heart in VPA-induced toxicity (Emekli-Alturfan et al. 2015). It is also known that EDA prevented inflammatory and oxidative reactions in the skin tissue after the VPA administration (Tunali-Akbay et al. 2017). In our study, biochemical results support histopathological results in the kidney tissue of rats given VPA. EDA which has an antioxidant effect decreased degenerative changes seen in the kidney tissue of VPA-given rats in agreement with biochemical results.

Conclusions

Reducing GSH levels, increasing LPO, PC, AOPP, and Hyp levels as well as altered enzymatic antioxidants levels in kidney tissues is harmonious with tissue damage and with histopathological changes (i.e. necrotic areas and disruption of epithelium in proximal and distal tubules, desquamated nuclei in tubule lumens) in kidney tissues, which is due to 500 mg/kg VPA administration to healthy rats.

We concluded that EDA, a robust antioxidant molecule, ameliorates against OS, thereby preventing renal injury induced by VPA when the following findings were observed: This is supported by the observation that rats treated with EDA showed recuperation in degenerative changes in their proximal and distal tubules, a decline in LPO, PC, and AOPP rates, and a return of nearly normal levels of marker enzymes in kidney tissues.

The usefulness of the present work is that 30 mg/kg EDA treatment to rats under OS conditions induced by VPA, biochemical results supported the findings showing histopathological changes.

The weaknesses/limitations of the current study are that the lack of glomerular filtration rate which is a test for determining to check out how well the kidneys of rats are working. Secondly, the accuracy of real-time PCR and/or Western blot results showing selected markers of OS and antioxidant factors cannot be confirmed due to financial problems.

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Data availability No datasets were generated or analysed during the current study.

Declarations

Ethical approval All experimental protocols were approved by the Animal Care and Use Committee of the Istanbul University, Istanbul, Turkey (ethics committee approval number 2010/54) and carried out in compliance with the ARRIVE guidelines. All animal experiments were conducted following the national guidelines and the relevant national laws on the protection of animals.

Competing interests The authors declare no competing interests.

References

Abdelkader NF, Elyamany M, Gad AM, Assaf N, Fawzy HM, Elesawy WH (2020) Ellagic acid attenuates liver toxicity induced by valproic acid in rats. J Pharmacol Sci 143:23–29. https://doi.org/10.1016/j.jphs.2020.01.007

Aebi H (1984) Catalase in vitro. Methods Enzymol 105:121–126. https://doi.org/10.1016/S0076-6879(84)05016-3

Al-Amoudi WM (2017) Protective effects of fennel oil extract against sodium valproate-induced hepatorenal damage in albino rats. Saudi J Biol Sci 24:915–924. https://doi.org/10.1016/j.sjbs.2016.10.021

Almikhlafi MA, Abouzied MM, Elbadawy HM, Eltahir HM, El-Sayed AAA, Alhaddad A, Wanas H, Mostafa MAH, Shehata AM (2023) Quercetin protective effects against hepatic and renal damage induced by an acute overdose of valproate. Lat Am J Pharm 42(3):1633–1644

Atallah M, Yamashita T, Hu X, Hu X, Abe K (2023) Edaravone confers neuroprotective, anti-inflammatory, and antioxidant effects on the fetal brain of a placental-ischemia mouse model. J Neuroimmune Pharmacol 18:640–656. https://doi.org/10.1007/s11481-023-10095-6

Azalim P, do Monte FM, Rendeiro MM, Liu X, O'Doherty GA, Fontes CF, Leitão SG, Quintas LEM, Noël F (2020) Conformational states of the pig kidney Na^+/K^+-ATPase differently affect bufadienolides and cardenolides: a directed structure-activity and structure-kinetics study. Biochem Pharmacol 171:113679. https://doi.org/10.1016/j.bcp.2019.113679

Azırak S, Taştemir Korkmaz D, Bilgiç S, Özgöçmen M, Özer MK (2022) Thymoquinone prevents valproic acid-induced nephrotoxicity in rat kidney. Eur J Biol Chem Sci 5(2):77–84. https://doi.org/10.46239/ejbcs.1123892

Bailly C (2019) Potential use of edaravone to reduce specific side effects of chemo-, radio- and immuno-therapy of cancers. Int Immunopharmacol 77:105967. https://doi.org/10.1016/j.intimp.2019.105967

