Original Article

Effect of Levo-Carnosine Co-Administration On Cisplatin-Induced Portal Inflammation And Fibrosis Of The Liver In BALB/c Mice

Sumyyia Bashir[1], Khadija Qamar[2], Sajid Ali[3], Tayyaba Faisal[4], Rabya Khalid[5], Aqsa Zahid[6]

Abstract

Objective: To investigate the effect of levocarnosine co-administration on portal inflammation and fibrotic alterations caused by cisplatin in the livers of BALB/c mice.

Methods: The study was carried out from December 2020 to April 2022 at the Department of Anatomy in collaboration with the Department of Physiology, Army Medical College in Rawalpindi. The study included 90 adult BALB/c mice; three groups of 30 mice each. Group A (Control) was not given any medication. Group B (Cisplatin) received intraperitoneal cisplatin 8 mg/kg body weight as a single weekly dose for four weeks, whereas Group C (Carnosine + Cisplatin) administered similar cisplatin treatment along with levo-carnosine 300 mg/kg body weight every day by oral lavage. Mice were then euthanized, a 2 ml of terminal blood sample was obtained by intracardiac puncture for biochemical analysis, and then the liver was dissected out, processed, and stained with Hematoxylin and Eosin (H&E) and Masson's trichrome stains to study portal inflammation and fibrosis, respectively.

Results: Absence of portal inflammation was observed in 86.7% of mice in group A, moderate to severe portal inflammation was observed in 66.7% of the specimens in group B, and mild portal inflammation was observed in 43.3% of the specimens in group C. Group B showed significantly ($p<0.001$) higher portal inflammation than the other three groups. Similarly, liver fibrosis was not observed in 93.3% of the mice in group A, mild to moderate fibrosis was observed in 83.3% of the specimens in group B, and mild fibrosis was noted in 26.7% of the mice in group C. Group C showed significantly ($p<0.001$) higher liver fibrosis than the other three groups. The mean serum albumin level was 2.36±0.12 g/dL, 1.71±0.12 g/dL, and 2.16±0.26 g/dL in groups A, B, and C, respectively. A significant ($p<0.001$) decrease in serum albumin was observed in group B when compared with groups A and C.

Conclusion: Cisplatin administration caused portal inflammation and fibrosis in the livers of mice. Levo-carnosine, when administered with cisplatin, reduced the toxic effects of cisplatin on the livers of mice.

Keywords: Antioxidants, Carnosine; Cisplatin; Inflammation; Liver disease.

Contributions:
SB KQ - Conception, Design
SB KQ SA RK AZ - Acquisition, Analysis, Interpretation
SB KQ SA RK – Drafting
SA TF AZ - Critical Review

All authors approved the final version to be published & agreed to be accountable for all aspects of the work.

Conflicts of Interest: None
Financial Support: None to report
Potential Competing Interests: None to report

Institutional Review Board Approval
ERC ID No: 11/2024/432
07-11-2024
Army Medical College, Rawalpindi

Review began 08/10/2025
Review ended 14/10/2025
Published 31/03/2026

How to cite this article: Bashir S, Qamar K, Ali S, Faisal T, Khalid R, Zahid A. Effect of Levo-Carnosine Co-Administration On Cisplatin-Induced Portal Inflammation And Fibrosis Of The Liver In BALB/c Mice. JRMC. 2026 Mar. 31;30(1).

https://doi.org/10.37939/jrmc.v30i1.3045

Introduction

The liver is the primary organ responsible for metabolising and eliminating foreign chemicals.[1] Consequently, it is regularly exposed to a wide range of potentially harmful substances and xenobiotics. As the primary site of metabolism and detoxification, the liver is prone to chemical accumulation, particularly from lipophilic medications, making it extremely vulnerable to drug-induced liver damage.[1] Approximately 32% of therapeutically approved drugs are later withdrawn from the market owing to their hepatotoxic side effects. Moreover, drug-induced hepatotoxicity accounts for 22% of clinical trial failures.[2] Some chemotherapeutic agents have well-characterized hepatotoxic side effects, ranging from inflammatory changes to liver failure. The primary cause is the production of toxic substances that trigger an immunological cascade or affect cell functioning by interacting with essential lipids, structural proteins, generating free radicals, or reducing antioxidant defense.[3]

