Pakistan Veterinary Journal

ISSN: 0253-8318 (PRINT), 2074-7764 (ONLINE)
DOI: 10.29261/pakvetj/2024.153

RESEARCH ARTICLE

Protective Effects of Selenium against Acrylamide-Induced Hepatotoxicity in Rats

Mehmet Enes Sozen[1*], Hasan Basri Savas[2] and Gokhan Cuce[3]

[1]Alanya Alaaddin Keykubat University, School of Medicine, Department of Histology and Embryology. Antalya, Turkiye; [2]Mardin Artuklu University, School of Medicine, Department of Medical Biochemistry. Mardin, Turkiye; [3]Necmettin Erbakan University, Meram School of Medicine, Department of Histology and Embryology. Konya, Turkiye
*Corresponding author: enes.sozen@alanya.edu.tr

ARTICLE HISTORY (23-396)

Received: September 9, 2023
Revised: February 26, 2024
Accepted: February 28, 2024
Published online: April 01, 2024

Key words:
Acrylamide
Selenium
Apoptosis
Oxidative damage.

A B S T R A C T

Acrylamide (ACR) is an organic chemical widely consumed worldwide, depending on the diet. ACR has toxic effects on the liver and other organs due to oxidative damage. The research is aimed to determine the effects of Selenium (Se) against ACR toxicity. 32 Wistar albino male rats were divided into Control, ACR, Se, and ACR+Se groups. After slaughter on the 28th day, the blood samples taken from the animals were tested for total oxidant status (TOS) and total antioxidant status (TAS) to assess oxidative stress. The liver tissue sections were evaluated for lymphocyte infiltration, hepatocyte degeneration, sinusoid dilatation, and congestion. IL-6, Bax, and Bcl-2 expression were evaluated with immunohistochemistry. While the ACR group's TOS and oxidative stress index (OSI) values were significantly higher than the control group's, there was no significant difference in the ACR+Se group's TOS and OSI values. The ACR group had a considerably higher histopathological score than the other groups. ACR increased IL-6, and Bax levels and decreased Bcl-2 levels compared to the control, Se, and ACR+Se groups. ACR increased oxidative stress significantly caused toxic effects, inflammation, and cell death in the liver. On the other hand, Se oral supplementation may protect against oxidative stress, toxic effects, inflammation, and cell death induced by ACR in the liver.

To Cite This Article: Sozen ME, Savas HB and Cuce G, 2024. Protective effects of selenium against acrylamide-induced hepatotoxicity in rats. Pak Vet J, 44(2): 274-279. http://dx.doi.org/10.29261/pakvetj/2024.153

INTRODUCTION

Acrylamide (ACR) occurs with the Maillard reaction and is caused by asparagines and sugar reacting, giving the food a brown color and distinctive flavor when cooked at high temperatures. ACR is formed while frying, baking, and roasting. Therefore, it can be found in bread, baked goods, biscuits, toast, cereals, breakfast cereals, french fries, and potato chips. Due to the prevalence of ACR in a wide range of consumed products, there is a significant risk of dietary exposure (Semla *et al*., 2017).

ACR causes toxic effects by reacting with DNA, hemoglobin, and enzymes in the circulatory system and various target organs (Ibrahim and Ibrahem 2020). The liver is vital for ACR metabolism and toxicity. ACR undergoes oxidative biotransformation by the enzyme cytochrome P450 (CYP2E1) and is converted to glycidamide, an epoxide derivative (Chen *et al.,* 2020). It has been shown that ACR causes injury in the brain, lungs, liver, kidney, heart, and testicles due to increased oxidative stress (Ghorbel *et al.,* 2015a; Semla *et al*., 2017; Koszucka *et al.,* 2020; Ghasemzadeh *et al.,* 2021). ACR also decreases antioxidant capacity, and trace element levels (Catalgol *et al.,* 2009; Yerlikaya and Yener 2013; Cerrah *et al.,* 2023).

Selenium (Se) is an essential trace element, that is vital in regulating the antioxidant system and it has been reported that ACR exposure decreased Se levels (Hayes *et al.,* 2020). Se takes place in the structure of glutathione peroxidase, which catalyzes the reduction reaction of glutathione and ensures its oxidation (Kiełczykowska *et al.,* 2018). Se also eliminates hydrogen peroxide and fatty acid hydroperoxides. Thus, Se contributes significantly to the viability and integrity of cells by protecting membrane lipids and helps prevent cell damage caused by free radicals (Ali *et al.,* 2014). For these reasons, Se may act as an essential protector in preventing oxidative stress and damage induced by ACR exposure.

