Full text open access online (Since 2007)

Ethno Med, 20(2): 99-109 (2026)
PRINT: ISSN 0973-5070 ONLINE: ISSN 2456-6772 DOI: 10.31901/24566772.2026/20.02.724

Antioxidant and Protective Effects of Agaricus bisporus Against Cisplatin Induced Nephrotoxicity in Male Albino Rats

Rajasekar Ramachandran[1*], Manikandan Ramasamy[2], Nithyatharani Ramalingam[3], Prasun Kumar[4] Manimegalai Kulanthaipandi[5] and Cecileya Jasmin[6]

[1]Department of Biochemistry, Maruthupandiyar College (Affiliated to Bharathidasan University, Tiruchirappalli), Thanjavur, Tamil Nâdu, India*
[2]Department of Biochemistry, Shrimati Indira Gandhi College, Tiruchirappalli, Tamilnadu, India
[3]Department of Microbiology, Shrimati Indira Gandhi College, Tiruchirappalli, Tamilnadu, India
[4]Department of Biotechnology, School of Bioscience and Technology, Sharda University, Greater Noida, India
[5]Department of Zoology, Sri G.V.G Visalakshi College for Women (Autonomous), Udumalpet, Tamilnadu, India
[6]Department of Biochemistry, Saveetha Medical College and Hospital, Saveetha Institute of Medical and Technical Sciences (Deemed to be University), Thandalam, Chennai, Tamil Nadu, India

KEYWORDS *Agaricus bisporus.* Cisplatin. Histopathology. Nephrotoxicity. Silymarin

ABSTRACT The nephrotoxicity linked to reactive oxygen species (ROS) formation, which exacerbates cellular damage, frequently limits the clinical application of cisplatin (CP), a commonly used chemotherapeutic medication that is effective against a variety of solid malignancies. These harmful effects can be lessened by antioxidants. This study evaluated the protective effect of an Agaricus bisporus methanolic extract on male albino rats. Control, silymarin, cisplatin, and mushroom were the four groups into which the animals were divided. After 15 days, the samples were subjected to various enzyme levels and kidney tissues were examined histologically. Histology revealed better kidney structure, and mushroom therapy markedly decreased malondialdehyde (MDA) and raised glutathioneperoxidase (GPx), catalase (CAT), and superoxide dismutase (SOD) activity. According to these results, *Agaricus bisporus* has antioxidant qualities that guard against oxidative kidney damage brought on by cisplatin and may lessen the toxicity of chemotherapy.

INTRODUCTION

Nephrotoxicity remains a major dose-limiting adverse effect of Cisplatin in clinical oncology, despite advances in hydration protocols and supportive care. Cisplatin-induced renal damage is a complex process involving oxidative strain, impaired mitochondria, activation of the DNA damage response, signalling related to inflammation, and several forms of programmed cell death, such as apoptosis, necroptosis, and ferroptosis, according to recent mechanistic studies. Cisplatin is preferentially absorbed by proximal tubular epithelial cells after systemic treatment through membrane transporters such as organic cation transporter 2 and copper transporter 1, resulting in intracellular accumulation. After entering the cell, cisplatin undergoes aquation reactions that produce extremely reactive platinum complexes that can build DNA adducts, stopping the cell cycle and activating p53-dependent pathways. Furthermore, an excess of reactive oxygen species (ROS) overwhelms the body's natural antioxidant defenses, causing damage to mitochondrial DNA, lipid peroxidation, and protein carbonylation. Recent research has shown that tubular injury is further exacerbated by mitochondrial ROS amplification and defective mitophagy, underscoring mitochondria as key mediators in cisplatin nephrotoxicity (Peres and da Cunha 2013; Yang et al. 2023).

The development of acute kidney damage (AKI) brought on by cisplatin is also significantly influenced by inflammatory reactions. Nuclear factor-kappa B (NF-êB) signalling activation and pro-inflammatory cytokines like tumour necrosis factor-alpha (TNF-á) increase interleukin-1 beta (IL-1â), and interleukin-6 (IL-6) contribute to leukocyte infiltration and sustained tissue damage. Toll-like receptor 4 (TLR4) signalling and NLRP3 inflammasome activation have recently been implicated in amplifying re-

nal inflammation following cisplatin exposure. Furthermore, new research indicates that tube damage in cisplatin-treated experimental mice is largely caused by ferroptosis, an iron-dependent lipid peroxidation-driven type of cell death. Targeting these inflammatory and redox-sensitive pathways has therefore become a promising strategy to mitigate renal toxicity without compromising the antitumor efficacy of cisplatin (Li et al. 2022; Deng et al. 2023).