Cisplatin is a potent inorganic platinum-based antineoplastic drug used to treat a wide variety of human malignancies.[4] The cytotoxic effect of cisplatin is attributed to the induction of apoptosis in cells via the production of DNA adducts. Despite being one of the most potent anti-cancer medications, cisplatin also causes damage to healthy tissues in many species.[5] Hepatocytes are susceptible to cisplatin-induced effects because of their propensity to accumulate in the liver. The mechanism underlying cisplatin-induced hepatotoxicity involves excessive synthesis of free radicals, which causes oxidative stress and inflammation. With cisplatin treatment, there have been reports of decreased glutathione levels, positive caspase-3 reactions, and loss of liver histoarchitecture.[6] Elevated levels of reactive oxygen species (ROS) negatively impact hepatocytes, leading to protein oxidation, glutathione depletion, membrane lipid peroxidation, and mitochondrial DNA damage, ultimately resulting in hepatocyte necrosis.[4]

Levo-carnosine, a dipeptide, is synthesised from a combination of L-histidine and beta-alanine amino acids. In the human body, it is highly concentrated in excitable tissues, including the brain, skeletal muscle, and heart. However, trace amounts are also present in the gastrointestinal system, kidneys, liver, and adipose tissue.[7] Levo-carnosine possesses scavenger properties for non-enzymatic free radicals. Its neuroprotective and anti-inflammatory potentials make it a potent antioxidant.[8] Additionally, it has many positive effects, such as the ability to buffer pH, chelate heavy metals, and inhibit antiglycation.[7] Levo-carnosine reduces the peroxidation of membrane lipids and prevents oxidative alterations of proteins exposed to hydroxyl radicals.[9] Levo-carnosine also exhibits anticancer effects by decreasing cancer cell proliferation in breast, ovarian, colon, and

leukemia cancer cell lines. This effect is attributed to its increased secretion of interleukin-10 (IL-10), granulocyte monocyte – colony stimulating factor (GM-CSF), and tumour necrosis factor-α (TNFα), and decreased secretion of IL-8.[10]
Cisplatin is a commonly used chemotherapeutic agent owing to its effectiveness, affordability, and ease of use; however, further research is warranted to reduce its negative effects on healthy tissues. Aiming to determine the role of levo-carnosine co-administration on cisplatin-induced portal inflammation and fibrosis in laboratory animal liver tissue, the current study was planned. We anticipate that the results of this study will serve as a useful tool for clinicians in evidence-based decision-making when using cisplatin as a chemotherapeutic agent to reduce drug-induced hepatotoxicity.