Apoptosis is a cell-suicide program that maintains tissue homeostasis. Apoptosis is required for various activities, including cell turnover, proper development, and chemical-induced cell death. Bax and Bcl-2 are members of the Bcl-2 protein family, which regulates apoptosis. Bax promotes apoptosis by triggering

mitochondrial cytochrome c release, whereas Bcl-2 inhibits it. The balance between Bax and Bcl-2 is critical in deciding whether a cell dies or survives (Elmore, 2007).

The present research aims to demonstrate the potential protective effects of Se against ACR toxicity in the liver and blood in a rat toxicity model.

MATERIALS AND METHODS

Study design: This experimental research received ethical approval from the SDU Animal Experiments Local Ethics Committee (decision 17/06, October 23, 2018). Thirty-two male Wistar-Albino rats, sourced from the SDU Experimental Animals Laboratory, were utilized for the study. The rats, aged 9-10 weeks, were stratified into four groups: Control, ACR, Se, and ACR+Se, and subjected to a 28-day experimental protocol. In the Control Group (n=8), oral administration of 1 ml saline solution occurred from day 0 to day 27. The ACR Group (n=8) received an oral solution of 20 mg/kg ACR dissolved in 0.5 cc saline over the same period. The Se Group (n=8) was orally administered a solution of 0.1 mg/kg Se, dissolved in 0.5 cc saline, from day 0 to day 27. The ACR+Se Group (n=8) underwent oral gavage of 20 mg/kg ACR (0.5 cc) and 0.1 mg/kg Se (0.5 cc) from day 0 to day 27. Intraperitoneal induction of general anesthesia (Ketamine 80-100 mg/kg/Xylazine 8-10 mg/kg) was conducted in all animals 24 hours post the final ACR administration (day 28). Following an abdominal incision, exsanguination was carried out by withdrawing blood from the Inferior Vena Cava.

Methods of biochemical analysis: Blood cell count analysis was performed with whole blood in tubes containing Ethylenediaminetetraacetic acid (EDTA). Lymphocytes, leukocytes, and neutrophils were counted. Whole blood taken gel biochemistry tubes were cooled and then centrifuged at 4,000 g for 10 minutes to obtain serum samples (Thermo Scientific SL 40R, USA). C reactive protein (CRP), Alanine aminotransferase (ALT), Aspartate aminotransferase (AST), and glucose levels were measured on an Abbott Architect c8000 clinical chemistry autoanalyzer using commercial kits (Abbott, Abbott Park, Illinois, USA). Commercial kits (Rel Assay Diagnostics, Mega Tip, Gaziantep, Turkey) based on a colorimetric method were then used to measure total oxidant status (TOS) (RL0024) and total antioxidant status (TAS) (RL0017) levels by a microplate reader (Biotek Synergy H1, USA). The TOS/TAS ratio was used to calculate the oxidative stress index (OSI) (Savas and Sayar 2021).

Histopathological evaluation of liver: Liver tissues were examined macroscopically after opening the abdomen, fixed, and embedded after processing. 5 µm thick sections were cut and stained with Hematoxylin and Eosin (H&E). Morphological changes were evaluated via a light microscope Nikon H5505 and DS-FI2 analysis system. Hepatocyte degeneration, sinusoid dilatation, lymphocyte infiltration, and congestion were evaluated in the sections. Histopathological changes were scored for each criterion (as absent =0, mild =1, moderate=2, or severe=3).

Immunohistochemistry: Xylene was used for deparaffinization for 30 min. After antigen retrieval, endogenous peroxidase activity was inhibited with 3% hydrogen peroxide ($3\%H_2O_2$). Super Block (ScyTek Laboratories, Logan, UT) prevented nonspecific antigen binding for 10 min. The sections were incubated for 60 min with Interleukin 6 (IL-6) (ab208113, Abcam, UK), Anti-B-cell lymphoma 2 (Bcl-2) (PA5-27094, Thermo Fisher Scientific, WA, USA), and Anti-Bcl-2-associated X protein (Bax) (ab7977, Abcam, UK) primary antibodies. The sections were treated with a secondary antibody for 20 min. Streptavidin-peroxidase was used to detect antigen-antibody complex for 20 min. 3-Amino-9-Ethylcarbazole (AEC) chromogen was added for 15 min. Mayer's hematoxylin was used for counterstaining for 5 min. Then sections staining were evaluated following criteria (0 expresses=no; 1 expresses=weak; 2 expresses=moderate; 3 expresses=intense).

Statistical analysis: For histological and biochemical examination, significant data were compared using one-way analysis of variance (ANOVA) with the Tukey post hoc test. The immunohistochemical analysis results were compared with the Kruskal–Wallis test. Differences between the pairs of groups were assessed with the Mann–Whitney U test. A p-value under 0.05 was considered statistically significant for all findings.