Bailly C, Hecquet PE, Kouach M, Thuru X, Goossens JF (2020) Chemical reactivity and uses of 1-phenyl-3-methyl-5-pyrazolone (PMP), also known as edaravone. Bioorg Med Chem 28(10):115463. https://doi.org/10.1016/j.bmc.2020.115463

Bayrak BB, Yilmaz S, Hacihasanoglu Cakmak N, Yanardag R (2021) The effects of edaravone, a free radical scavenger, in lung injury induced by valproic acid demonstrated via different biochemical parameters. J Biochem Mol Toxicol 35(9):e22847. https://doi.org/10.1002/jbt.22847

Beutler E (1971) Red cell metabolism. A manual of biochemical methods, 12th edn. Academic, London

Beutler E (1975) Reduced glutathione (GSH). In: Bergmeyer HV (ed) Red blood cell metabolism. A manual of biochemical methods, 2nd edn. Grune and Stratton, New York, pp 112–114

Celik E, Tunali S, Gezginci-Oktayoglu S, Bolkent S, Can A, Yanardag R (2021) Vitamin U prevents valproic acid-induced liver injury through supporting enzymatic antioxidant system and increasing hepatocyte proliferation triggered by inflammation and apoptosis. Toxicol Mech Method 31(8):600–608. https://doi.org/10.1080/15376516.2021.1943089

Çelik Ç, Bayrak BB, Hacıhasanoğlu Çakmak N, Yanardağ R (2022) Protective effect of edaravone on rat testis after valproic acid treatment. J Res Pharm 26(1):52–62. https://doi.org/10.29228/jrp.103

Ceron JJ, Tecles F, Tvarijonaviciute A (2014) Serum paraoxonase 1 (PON1) measurement: an update. BMC Vet Res 10:74. https://doi.org/10.1186/1746-6148-10-74

Chaudhary S, Ganjoo P, Raiusddin S, Parvez S (2015) Nephroprotective activities of quercetin with potential relevance to oxidative stress induced by valproic acid. Protoplasma 252:209–217. https://doi.org/10.1007/s00709-014-0670-8

Chinard AS (1952) Photometric estimation of proline and ornithine. J Biol Chem 199:91–95

Cissell DD, Link JM, Hu JC, Athanasiou KA (2017) A modified hydroxyproline assay based on hydrochloric acid in Ehrlich's solution accurately measures tissue collagen content. Tissue Eng Part C Methods 23:243–250. https://doi.org/10.1089/ten.tec.2017.0018

Corte ED, Stirpe F (1968) Regulation of xanthine oxidase in rat liver: modifications of the enzyme activity of rat liver supernatant on storage at 20 degrees. Biochem J 108:349–351. https://doi.org/10.1042/bj1080349

Cruz MP (2018) Edaravone (Radicava): a novel neuroprotective agent for the treatment of amyotrophic lateral sclerosis. Pharm Ther 43:25–28

Dagsuyu E, Magaji UF, Sacan O, Yanardag R (2023) *Moringa oleifera* ethanolic extract prevents oxidative damage on lens caused by sodium valproate used in epilepsy treatment. Eur J Biol 82(2):289–295. https://doi.org/10.26650/EurJBiol.2023.1328517

Dash RP, Babu RJ, Srinivas NR (2018) Two decades-long journey from riluzole to edaravone: revisiting the clinical pharmacokinetics of the only two amyotrophic lateral sclerosis therapeutics. Clin Pharmacokinet 57:1385–1398. https://doi.org/10.1007/s40262-018-0655-4

Deschenes PC, Autmizguine J, Major P, Kleiber N (2020) Valproic acid induced pancreatitis presenting with decreased level of consciousness in a child with tuberous sclerosis complex. J Pediatr Pharmacol Ther 25(3):256–260. https://doi.org/10.5863/1551-6776-25.3.256

Drokov AP, Lipatova LV, Shnayder NA, Nasyrova RF (2020) Pharmacogenetic markers for metabolic impairments in treatment with valproic acid. Neurosci Behav Physiol 50:13–19. https://doi.org/10.1007/s11055-019-00861-6

El-Shenawy NS, Hamza RZ (2016) Nephrotoxicity of sodium valproate and protective role of l-cysteine in rats at biochemical and histological levels. J Basic Clin Physiol Pharmacol 27:497–504. https://doi.org/10.1515/jbcpp-2015-0106