Materials And Methods

This laboratory-based experimental study was conducted from December 2020 to April 2022 at the Department of Anatomy in collaboration with the Department of Physiology, Army Medical College, Rawalpindi. The study commenced after obtaining formal approval from the Institutional Review Board (ERC ID N0. 11/2024/432 dated 07 November 2024). A total of 90 adult BALB/c mice were used in the study, which were procured from the National Institute of Health (NIH), Islamabad. The sampling technique was non-probability consecutive sampling. Healthy mice of both genders, aged 8-10 weeks, weighing 25-30 met the inclusion criteria. Mice with liver disease (as determined by serum alanine aminotransferase levels at the beginning of the trial based on a sample of blood from the rat tail vein) were excluded from the study.
Mice were housed in individual cages (seven to eight mice per cage) with bedding made of wooden shavings, and the room temperature was maintained at 22±3 °C. They were exposed to a dark and light cycle of 12 h every day. Housing and procedural protocols for mice were performed in compliance with the Institute's Ethical Review Committee approval and standard guidelines for experimental animal care.[11] By random distribution, mice were distributed into three groups of 30 each (n=30). Group A was the control, Group B was cisplatin, and Group C was carnosine + cisplatin. Animals had unrestricted access to water and a regular chow diet throughout the experimental period. Group-A mice were not administered any medication. Mice in Group B received an intraperitoneal injection of cisplatin at a dose of 8 mg/kg body weight once a week for four weeks (days 7, 14, 21, and 28).[12] In contrast, mice in Group C were administered a similar dose of cisplatin as Group B plus 300 mg/kg body weight of levo-carnosine (dissolved in water) orally every day by oral lavage for four weeks. [13]
At the end of the trial period, the mice were euthanised by an overdose of diethyl ether anaesthesia.[14] A 2-mL terminal blood sample was obtained by intracardiac puncture for biochemical analysis. Dissection was performed by cutting the skin and muscles through a midline abdominal incision. A skilled veterinarian at the National Institute of Health, Islamabad, performed the animal dissection. The abdominal viscera were made visible, and the liver was identified as being located beneath the diaphragm on the right side. The hepatic vein and inferior vena cava were sectioned, the peritoneal connections were severed, and the liver was dissected. After being cleaned with saline, the weight and volume of the liver were recorded, and the organ was placed in labelled containers filled with 10% formalin solution that was sufficient to completely submerge the tissue. The liver was processed and sectioned using a rotary microtome into 5-µm-thick slices after being left for 24 h to allow settling. After floating the sections on a warm water bath at 45 °C, they were mounted on glass slides and stained with haematoxylin and eosin (H&E) and Masson's trichrome stain to examine portal inflammation and collagen fibres (fibrosis), respectively.
Portal inflammation was defined as mononuclear cell (lymphocytes and monocytes) infiltration within the portal areas. Scoring was based on the modified Ishak Histology Activity Index.[15] Grade-0 indicated no inflammation; Grade-1 indicated mild inflammation (<20% of a portal area of inflammatory cells in some 20-40% of all portal areas); Grade-2 indicated moderate inflammation (20-50% of a portal area of inflammatory cells in some 20-40% of all portal areas); and Grade-3 indicated marked inflammation (>50% of a portal area of inflammatory cells in almost all areas).
For the analysis of fibrosis**, collagen** deposition was observed in blue within the portal areas and was semi-quantitatively assessed and scored using the modified Ishaq staging.[16] Grade-0 indicated no fibrosis; Grade-1 indicated portal fibrosis (fibrous expansion of the portal area with or without short fibrous septa in <50% of portal areas); Grade-2 indicated fibrous septa (portal fibrosis with portal-to-portal and portal-to-central bridging in >50% of the portal area); and Grade-3 indicated marked bridging with nodules in all portal areas.
The obtained blood sample was centrifuged at 3000 rpm for 15 min, and the collected serum was used for the quantitative assessment of serum albumin using commercial kits on a fully automated clinical chemistry analyser (Vitalab Selectra E).
Data analysis was done using IBM (International Business Machine) SPSS (Statistical Package for the Social Sciences) version 25. Qualitative variables (portal inflammation and fibrosis of the liver) are expressed as frequency and percentage; quantitative variables (age of mice, initial weight of mice, final weight of mice, weight of liver, relative tissue body weight index [RTBWI], volume of liver, and serum albumin) are described using descriptive statistics, that is, mean ± standard deviation (SD). One-way analysis of variance followed by Tukey's post hoc test was used to compare the differences in quantitative variables, while the chi-square test was used to assess qualitative data. Statistical significance was set at $P \leq 0.05$.