RESULTS

Biochemical parameters: TAS values in the Se group were significantly higher than the control group ($p=0.01$). There was no significant difference in the ACR+Se and ACR group between the control group in terms of TAS values ($p>0.05$). TOS ($p=0.003$) and OSI ($p=0.006$) values were significantly higher in the ACR group compared to the control group. There was no significant difference in the Se and ACR+Se groups compared to the control group in terms of TOS and OSI values ($p> 0.05$). In the ACR group, there was a statistically significant increase in AST and ALT compared to the control group, but AST and ALT levels were significantly lower in the ACR+Se group ($p<0.0001$). Although glucose levels were slightly elevated in the ACR group compared to the control group, glucose levels decreased significantly in the ACR+Se group ($p<0.022$) (Table 1).

Histopathological evaluation: Liver sections were used to examine histopathological changes. The typical histological structure was seen in the control and Se group. The histopathological score was significantly higher in the ACR group compared to other groups ($p<0.05$). The ACR+Se group had a significantly higher histopathological score than the control and Se groups ($p<0.05$), and there were no differences between the control and Se groups (Fig. 1 and 2).

Immunohistochemistry: ACR caused increased expression of IL-6 (Table 2, Fig. 3) compared to the other groups. IL-6 expression was significantly higher in the ACR+Se group compared to the control group. Also, no statistically significant difference between the control and the Se group was observed. ACR caused increased expression of Bax (Table 3, Fig. 3) compared to the other groups.

Table 1: Comparison of Biochemical and Oxidative Stress Parameters

Parameters	Units	Control; mean±SD	ACR; mean±SD	Se; mean±SD	ACR+Se; mean±SD	p
Lymphocytes	Cells per microliter of blood	1.779±0.899	1.488±0.726	2.713±1.269	1.295±0.665	0.022
Neutrophils	Cells per microliter of blood	1.364±0.578	1.188±0.712	1.816±1.025	0.804±0.548	0.074
Neutrophils/ Lymphocytes	Ratio	0.841±0.307	0.840±0.296	0.714±0.301	0.690±0.467	0.736
Leukocytes	Cells per microliter of blood	3.275±1.509	2.885±1.473	4.714±2.101	2.528±1.151 !	0.053
CRP	mg/l	0.380±0.047	0.490±0.112	0.480±0.089	0.463±0.151	0.175
Glucose	mg/dl	214.550±21.200	*245.025±30.708*	*189.800±27.881* #	191.800±59.331	0.022
AST	IU/l	55.300±2.435	76.300±7.344 *	63.80±2.659 *#	66.80±3.495 * #	0.0001
ALT	U/l	27.550±7.270	39.300±5.726 *	27.85±5.42 #	21.10±4.25 #	0.000
Total Antioxidant Status	mmol Trolox equivalent/l	0.909±0.06	0.991±0.12	1.122±0.177 #	0.988±0.07	0.01
Total Oxidant Status	µmol H_2O_2 equivalent/l	27.164±2.523	37.887±2.355 *	31.179±7.191	29.381±7.376	0.003
Oxidative Stress Index	Ratio	30.00±3.376	39.023±7.547 *	28.339±6.919	29.444±6.025	0.006

Acrylamide (ACR), Selenium (Se), Alanine aminotransferase (ALT) and aspartate aminotransferase (AST); *: Statistically significant difference with the control group ($p<0.05$); #: Statistically significant difference with the acrylamide group ($p<0.05$); !: Statistically significant difference with the selenium group ($p<0.05$).

Table 2: Immunohistochemical analysis of liver IL-6 staining

Group	n	Mean	Median	25%	75%	Min	Max
Control	8	0.25	0	0	0.75	0	1
Se	8	0.37	0	0	0.75	0	2
ACR	8	2.75	3	2.25	3	2	3
ACR+Se	8	0.87	1	1	1	0	1

A significant difference in the IL-6 was detected between the Control group- Acrylamide (ACR) group, the Control group - ACR+Selenium (Se) group, the Se group- ACR group, and between the ACR group and the ACR+Se group ($p<0.05$).

Table 3: Immunohistochemical analysis of liver Bax staining

Group	n	Mean	Median	25%	75%	Min	Max
Control	8	0.25	0	0	0.75	0	1
Se	8	0.12	0	0	0	0	1
ACR	8	2.87	3	3	3	2	3
ACR+Se	8	1	1	1	1	0	2

A significant difference in the Bax was detected between the Control group- Acrylamide (ACR) group, Control group- ACR+Selenium (Se) group, Se group- ACR group, Se group- ACR +Se group, and between the ACR group and ACR+Se group ($p<0.05$).