Emekli-Alturfan E, Alev B, Tunali S, Oktay S, Tunali-Akbay T, Koc Ozturk L, Yanardag R, Yarat A (2015) Effects of edaravone on

cardiac damage in valproic acid induced toxicity. Ann Clin Lab Sci 45(2):166–172

Ertik O, Magaji UF, Sacan O, Yanardag R (2023) Effect of *Moringa oleifera* leaf extract on valproate-induced oxidative damage in muscle. Drug Chem Toxicol 46(6):1212–1222. https://doi.org/10.1080/01480545.2022.2144876

Furlong CE, Richter RJ, Seidel SJ (1988) Role of genetic polymorphism of human plasma paraoxonase/arylesterase in hydrolysis of the insecticide metabolites chlorpyrifos oxon and paraoxon. Am J Hum Gen 43:230–238

Gad AM (2018) Study on the influence of caffeic acid against sodium valproate–induced nephrotoxicity in rats. J Biochem Mol Toxicol 32:e22175. https://doi.org/10.1002/jbt.22175

Gezginci-Oktayoglu S, Turkyilmaz IB, Ercin M, Yanardag R, Bolkent S (2016) Vitamin U has a protective effect on valproic acid-induced renal damage due to its anti-oxidant, anti-inflammatory, and anti-fibrotic properties. Protoplasma 253:127–135. https://doi.org/10.1007/s00709-015-0796-3

Gyurászová M, Gurecká R, Bábíčková J, Tóthová Ľ (2020) Oxidative stress in the pathophysiology of kidney disease: implications for noninvasive monitoring and identification of biomarkers. Oxid Med Cell Longev 2020:5478708. https://doi.org/10.1155/2020/5478708

Hacihasanoglu Cakmak N, Yanardag R (2015) Edaravone, a free radical scavenger, protects liver against valproic acid induced toxicity. J Serb Chem Soc 80:627–637. https://doi.org/10.2298/JSC140812123C

Hassanein EH, Mohamed WR, Hussein RM, Arafa ESA (2023) Edaravone alleviates methotrexate-induced testicular injury in rats: implications on inflammation, steroidogenesis, and Akt/p53 signaling. Int Immunopharmacol 117:109969. https://doi.org/10.1016/j.intimp.2023.109969

Heidari R, Jafari F, Khodaei F, Shirazi Yeganeh B, Niknahad H (2018) Mechanism of valproic acid-induced Fanconi syndrome involves mitochondrial dysfunction and oxidative stress in rat kidney. Nephrol (Carlton) 23:351–361. https://doi.org/10.1111/nep.13012

Heidari R, Mandegani L, Ghanbarinejad V, Siavashpour A, Ommati MM, Azarpira N, Najibi A, Niknahad H (2019) Mitochondrial dysfunction as a mechanism involved in the pathogenesis of cirrhosis-associated cholemic nephropathy. Biomed Pharmacother 109:271–280. https://doi.org/10.1016/j.biopha.2018.10.104

Jutrić D, Đikić D, Boroš A, Odeh D, Gračan R, Beletić A, Jurčević IL (2023) Combined effects of valproate and naringin on kidney antioxidative markers and serum parameters of kidney function in C57BL6 mice. Arh Hig Rada Toksikol 74(3):218–223

Kamogawa E, Sueishi Y (2014) A multiple free-radical scavenging (MULTIS) study on the antioxidant capacity of a neuroprotective drug, edaravone as compared with uric acid, glutathione, and trolox. Bioorg Med Chem Lett 24:1376–1379. https://doi.org/10.1016/j.bmcl.2014.01.045

Kandemir FM, Ileriturk M, Gur C (2022) Rutin protects rat liver and kidney from sodium valproate-induce damage by attenuating oxidative stress, ER stress, inflammation, apoptosis and autophagy. Mol Biol Rep 49(7):6063–6074. https://doi.org/10.1007/s11033-022-07395-0

Kapucu A, Kaptan Z, Dar KA, Kaleler I, Gülay Ü (2021) Effects of erythropoietin pretreatment on liver, kidney, heart tissue in pentylentetrazol-induced seizures; evaluation in terms of oxidative markers, prolidase and sialic acid. J Ist Fac Med 84(4):464–471. https://doi.org/10.26650/IUITFD.2021.883402