Results

The mean age of the animals at the commencement of the study was not significantly different between the groups (Table 1). The initial weight of the mice was 26.7±0.75 g, 27.0±0.87 g, and 26.9±0.85 g in groups A, B, and C, respectively, which was not significantly different (Table 1). The final weight of the mice after a four-week treatment period before sacrifice was 30.03±1.13 g in group A, 25.73±0.83 g in group B, and 29.2±1.35 g in group C ($p<0.001$). A post hoc comparison revealed a significant ($p<0.001$) decrease in the final weight of mice in group B compared to groups A and C. Significant differences ($p=0.014$) in weight were also noted among the other two experimental groups (Table 1). The weight of the liver was 1.6±0.14g, 1.4±0.11g, and 1.56±0.09g in groups B and C, respectively. Statistical analysis showed a significant ($p<0.001$) difference among the groups (Table 2). The weight of the liver was considerably lower ($p<0.001$) in group B than in the other two groups. In comparison to group A, the mean liver weight in group C was also low, but not significant ($p=0.286$) (Table 2). The RTBWI was 5.34±0.5 g in group A, 5.46±0.36 g in group B, and 5.33±0.23g in group C. The mean RTBWI in group B was slightly higher than that in the other groups but was not significantly different (Table 2). The mean liver volume was 1.92±0.17 cm^3, 1.69±0.13 cm^3, and 1.87±0.11 cm^3 in groups B and C, respectively ($p<0.001$). Liver volume was significantly lower ($p<0.001$) in group B than in the other two groups. The liver volume of group C was also low when compared to that in group A, but was not significant ($p=0.286$) (Table 2). The mean serum albumin level was 2.36 ±0.12 g/dL in group A, 1.71 ±0.12 g/dL in group B, and 2.16 ±0.26 g/dL in group C ($p<0.001$). A significant ($p<0.001$) decrease in serum albumin level was observed in group B compared to groups A and C. The difference in serum albumin between group C and group A was also significant ($p<0.001$) (Table 2).

Table 1: Intergroup comparison of age, initial weight, and final weight of mice between control group A and experimental groups B and C

Parameter	Groups	Mean ± SD	Statistical Significance (p-value)		
			Group A/B	Group B /C	Group A /C
Age of Mice (weeks)	A	8.6 ± 0.68	0.642	0.931	0.852
	B	8.77 ± 0.73			
	C	8.73 ± 0.79			
Initial weight of Mice (g)	A	26.7 ± 0.75	0.340	0.885	0.616
	B	27.0 ± 0.87			
	C	26.9 ± 0.85			
Final weight of Mice (g)	A	30.03 ± 1.13	0.000	0.000	0.014
	B	25.73 ± 0.83			
	C	29.2 ± 1.35			

Table 2: Intergroup comparison of the weight of liver, RTBWI, volume of liver, and serum albumin levels between control group A and experimental groups B and C

Parameter	Groups	Mean ± SD	Statistical Significance (p-value)		
			Group A/B	Group B /C	Group A /C
Weight of liver (g)	A	1.6 ± 0.14	0.000	0.000	0.286
	B	1.4 ± 0.11			
	C	1.56 ± 0.09			
RTBWI (g)	A	5.34 ± 0.5	0.463	0.381	0.989
	B	5.46 ± 0.36			
	C	5.33 ± 0.23			
Volume of liver (cm^3)	A	1.92 ± 0.17	0.000	0.000	0.286
	B	1.69 ± 0.13			
	C	1.87 ± 0.11			
Serum Albumin (g/dl)	A	2.36 ± 0.12	0.000	0.000	0.000
	B	1.71 ± 0.12			
	C	2.16 ± 0.26			

Microscopic examination of haematoxylin and eosin-stained liver sections revealed the absence of portal inflammation (grade 0) in 86.7% and grade-1 (mild) portal inflammation in 13.3% of mice in group A. In group B, grade-1 (mild) portal inflammation was observed in 20% of liver specimens, grade-2 (moderate) portal inflammation was observed in 50% of specimens, grade-3 (severe) portal inflammation was observed in 16.7% of specimens, whereas 13.3% of specimens were found normal in this group (Table 3 and Figure 1). (Table 3 and Figure 1). In the experimental group C, grade-1 (mild) portal inflammation was observed in 43.3% liver specimens, grade-2 (moderate) portal inflammation was observed in 20% liver specimens, whereas 37.7% of specimens were found normal in this group (Table 3 and Figure 1). The results in intergroup comparison showed a significant difference (Table 3). On histological examination of Masson's trichrome-stained liver sections, the absence of liver fibrosis (grade 0) was observed in 93.3% of mice, while grade-1 fibrosis was observed in 6.7% of animals in group A. In experimental group B, 60% of mice had grade-1 fibrosis, 23.3% had grade-2 fibrosis, and 16.7% of specimens in this group revealed no fibrosis (Table II and Figure 1). In experimental group C, 26.7% of mice showed grade-1 fibrosis, and 73.3% of animals showed no liver fibrosis in this group (Table 3 and Figure 1). The results in intergroup comparison showed a significant difference (Table 3).