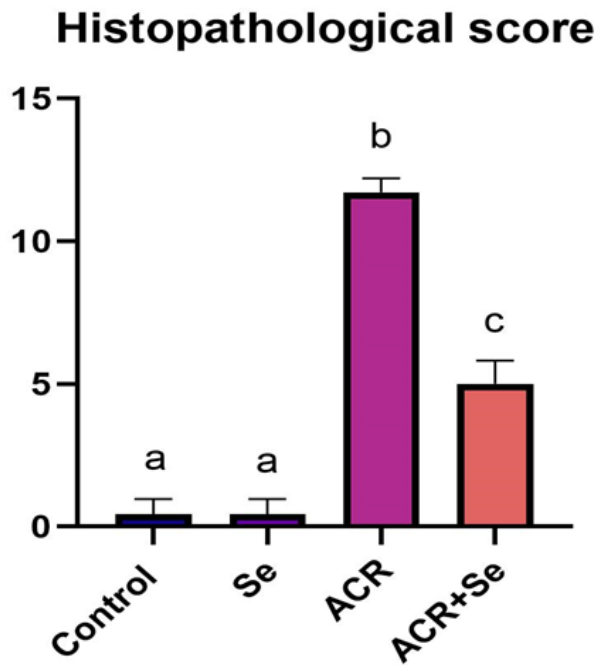

Fig. 1: Histopathological scores of liver tissue that consists of hepatocyte degeneration, sinusoid dilatation, lymphocyte infiltration, and congestion. Different letters indicate statistical significance ($p<0.05$). Compared by one-way analysis of variance (ANOVA) with the Tukey post hoc test.

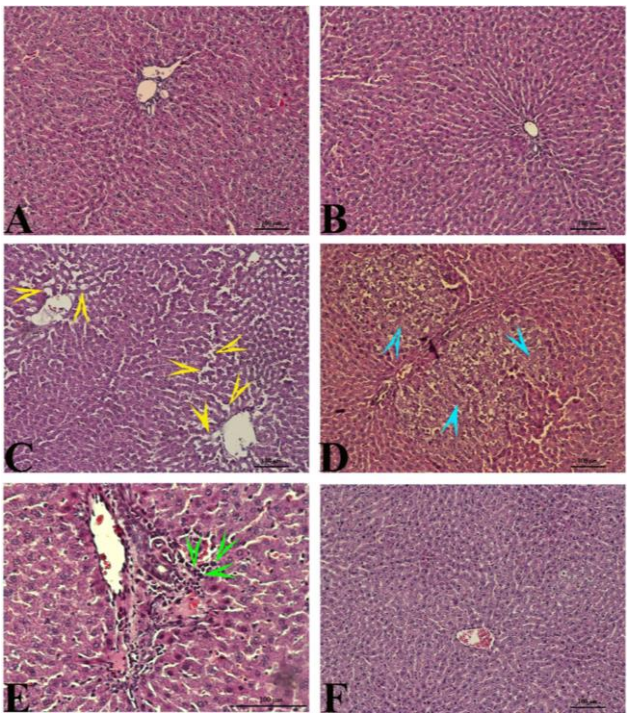

Fig. 2: Hematoxylin-Eosin (H&E)-stained liver sections. A: Control group: No significant pathological condition is observed in this group. It represents the normal liver morphology. B: Selenium (Se) Group: This group exhibits a morphology similar to the control group. C-D-E Acrylamide (ACR) Group: Histopathological changes caused by the ACR exposure. C: Blonde arrows indicate sinusoid dilatation. D: Blue arrows indicate; hepatocyte degeneration. E: Green arrows indicate; lymphocyte infiltration. F: ACR+Se Group: there may be a protective effect of selenium against acrylamide effects. Scale bar = 100 µm.

Bax expression was significantly higher in the ACR+Se group compared to the control and Se groups. Also, no statistically significant difference between the control and Se group was observed. ACR caused decreased expression of Bcl-2 (Table 4, Fig. 3) compared to the control, Se, and ACR+Se groups.