Kikuchi K, Takeshige N, Miura N, Morimoto Y, Ito T, Tancharoen S, Miyata K, Kikuchi C, Iida N, Uchikado H, Miyagi N, Shiomi N, Kuramoto T, Maruyama I, Morioka M, Kawahara KI (2012) Beyond free radical scavenging: beneficial effects of edaravone (Radicut) in various diseases (review). Exp Ther Med 3:3–8. https://doi.org/10.3892/etm.2011.352

Kikuchi K, Tancharoen S, Takeshige N, Yoshitomi M, Morioka M, Murai Y, Tanaka E (2013) The efficacy of edaravone (radicut), a free radical scavenger, for cardiovascular disease. Int J Mol Sci 14(7):13909–13930. https://doi.org/10.3390/ijms140713909

Knights MJ, Finlay E (2014) The effects of sodium valproate on the renal function of children with epilepsy. Pediatr Nephrol 29:1131–1138. https://doi.org/10.1007/s00467-013-2512-x

Koike N, Sasaki A, Murakami T, Suzuki K (2021) Effect of edaravone against cisplatin-induced chronic renal injury. Drug Chem Toxicol 44(4):437–446. https://doi.org/10.1080/01480545.2019.1604740

Ledwozyw A, Michalak J, Stephien A, Kadziolka A (1986) The relationship plasma triglycerides, cholesterol, total lipids and lipid peroxidation products during human atherosclerosis. Clin Chim Acta 155:275–284

Lee M, Ahn C, Kim K, Jeung E-B (2023) Mitochondrial toxic effects of antiepileptic drug valproic acid on mouse kidney stem cells. Toxics 11(5):471. https://doi.org/10.3390/toxics11050471

Levine RL, Garland D, Oliver CN, Amici A, Climent I, Lenz AG, Ahn BW, Shaltiel S, Stadtman ER (1990) Determination of carbonyl content in oxidatively modified proteins. Method Enzymol 186:464–478. https://doi.org/10.1016/0076-6879(90)86141-H

Levine RL, Williams JA, Stadtman EP, Shacter E (1994) Carbonyl assays for determination of oxidatively modified proteins. Method Enzymol 233:346–357. https://doi.org/10.1016/S0076-6879(94)33040-9

Lin YL, Bialer M, Cabrera RM, Finnell RH, Wlodarczyk BJ (2019) Teratogenicity of valproic acid and its constitutional isomer, amide derivative valnoctamide in mice. Birth Defects Res 111:1013–1023. https://doi.org/10.1002/bdr2.1406

Liu J, Lilly MN, Shapiro JI (2018) Targeting Na/K-ATPase signaling: a new approach to control oxidative stress. Curr Pharm Des 24:359–364. https://doi.org/10.2174/1381612824666180110101052

Lowry OH, Rosebrough NJ, Farr AL, Randall RJ (1951) Protein measurement with the Folin phenol reagent. J Biol Chem 193:265–275. https://doi.org/10.1016/S0021-9258(19)52451-6

Maneenin C, Burawat J, Maneenin N, Nualkaew S, Arun S, Sampannang A, Iamsaard S (2018) Antioxidant capacity of *Momordica charantia* extract and its protective effect on testicular damage in valproic acid-induced rats. Int J Morphol 36:447–453

Maneenin C, Lapyuneyong N, Tongpan S, Yannasithinon S, Burawat J, Maneenin N, Sukhorum W, Arun S, Iamsaard S (2019) The alterations of microvasculature, tyrosine phosphorylation, and lipid peroxidation in kidney of rats treated with valproic acid. Int J Morphol 37:65–70

Mylroie AA, Collins H, Umbles C, Kyle J (1986) Erythrocyte superoxide dismutase activity and other parameters of copper status in rats ingesting lead acetate. Toxicol Appl Pharmacol 82:512–520. https://doi.org/10.1016/0041-008X(86)90286-3

Nishino T, Okamoto K, Eger BT, Pai EF, Nishino T (2008) Mammalian xanthine oxidoreductase - mechanism of transition from xanthine dehydrogenase to xanthine oxidase. FEBS J 275:3278–3289. https://doi.org/10.1111/j.1742-4658.2008.06489.x