Table 3: Intergroup comparison of portal inflammation and fibrosis of the liver between control group A and experimental groups B and C

Parameter	Findings	Group-A	Group-B	Group-C	Statistical Significance (p-value)		
					Group A/B	Group B /C	Group A /C
Portal Inflammation (%)	Grade-0	86.7	13.3	36.7	0.000	0.002	0.000
	Grade-1	13.3	20	43.3			
	Grade-2	-	50	20			
	Grade-3	-	16.7	-			
Fibrosis of Liver (%)	Grade-0	93.3	16.7	73.3	0.000	0.000	0.080
	Grade-1	6.7	60	26.7			
	Grade-2	-	23.3	-			
	Grade-3	-	-	-			

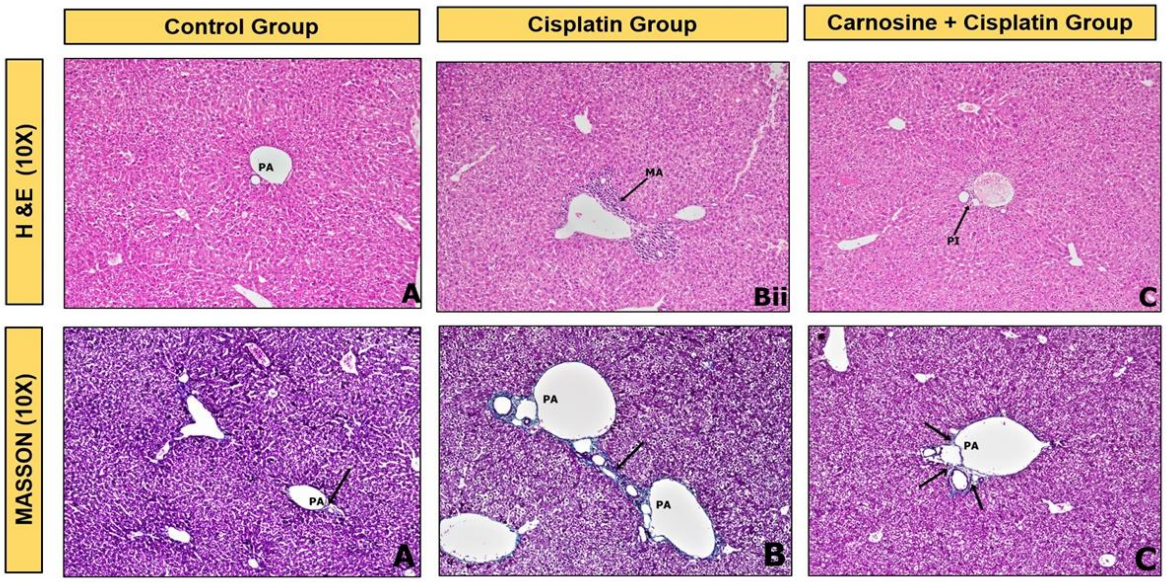

Figure 1. H&E-stained liver sections showing no portal inflammation in the control group (A), marked portal inflammation in the cisplatin group (B), and reduced portal inflammation in the carnosine + cisplatin group (C). Masson's trichrome-stained liver sections showing scant collagen fibers in the control group (A), markedly increased collagen fibers in the cisplatin group (B), and mild collagen deposition in the carnosine + cisplatin group (C). (x 10). MA, marked; PA, portal area; PI, portal inflammation

Discussion

In the current study, significant histomorphological abnormalities in the mouse liver were noted following cisplatin therapy. When levocarnosine and cisplatin were administered together, these abnormalities were reduced. Levocarnosine has been proven useful in the prevention and treatment of certain cancers. Oral carnosine supplementation tends to increase its bioavailability and accessibility, making it the focus of experimental and interventional trials.[9]