DISCUSSION

ACR is an important chemical that can lead to a serious increase in oxidative stress and toxic effects due to widespread exposure through nutrition. On the other hand,

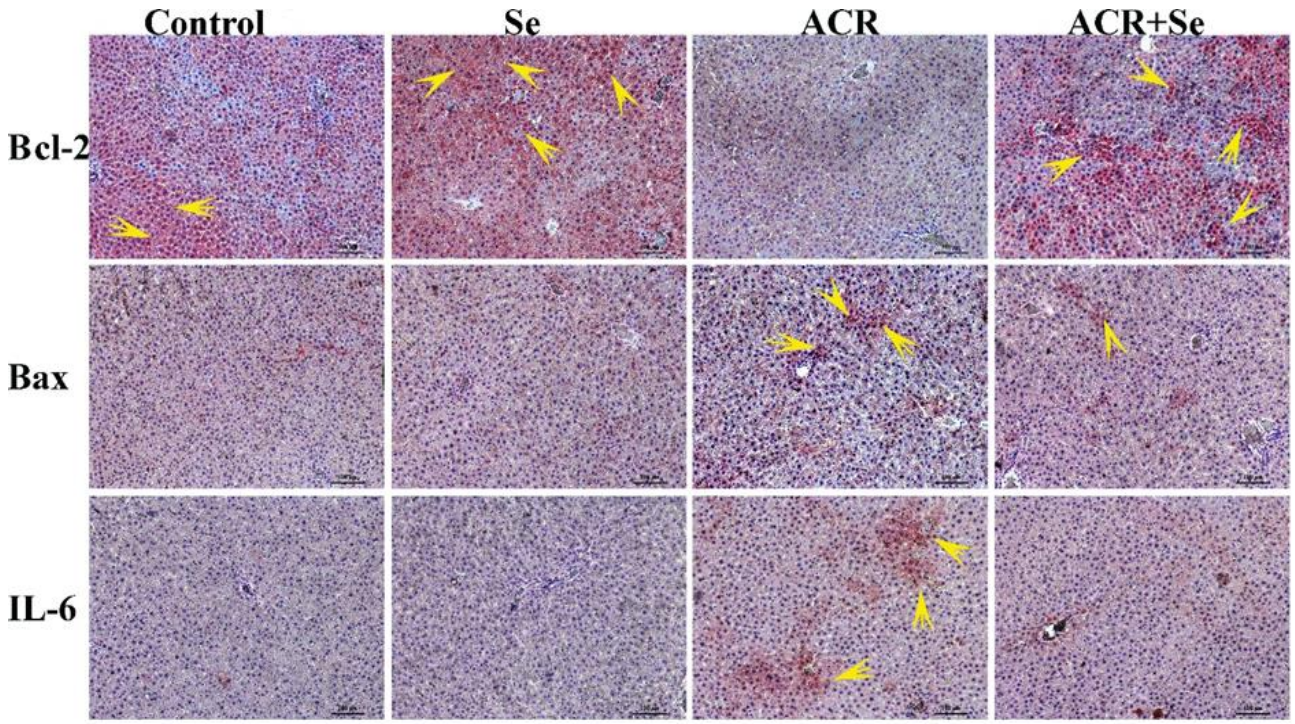

Fig. 3: Liver sections of immunohistochemical staining of Bcl-2, Bax, and IL-6.

Table 4: Immunohistochemical analysis of liver Bcl-2 staining

Group	n	Mean	Median	25%	75%	Min	Max
Control	8	2.87	3	3	3	2	3
Se	8	2.75	3	2.25	3	2	3
ACR	8	1.12	1	1	1	1	2
ACR+Se	8	2.37	2	2	3	2	3

A significant difference in the Bcl-2 was detected between the Control group- Acrylamide (ACR) group, the Selenium (Se) group- ACR group, and between the ACR group and the ACR+Se group ($p < 0.05$).

Se is a trace element with essential roles in the antioxidant system and positively affects immunity. For this reason, Se might protect against the toxic effects of ACR. Our experimental toxicity model aims to demonstrate the possible protective effects of Se, against the potential toxic effects of oral ACR exposure on the liver in rats.

ACR results in a variety of histological abnormalities in the liver (Gedik *et al.,* 2017; Acaroz *et al.,* 2018; Belhadj *et al.,* 2019). ACR causes hepatocyte degeneration, congestion, and lymphocyte infiltration (Belhadj *et al.,* 2019), necrosis (Gedik *et al.,* 2017), sinusoidal dilatation, hyperemia, and Kupffer cell activation (Acaroz *et al.,* 2018). The histopathological examination of the liver demonstrates that the ACR+Se group has a decreased histopathological score compared to the ACR group. It's thought that Se has a protective effect against ACR toxicity in the liver. These findings are in accordance with other studies that antioxidant substances were used against ACR toxicity and the damage caused by ACR in the liver was ameliorated by antioxidant substances (Altinoz *et al.,* 2015; Ansar *et al.,* 2016; Gedik *et al.,* 2017; Acaroz *et al.,* 2018).