Ojo OB, Olagunju GB, Olajide AO, Jegede ME, Fakorede AS, Crown OO, Olaleye MT, Akinmoladun AC (2022) *Spondias mombin* leaf extract ameliorates cerebral ischemia/reperfusion-induced cardiohepatorenal oxidative stress in rats. Phytomed Plus 2(1):100196. https://doi.org/10.1016/j.phyplu.2021.100196

Oktay S, Alev B, Tunali S, Emekli-Alturfan E, Tunali-Akbay T, Koc Ozturk L, Yanardag R, Yarat A (2015) Edaravone ameliorates the adverse effects of valproic acid toxicity in small intestine. Hum Exp Toxicol 34:654–661. https://doi.org/10.1177/0960327114554047

Oktay S, Alev B, Koc Ozturk L, Tunali S, Demirel S, Emekli-Alturfan E, Tunali-Akbay T, Akyuz S, Yanardag R, Yarat A (2016) Edaravone ameliorates valproate-induced gingival toxicity by reducing oxidative-stress, inflammation and tissue damage. Marmara Pharm J 20:232–240. https://doi.org/10.12991/mpj.20162096267

Oktay S, Alev-Tüzüner B, Tunali S, Ak E, Emekli-Alturfan E, Tunali-Akbay T, Koç-Öztürk L, Çetinel Ş, Yanardag R, Yarat A (2017) Investigation of the effects of edaravone on valproic acid induced tissue damage in pancreas. Marmara Pharm J 21:570–577. https://doi.org/10.12991/marupj.319312

Oztay F, Tunali S, Kayalar O, Yanardag R (2020) The protective effect of vitamin U on valproic acid-induced lung toxicity in rats via amelioration of oxidative stress. J Biochem Mol Toxicol 34(12):e22602. https://doi.org/10.1002/jbt.22602

Oztopuz O, Turkon H, Buyuk B, Coskun O, Sehitoglu MH, Ovali MA, Uzun M (2020) Melatonin ameliorates sodium valproate-induced hepatotoxicity in rats. Mol Biol Rep 47:317–325. https://doi.org/10.1007/s11033-019-05134-6

Paglia DE, Valentine WN (1967) Studies on the quantitative and qualitative characterization of erythrocyte glutathione peroxidase. J Lab Clin Med 70:158–169. https://doi.org/10.5555/uri:pii:0022214367900765

Permpongpaiboon T, Nagila A, Pidetcha P, Tuangmungsakulchai K, Tantrarongroj S, Porntadavity S (2011) Decreased paraoxonase 1 activity and increased oxidative stress in low lead-exposed workers. Hum Exp Toxicol 30:1196–1203. https://doi.org/10.1177/0960327110388536

Petrushanko IY, Yakushev S, Mitkevich VA, Kamanina YV, Ziganshin RH, Meng X, Anashkina AA, Makhro A, Lopina OD, Gassmann M, Makarov AA, Bogdanova A (2012) S-glutathionylation of the Na,K-ATPase catalytic α subunit is a determinant of the enzyme redox sensitivity. J Biol Chem 287:32195–32205. https://doi.org/10.1074/jbc.M112.391094

Pirinççi N, Kaba M, Geçit İ, Güneş M, Yüksel MB, Tanık S, Arslan A, Demir H (2016) Serum prolidase activity, oxidative stress, and antioxidant enzyme levels in patients with renal cell carcinoma. Toxicol Indust Health 32:193–999. https://doi.org/10.1177/0748233713498924

Ratliff BB, Abdulmahdi W, Pawar R, Wolin MS (2016) Oxidant mechanisms in renal injury and disease. Antioxid Redox Signal 25(3):119–146. https://doi.org/10.1089/ars.2016.6665

Reedy GK, Enwemeka CS (1996) A simplified method for the analysis of hydroxyproline in biological tissues. Clin Biochem 29:225–229. https://doi.org/10.1016/0009-9120(96)00003-6

Richens A, Ahmad S (1975) Controlled trial of sodium valproate in severe epilepsy. Br Med J 4:255–256. https://doi.org/10.1136/bmj.4.5991.255

Ridderstap AS, Bonting SL (1969) Na^+-K^+-activated ATPase and exocrine pancreatic secretion in vitro. Am J Physiol 217:1721–1727. https://doi.org/10.1152/ajplegacy.1969.217.6.1721