The mice in Group B, who were treated with cisplatin alone, showed significant weight loss. Our findings are analogous to the study carried out by Sioud et al., reporting weight reduction in animals by 10.9% following cisplatin administration.[16] Whereas the animals' weight change in Group C of the present study was reported as weight gain, this is attributed to the effect of levo-carnosine. In the current study, RTBWI was calculated for the liver, and it showed that RTBWI in the cisplatin-treated group is increased when compared to the other two groups. This increase in the RTBWI was noticed because the decrease in the mice's body weight in this group was relatively greater compared to the decrease in the weight of the liver, which resulted in increased RTBWI in this group. Our results are comparable to the findings of a study performed by Hwang et al., in which the authors reported increased relative weight of the liver by intraperitoneal injection of cisplatin.[17] Reduction in volume of liver was also noted in Group B, which is attributable to decreased weight of the liver and cisplatin-induced fibrosis. *Levo*-carnosine supplementation has significantly improved the volume of the liver in study Group C. Zhu et al., in their study, have mentioned a reduction in liver volume as a result of liver fibrosis, which is consistent with the findings of the present study.[18]

Histological examination of liver sections revealed marked portal inflammation in group B. Cisplatin increases TNF-α levels by activating Kupffer cells, which in turn leads to inflammation by activating macrophages and releasing inflammatory cytokines. In addition to decreasing the transcription levels of NF-κB -p65 and caspase-1, cisplatin increases cyclooxygenase-2 (COX-2) levels and IL-1β and the activity of caspase-3, suggesting a general trend toward inflammation and a rise in the apoptotic shift.[19] Similar findings were reported by Zhang et al., who assessed the ameliorating effect of ellagic acid on cisplatin-induced hepatotoxicity, and found that cisplatin caused inflammatory infiltration in the portal areas of the mouse liver when administered at a dose of 20 mg/kg/day for two days.[20] L-carnosine treatment significantly decreased portal inflammation in group C, thus demonstrating its role as an anti-inflammatory agent. Our results are in accordance with those of a previous study by Liu et al., who evaluated the role of carnosine in alleviating inflammation and fibrosis in a streptozotocin-induced diabetic mouse model.[21] The authors showed that carnosine supplementation ameliorates tissue inflammation by reducing IL-1β and TNF-α.

In the current study, significant liver fibrosis was observed in mice treated with cisplatin. Cisplatin has been implicated in hepatic fibrosis in previous studies.[22] Increased production of reactive oxygen species (ROS) from cisplatin-induced liver damage triggers an inflammatory response, which in turn activates hepatic stellate cells (HSCs) to become fibrogenic myofibroblasts. Nimbalkar et al. mentioned that cisplatin increases the deposition of collagen fibres and fibrous expansion of the portal areas, causing alterations in the architecture of the liver,[23] which initially can be represented as a change in hepatic lobule size, as revealed in our study. However, as demonstrated in our study, group C liver fibrosis was protected by levocarnosine, which helped restore the hepatic lobule size. Mousa et al. concluded in their study that carnosine, being a scavenger of ROS, reduces the inflammatory cytokines like TNFα and, in turn, has an anti-fibrotic effect by reducing the effects of IL-1α on the liver.[24]

The current study observed decreased serum albumin levels in Group B in relation to Groups A and C. The cisplatin-induced damage to the hepatocytes, followed by the development of fibrosis, is a plausible cause of this finding. Our results of decreased albumin levels as a result of hepatotoxicity are in accordance with those of Chang et al., who also reported similar findings as a result of hepatotoxicity caused by exposure to low-level vinyl chloride in mice.[25] *Levo*-carnosine supplementation significantly improved serum albumin levels in Group C. Our results are

in accordance with those of Ali et al., who also reported a significant improvement in serum albumin levels as a result of levo-carnosine supplementation along with cisplatin in Sprague–Dawley rats.[3]

The current study demonstrated that cisplatin chemotherapy causes inflammatory changes and fibrosis in the liver. Because of its anti-inflammatory properties and reduced generation of reactive oxygen species, the co-administration of levo-carnosine with cisplatin has shown significant improvement in liver histopathological changes. Based on these findings, the present study suggests that levo-carnosine may be used as an adjunct while treating malignancies with cisplatin to reduce hepatotoxicity. The limitations of the study were our inability to precisely assess the types and concentrations of proinflammatory markers causing portal inflammation and fibrotic changes in the liver.