Bax is a proapoptotic effector protein that disrupts the mitochondrial outer membrane and causes a pore formation. The proapoptotic factors like cytochrome c are released into the cytosol by these pores. Then, these factors cause activation of the caspase cascade and lead to cell death. Bcl-2 is an antiapoptotic effector protein that binds and inhibits Bax and suppresses cell death (Peña-Blanco and García-Sáez 2018). Our results showed that ACR increased Bax levels and decreased Bcl-2 levels, indicating increased cell death. Our findings are consistent with other studies, which showed that ACR has apoptotic effects in hepatocytes (Seydi *et al.,* 2015; Ghasemzadeh *et al.,* 2021). It has been found that 50 mg/kg i.p. ACR exposure caused apoptosis in the liver for 11 days in the liver and 0.6 mg/kg i.p. Se or 200 mg/kg vitamin E supplementation decreased this effect (Ghasemzadeh *et al.,* 2021). In the present study, although there was a lower dose of ACR exposure, ACR caused apoptosis in the liver. The reason for this may be the supplementation of ACR at a low dose but for a longer period in our study. On the other hand, ACR+Se decreased Bax levels and increased Bcl-2 levels compared to ACR group; these findings indicate cell survival. In this way, it's found that Se suppresses cell death caused by ACR.

IL-6 is a proinflammatory cytokine and plays a vital role in inflammation (Drutskaya *et al.,* 2018). Results of this study showed that ACR increased the IL-6 levels and caused inflammation, and we know that suppressing inflammation in the liver decreases liver injury (Calis *et al.,* 2022; Dasdelen *et al.,* 2023). Like other studies (Alturfan *et al.,* 2012; Ghorbel *et al.,* 2015b; Elhelaly *et al.,* 2019; Cerrah *et al.,* 2023), our results showed that ACR increased IL-6 concentrations. In our study, Se supplementation decreased IL-6 levels and suppressed inflammation caused by ACR in the liver.

About 90% of inhaled oxygen is used for oxidative phosphorylation in humans. The oxygen molecule is a strong oxidizer. Oxygen routinely forms some superoxide, hydrogen peroxide, and hydroxyl radicals during metabolic processes. The organism's antioxidant system tries to eliminate these radicals' harmful effects. If this balance between the oxidant-antioxidant system is disturbed, oxidative stress causes damage to cells and tissues. The measurement of TOS, TAS, and OSI values

are important to indicate oxidative damage. OSI increase can be evaluated as an indicator of significant oxidative stress and damage (Seydi *et al.,* 2015). It has been reported that 20 mg/kg oral ACR supplementation increased liver TOS levels and decreased TAS levels (Cerrah *et al.,* 2023). In parallel with this study, it was seen in our research that the ACR group caused significantly higher OSI in blood compared to the control group (p=0.032). With the Se supplementation, the OSI values were decreased to a value lower than the control group, and the statistically significant difference with the control group disappeared (p=0.998). For this reason, it can be said that ACR causes a significant increase in the OSI, and additional Se balances the increase in oxidative stress.

In a study, Se significantly increased the levels of glutathione (GSH) and glutathione peroxidase (GPx) and decreased the levels of malondialdehyde (MDA) compared to the group that received only ACR (Teodor *et al.,* 2011). This study in the literature supports our findings in terms of oxidative stress reduction and antioxidant capacity increase with the addition of Se in ACR toxicity, although the measured parameters are different. In a recent study, ACR induced oxidative stress; in the same way, these were suppressed by high doses of Se (Sengul *et al.,* 2021). In this respect, the results of the research support our findings of increased oxidative stress, although different parameters have been measured. A recent experimental study demonstrated that Se is an essential micronutrient with significant antioxidant activity that may be beneficial in reducing ACR formation during high-temperature roasting (Alafeef *et al.,* 2020). These results support our research. A similar recent study revealed that Se nanoparticles (Ch-SeNPs) stabilized with chitosan have strong antioxidant effects, can function stably, and have a protective potential against ACR toxicity in rats (Khiralla *et al.,* 2020). Considering the results of our research showing the properties of Se to reduce ACR toxicity and protect the liver, the application of Se to foods containing ACR in limited numbers should be increased and expanded.

It has been shown that ACR causes an increase in oxidative stress and decreases antioxidant capacity, and naringin reduces the oxidative stress induced by ACR (Gelen *et al.,* 2022). These research results are consistent with our results.

ALT and aspartate AST are general indicators of liver tissue damage. The fact that the AST and ALT values measured in our study increased in the ACR group with a statistically significant difference with the control group and decreased with the addition of Se, indicating that ACR causes liver tissue damage, and Se supplementation has a positive effect on preventing this damage. In this respect, our research is compatible with the literature (Teodor *et al.,* 2011; Sengul *et al.,* 2021).

As it can be understood from the articles we compared in the discussion, the protective role of Se in ACR toxicity has been investigated before (Teodor *et al.,* 2011). However, our research's original aspect is examining the liver tissue for histopathological changes. Unlike the previous ones, it provides a comprehensive evaluation opportunity by measuring TAS and TOS, a more general indicator, and calculating the OSI instead of measuring a few of the antioxidant enzymes and oxidative damage products.