Sharma S, Kaur T, Sharma AK, Singh B, Pathak D, Yadav HN, Singh AP (2021) Betaine attenuates sodium arsenite-induced renal dysfunction in rats. Drug Chem Toxicol 45(6):2488–2498. https://doi.org/10.1080/01480545.2021.1959699

Sokmen BB, Tunali S, Yanardag R (2012) Effects of vitamin U (S-methyl methionine sulphonium chloride) on valproic acid induced liver injury in rats. Food Chem Toxicol 50:3562–3566. https://doi.org/10.1016/j.fct.2012.07.056

Tavafi M, Ahmadvand H, Tamjidipour A, Hasanvan A (2020) Rosmarinic acid ameliorates renal ischemia reperfusion damage in rats. J Nephropharmacol 9:e15. https://doi.org/10.15171/npj.2020.15

Tunali S (2014) The effects of vitamin B6 on lens antioxidant system in valproic acid-administered rats. Hum Exp Toxicol 33:623–628. https://doi.org/10.1177/0960327113506233

Tunali-Akbay T, Alev B, Tunali S, Oktay S, Emekli-Alturfan E, Ipekci H, Ozturk LK, Yanardag R, Yarat A (2017) Protective role of edaravone against valproic acid induced changes in skin. Indian J Exp Biol 55:286–291

Türe E, Yazar A (2020) Sodium valproate and levetiracetam treatment in children: their effects on serum paraoxonase/arylesterase activities. Techniques Neurosurg Neurol 3(3). https://doi.org/10.31031/TNN.2020.03.000561. TNN.000561.2020

Turkyilmaz IB, Altas N, Arisan I, Yanardag R (2021) Effect of vitamin B_6 on brain damage in valproic acid induced toxicity. J Biochem Mol Toxicol 35(9):e22855. https://doi.org/10.1002/jbt.22855

Verma AK, Chandra S, Singh RG, Singh TB, Srivastava S, Srivastava R (2014) Serum prolidase activity and oxidative stress in diabetic nephropathy and end stage renal disease: a correlative study with glucose and creatinine. Biochem Res Int 2014:291458. https://doi.org/10.1155/2014/291458

Watanabe K, Tanaka M, Yuki S, Hirai M, Yamamoto Y (2018) How is edaravone effective against acute ischemic stroke and amyotrophic lateral sclerosis? J Clin Biochem Nutr 2018(62):20–38. https://doi.org/10.3164/jcbn.17-62

Wei H, Frenkel K (1991) In vivo formation of oxidized DNA bases in tumor promoter-treated mouse skin. Cancer Res 51(16):4443–4449

Wilk P, Wator E, Weiss MS (2021) Prolidase– a protein with many faces. Biochimie 183:3–12. https://doi.org/10.1016/j.biochi.2020.09.017

Witko-Sarsat V, Friedlander M, Capeillère-Blandin C, Nguyen-Khoa T, Nguyen AT, Zingraff J, Jungers P, Béatrice Descamps B (1996) Advanced oxidation protein products as a novel marker of oxidative stress in uremia. Kidney Int 49:1304–1313. https://doi.org/10.1038/ki.1996.186

Wu MY, Chang FY, Ke JY, Chen CS, Lin PC, Wang TS (2020) Valproic acid-induced hyperammonemic encephalopathy in a patient with bipolar disorder: a case report. Brain Sci 10:E187. https://doi.org/10.3390/brainsci10030187

Yang Z, Zhao Y, He Y, Liu W, Yu Q (2023) Pyroptosis mediated renal injury caused by chronic intermittent hypoxia and the intervention effect of edaravone in rats. Chin J Clin Pharmacol Ther 28(1):10–18. https://doi.org/10.12092/j.issn.1009-2501.2023.01.002

Zeng Y, Zhu G, Zhu M, Song J, Cai H, Song Y, Wang J, Jin M (2022) Edaravone attenuated particulate matter-induced lung inflammation by inhibiting ROS-NF-κB signaling pathway. Oxid Med Cell Longev 2022:6990884. https://doi.org/10.1155/2022/6908884

Zhao X, Zhang E, Ren X, Bai X, Wang D, Bai L, Luo D, Guo Z, Wang Q, Yang J (2020) Edaravone alleviates cell apoptosis and mitochondrial injury in ischemia–reperfusion-induced kidney injury via the JAK/STAT pathway. Biol Res 53(1):28. https://doi.org/10.1186/s40659-020-00297-0