Conclusions

Cisplatin administration causes portal inflammation and fibrosis in the liver. Levo-carnosine, when administered with cisplatin, reduces the toxic effects of cisplatin on the liver of mice.

Author Information

1. Assistant Professor, Army Medical College 2. Professor, Army Medical College 3,4. Assistant Professor, Army Medical College 5. Demonstrator, Army Medical College 6. Assistant Professor, Army Medical College/RIHS.
Corresponding author: Dr. Sumyyia Bashir sumayyab88@gmail.com

References

1. Björnsson H, Björnsson E. Drug-induced liver injury: pathogenesis, epidemiology, clinical features, and practical management. Eur J Intern Med. 2022; 97:26-31. https://doi.org/10.1016/j.ejim.2021.10.035
2. Zhang CJ, Meyer SR, O'Meara MJ, Huang S, Capeling MM, Ferrer-Torres D, et al. A human liver organoid screening platform for DILI risk prediction. J Hepatol. 2023; 78(5):998-1006. https://doi.org/10.1016/j.jhep.2023.01.019
3. Ali S, Rasul A, Latif N, Aleem SB, Zia R, Khan B. Effect of levo-carnosine on biomarkers of oxidative stress and hepatotoxicity in cisplatin-treated male Sprague Dawley rats. Pak Armed Forces Med J. 2022; 72(4):1334-8.
4. Habib SA, Suddek GM, Rahim MA, Abdelrahman RS. The protective effect of protocatechuic acid on hepatotoxicity induced by cisplatin in mice. Life Sci. 2021; 277:119485. https://doi.org/10.1016/j.lfs.2021.119485
5. Sherif IO. Hepatoprotective effect of arjunolic acid against cisplatin-induced hepatotoxicity: Targeting oxidative stress, inflammation, and apoptosis. J Biochem Mol Toxicol. 2021; 35(4):e22714. https://doi.org/10.1002/jbt.22714
6. Taghizadeh F, Hosseinimehr SJ, Zargari M, Karimpour Malekshah A, Mirzaei M, Talebpour Amiri F. Alleviation of cisplatin-induced hepatotoxicity by gliclazide: Involvement of oxidative stress and caspase-3 activity. Pharmacol Res Perspect. 2021; 9(3):e00788. https://doi.org/10.1002/prp2.788
7. Wang Q, Sadati S, Kabthymer RH, Gadanec LK, Lawton A, Tripodi N, et al. The impact of carnosine on biological ageing–A geroscience approach. Maturitas. 2024; 189:108091. https://doi.org/10.1016/j.maturitas.2024.108091
8. Cesak O, Vostalova J, Vidlar A, Bastlova P, Student V Jr. Carnosine and beta-alanine supplementation in human medicine: narrative review and critical assessment. Nutrients. 2023; 15(7):1770. https://doi.org/10.3390/nu15071770
9. Jukić I, Kolobarić N, Stupin A, Matić A, Kozina N, Mihaljević Z, et al. Carnosine, small but mighty—prospect of use as a functional ingredient for functional food formulation. Antioxidants (Basel). 2021; 10(7):1037. https://doi.org/10.3390/antiox10071037
10. Prakash MD, Fraser S, Boer JC, Plebanski M, de Courten B, Apostolopoulos V. Anti-Cancer Effects of Carnosine-A Dipeptide Molecule. Molecules. 2021; 26(6):1644. https://doi.org/10.3390/molecules26061644
11. Kiani AK, Pheby D, Henehan G, Brown R, Sieving P, Sykora P, et al. Ethical considerations regarding animal experimentation. J Prev Med Hyg. 2022; 63(2 Suppl 3):E255-E266. https://doi.org/10.15167/2421-4248/jpmh2022.63.2s3.2768
12. Yu JB, Lee DS, Padanilam BJ, Kim J. Repeated Administration of Cisplatin Transforms Kidney Fibroblasts through G2/M Arrest and Cellular Senescence. Cells. 2022;11(21):3472. https://doi.org/10.3390/cells11213472
13. Park J, Jang J, Cha SR, Baek H, Lee J, Hong SH, et al. L-carnosine Attenuates Bleomycin-Induced Oxidative Stress via NFκB Pathway in the Pathogenesis of Pulmonary Fibrosis. Antioxidants (Basel). 2022;11(12):2462. https://doi.org/10.3390/antiox11122462
14. Tobar Leitão SA, Soares DDS, Carvas Junior N, Zimmer R, Ludwig NF, Andrades M. Study of anesthetics for euthanasia in rats and mice: A systematic review and meta-analysis on the impact upon biological outcomes (SAFE-RM). Life Sci 2021; 284:119916. https://doi.org/10.1016/j.lfs.2021.119916
15. Krishna M. Histological Grading and Staging of Chronic Hepatitis. Clin Liver Dis (Hoboken). 2021; 17(4):222-226. https://doi.org/10.1002/cld.1014
16. Sioud F, Ben Toumia I, Lahmer A, Khlifi R, Dhaouefi Z, Maatouk M, et al. Methanolic extract of Ephedra alata ameliorates cisplatin-induced nephrotoxicity and hepatotoxicity by reducing oxidative stress and genotoxicity. Environ Sci Pollut Res Int 2020; 27(11):12792-801. https://doi.org/10.1007/s11356-020-07904-3
17. Hwang D-B, Won D-H, Shin Y-S, Kim S-Y, Kang B-C, Lim K-M, et al. Ccrn4l as a pre-dose marker for prediction of cisplatin-induced hepatotoxicity susceptibility. Free Radic Biol Med 2020; 148:128-39. https://doi.org/10.1016/j.freeradbiomed.2020.01.003
18. Zhu T, Li Z, Yuan J, Huang K, Chen G, Guo R, et al. Characteristics of liver volume and pathological changes with different stages of liver fibrosis in chronic liver disease. Zhonghua Gan Zang Bing Za Zhi 2024; 32(6):517-24. https://doi.org/10.3760/cma.j.cn501113-20231219-00286
19. Esmat MA, Osman A, Hassan RE, Hagag SA, El-Maghraby TK. Hepatoprotective effect of ferulic acid and/or low doses of γ-irradiation against cisplatin-induced liver injury in rats. Hum Exp Toxicol 2022; 41:09603271221136205. https://doi.org/10.1177/09603271221136205
20. Zhang X, Liu Y, Liu M, Ma Q, Hao Z, Tang S, et al. Ellagic acid supplementation ameliorates cisplatin-induced liver injury in mice by inhibiting the NF-κB pathway and activating the Nrf2/HO-1 pathway. One Health Adv 2024; 2(1):20. https://doi.org/10.1186/s44280-024-00056-w