Conclusions: The histopathological and biochemical evaluation showed that ACR has toxic effects on the liver, and Se supplementation decreases these toxic effects. ACR appears to increase oxidative stress and produce toxic effects significantly. Se is known to be a powerful antioxidant trace element. Oxidative stress has an important role in forming damage and diseases in ACR toxicity. Our research showed that Se has a protective effect on the liver against ACR toxicity. Se supplementation may be protective against toxic effects of ACR such as oxidative stress, inflammation, and apoptosis. Our data should be supported by further clinical studies on volunteers with identified dietary ACR exposure to demonstrate the antioxidant, anti-apoptotic, and anti-inflammatory effects of Se in humans. New research should be conducted for this purpose.

Ethical issue: This experimental research was carried out in the licensed Süleyman Demirel University Animal Experiments Laboratory, with the permission of the Ethics Committee to be obtained from the Animal Experiments Local Ethics Committee of the Rectorate of Süleyman Demirel University (decision 17/06, October 23, 218).

Conflicts of interest: The authors declare no conflict of interest.

Acknowledgments: The authors gratefully acknowledge the Alanya Alaaddin Keykubat University Scientific Research Projects Coordination Office for financial support and Halil Ascı, Mehtap Savran, Fatma Nihan Cankara, İbrahim Aydın Candan, and Yasemin Sahin for contributions. Alanya Alaaddin Keykubat University, Scientific Research Projects Coordination Office, supported this project (Project No: 2019-04-01-MAP03). The preliminary data of this research were presented in summary at the International Harran Health Sciences Congress and 4th International Hippocrates Congress on Medical and Health Sciences in 2020.

Authors contribution: HBS and MES conceived and designed the study. HBS and MES executed the experiment. HBS analyzed the sera. MES and GC analyzed tissue samples. All authors analyzed the results, the paper was critically edited for key intellectual content, and the final version was approved.

REFERENCES

Acaroz U, Ince S, Arslan-Acaroz D, *et al.,* 2018. The ameliorative effects of boron against acrylamide-induced oxidative stress, inflammatory response, and metabolic changes in rats. Food Chem Toxicol 118: 745-752.

Alafeef AK, Ariffin F and Zulkurnain M, 2020. Organic Selenium as antioxidant additive in mitigating acrylamide in coffee beans roasted via conventional and superheated steam. Foods 9: 1197.

Ali MA, Aly EM and Elawady AI, 2014. Effectiveness of selenium on acrylamide toxicity to retina. Int J Ophthalmol 7: 614-620.

Altinoz E, Turkoz Y and Vardi N, 2015. The protective effect of N-acetylcysteine against acrylamide toxicity in liver and small and large intestine tissues. Bratisl Lek Listy 116: 252-258.

Alturfan AA, Tozan-Beceren A, Sehirli AO, *et al.,* 2012. Resveratrol

ameliorates oxidative DNA damage and protects against acrylamide-induced oxidative stress in rats. Mol Biol Rep 39: 4589-4596.

Ansar S, Siddiqi NJ, Zargar S, *et al.,* 2016. Hepatoprotective effect of Quercetin supplementation against Acrylamide-induced DNA damage in Wistar rats. BMC Complement Altern Med 16: 327.

Belhadj BA, Dilmi BA, Mezaini A, *et al.,* 2019. Effect of oral exposure to acrylamide on biochemical and hematologic parameters in Wistar rats. Drug Chem Toxicol 42: 157-166.

Calis Z, Dasdelen D, Baltaci AK, *et al.,* 2022. Naringenin prevents inflammation, apoptosis and DNA damage in Potassium Oxonate-induced hyperuricemia in rat liver tissue: Roles of Cytochrome C, NF-κB, Caspase-3 and 8-Hydroxydeoxyguanosine. Metab Syndr Relat Disord 20: 473-479.

Catalgol B, Özhan G and Alpertunga B, 2009. Acrylamide-induced oxidative stress in human erythrocytes. Human & Experimental Toxicology 28: 611-617.

Cerrah S, Ozcicek F, Gundogdu B, *et al.,* 2023. Carvacrol prevents acrylamide-induced oxidative and inflammatory liver damage and dysfunction in rats. Front Pharmacol 14: 1161448.

Chen X, Jia W, Wang Q, *et al.,* 2020. Protective effect of a dietary flavonoid-rich antioxidant from bamboo leaves against internal exposure to acrylamide and glycidamide in humans, Food Funct 11: 7000–7011.

Dasdelen D, Cetin N, Menevse E, *et al.,* 2023. Effects of putrescine on oxidative stress, spermidine/spermine-N(1)-acetyltransferase, inflammation and energy levels in liver and serum in rats with brain ischemia-reperfusion. Physiol Int 110: 34-45.

Drutskaya MS, Gogoleva VS, Atretkhany KN, *et al.,* 2018. Proinflammatory and immunoregulatory functions of Interleukin 6 as identified by reverse genetics. Mol Biol 52: 963-974.

Elhelaly AE, AlBasher G, Alfarraj S, *et al.,* 2019. Protective effects of hesperidin and diosmin against acrylamide-induced liver, kidney, and brain oxidative damage in rats. Environ Sci Pollut Res Int 26: 35151-35162.

Elmore S, 2007. Apoptosis: a review of programmed cell death. Toxicol Pathol 35: 495-516.

Gedik S, Erdemli ME, Gul M, *et al.,* 2017. Hepatoprotective effects of crocin on biochemical and histopathological alterations following acrylamide-induced liver injury in Wistar rats. Biomed Pharmacother 95: 764-770.

Gelen V, Yıldırım S, Şengül E, *et al.,* 2022. Naringin attenuates oxidative stress, inflammation, apoptosis, and oxidative DNA damage in acrylamide-induced nephrotoxicity in rats. Asian Pac J Trop Biomed 12: 223-232.

Ghasemzadeh RM, Cheraghi FH, Hosseinzadeh H, *et al.,* 2021. Protective effects of selenium on acrylamide-induced neurotoxicity and hepatotoxicity in rats. Iran J Basic Med Sci 24: 1041-1049.

Ghorbel I, Elwej A, Jamoussi K, *et al.,* 2015a. Potential protective effects of extra virgin olive oil on the hepatotoxicity induced by co-exposure of adult rats to acrylamide and aluminum. Food Funct 6: 1126-1135.

Ghorbel I, Maktouf S and Kallel C, 2015b. Ellouze Chaabouni S, Boudawara T, Zeghal N. Disruption of erythrocyte antioxidant defense system, hematological parameters, induction of pro-inflammatory cytokines and DNA damage in liver of co-exposed rats to aluminium and acrylamide. Chem Biol Interact 5:31-40.

Hayes JD, Dinkova-Kostova AT and Tew KD, 2020. Oxidative Stress in Cancer. Cancer Cell 38:167-197.

Ibrahim MA and Ibrahem MD, 2020. Acrylamide-induced hematotoxicity, oxidative stress, and DNA damage in liver, kidney, and brain of catfish (Clarias gariepinus), Environ Toxicol 35: 300–308.

Khiralla G, Elhariry H and Selim SM, 2020. Chitosan-stabilized selenium nanoparticles attenuate acrylamide-induced brain injury in rats. J Food Biochem 4: e13413.

Kiełczykowska M, Kocot J, Paździor M, *et al.,* 2018. Selenium - a fascinating antioxidant of protective properties. Adv Clin Exp Med 27 : 245-255.

Koszucka A, Nowak A, Nowak I, *et al.,* 2020. Acrylamide in human diet, its metabolism, toxicity, inactivation and the associated European Union legal regulations in food industry. Crit Rev Food Sci Nutr 60: 1677-1692.

Peña-Blanco A and García-Sáez AJ, 2018. Bax, Bak and beyond - mitochondrial performance in apoptosis. FEBS J 285: 416-431.

Savas HB and Sayar E, 2021. Oxidant antioxidant balance and trace elements in children with functional dyspepsia. Turkiye Klinikleri J Med Sci 41: 70-79.

Semla M, Goc ZM, Martiniaková R, *et al.,* 2017. Acrylamide: A common food toxin related to physiological functions and health. Physiol Res 66: 205–217.

Sengul E, Gelen V, Yildirim S, *et al.,* 2021. The effects of selenium in acrylamide-induced nephrotoxicity in rats: Roles of oxidative stress, inflammation, apoptosis, and DNA damage. Biol Trace Elem Res 199: 173-184.

Seydi E, Rajabi M, Salimi A, *et al.*, 2015. Involvement of mitochondrial-mediated caspase-3 activation and lysosomal labilization in acrylamide-induced liver toxicity. Toxicol Environ Chem 97: 563–575.

Teodor V, Cuciureanu M, Filip C, *et al.,* 2011. Protective effects of selenium on acrylamide toxicity in the liver of the rat. Effects on the oxidative stress. Rev Med Chir Soc Med Nat Iasi 115: 612-618.

Yerlikaya FH and Yener Y, 2013. The dietary acrylamide intake adversely affects the serum trace element status. Biol Trace Elem Res 152: 75-81