21. Liu X-q, Jiang L, Lei L, Nie Z-y, Zhu W, Wang S, et al. Carnosine alleviates diabetic nephropathy by targeting GNMT, a key enzyme mediating renal inflammation and fibrosis. Clin Sci 2020; 134(23):3175-93. https://doi.org/10.1042/cs20201207
22. Bademci R, Erdoğan MA, Eroğlu E, Meral A, Erdoğan A, Atasoy Ö, et al. Demonstration of the protective effect of ghrelin in the livers of rats with cisplatin toxicity. Hum Exp Toxicol 2021; 40(12):2178-87. https://doi.org/10.1177/09603271211026722
23. Nimbalkar V, Vyawahare N. L-carnitine Protects Cisplatin-Induced Liver Fibrosis in Experimental Animals via Reducing Oxidative and Nitrosative Stress. Int J Pharmaceut Invest 2022; 12(3):370-4. https://doi.org/10.5530/ijpi.2022.3.62
24. Mousa AM, Aldebasi YH. L-carnosine mitigates interleukin-1α-induced dry eye disease in rabbits via its antioxidant, anti-inflammatory, antiapoptotic, and antifibrotic effects. Cutan Ocul Toxicol 2021; 40(3):241-51. http://doi:10.1080/15569527.2021.1935995
25. Chang LT, Lee YL, Yuan TH, Chang JH, Chang TY, Lee CH, et al. Hepatotoxicity caused by repeated and subchronic pulmonary exposure to low-level vinyl chloride in mice. Atmosphere 2021; 12(5):596. https://doi.org/10.3390/atmos12050596