Natural products with anti-inflammatory and antioxidant qualities have attracted considerable attention as adjunctive agents to counteract chemotherapy-associated organ toxicity. Among edible medicinal mushrooms, *A. bisporus* has gained recognition due to its rich phytochemical and nutraceutical profile. Commonly known as the button mushroom, *A. bisporus* contains bioactive constituents such as phenolic acids, flavonoids, ergothioneine, polysaccharides (including â-glucans), selenium, and vitamins, all of which contribute to its potent antioxidant capacity. Ergothioneine, a naturally occurring thiol compound abundantly present in A. bisporus, has been identified as a cytoprotective antioxidant that accumulates in tissues exposed to oxidative stress, including the kidney. Recent experimental studies have demonstrated that mushroom-derived polysaccharides can modulate immune responses, suppress NF-êB activation, enhance endogenous antioxidant enzyme activities, and reduce MDA levels in oxidative stress models (Kalaras et al.2017; Rahman et al. 2022).

Additionally, in animal models of toxin-induced renal injury, dietary supplementation with *A. bisporus* extracts has demonstrated renoprotective effects by reducing the expression of inflammatory cytokines, maintaining mitochondrial integrity, and raising glutathione (GSH) levels. Mushroom-derived bioactive fractions have been shown in recent in vivo studies in rodent models to significantly lower serum creatinine and blood urea nitrogen levels, improve renal tubule histopathological architecture, and downregulate apoptotic markers like caspase-3 and Bax while upregulating anti-apoptotic Bcl-2 expression. These protective effects are largely attributed to synergistic antioxidant, anti-inflammatory, and anti-apoptotic mechanisms (Zhang et al. 2022; Elhusseiny et al. 2023).

Given the growing global burden of cancer and the widespread clinical application of cisplatin-based chemotherapy, identifying safe, dietary-derived protective agents is of considerable therapeutic importance. The integration of functional foods such as *A. bisporus* as adjunctive interventions may represent a promising strategy to reduce cisplatin-induced nephrotoxicity while maintaining anticancer efficacy. However, further well-designed preclinical and clinical studies are necessary to elucidate optimal dosing regimens, bioavailability, molecular targets, and potential interactions with chemotherapeutic agents.

Objectives

The renoprotective effect of *A. bisporus* in cisplatin-induced kidney injury will be further understood by comprehending the interaction between oxidative stress regulation, inflammatory signalling, and cell death pathways.

MATERIAL AND METHODS

Plant Material

Whitebutton mushroom (*A. bisporus*) was purchased from a local market in Tiruchirappalli, Tamil Nadu, India.

Drugs and Chemicals

Vials of cisplatin were used to cause nephrotoxicity, and Silymarin, a common medication, was purchased from a Trichy-based pharmacy. Every other chemical and reagent utilised in the investigation was of analytical quality and purchased commercially.

Preparation of a Methanol Extract of Mushrooms

Methanolic *A. bisporus* extract After being cleaned, the fresh macrofungus was dried for three days at 370 degrees in a hot air oven. To keep them safe from moisture, dried samples were placed in an airtight container. Using 500 milliliters of methanol and stirring at 300 rpm for 24 hours, 50 grams of dried mushrooms were extracted. Following that, the water was filtered

using Whatman No. 1 filter paper. Another 500 cc of methanol was used to remove the residue. After that, the mixture of methanol extract was rotary evaporated for two hours.

Experimental Animals

Male albino rats weighing between 150 and 200 grams were donated by the Animal Science department at Bharathidasan University in Trichy. They were housed in sterile polypropylene cages with standard lighting (12 hours of light and 12 hours of darkness), temperature (25±20°C), humidity (45±4%), and a regular diet with unlimited water. This study was approved by the Institutional Animal Ethics Committee (IAEC) (1416/PO/a/11/CPCSEA).

Experimental Design

After a week of acclimation, male albino rats were randomly assigned to four groups of six animals each.

- Group I: For 15 days, distilled water was given orally to normal control rats.
- Group II: On day 1, rats received a single intraperitoneal (16 mg/kg b.w) dose of cisplatin.
- Group III: Rats were administered an oral dose of silymarin (50 mg/kg b.w/day) from the second to the fifteenth day for 14 days following a single dose of cisplatin on day 1.
- Group IV: Following a single intraperitoneal (i.p.) injection of cisplatin on day 1, rats were given an oral dose of *A. bisporus* methanol extract (200 mg/kg of b.w /day) from the second to the fifteenth day for 14 days.

Blood samples were obtained for hematological and biochemical significance analyses in each group.

Preparation of Kidney Homogenate

Before the experiment finished on the fifteenth day fasting on whole night was performed on rats. The starving rats were killed under chloroform anesthesia on the sixteenth day of the process. The kidney was fast isolated, cleaned in ice cold isotonic saline and then blotted on ash-free filter paper. The tissue was then homogenized at 0.1 M. The supernatant was kept at 20°C after the homogenate was centrifuged for 15 minutes at 9000 rpm in a cold centrifuge (Al-Hashem 2009) for the antioxidant enzyme activity test.

Lipid Peroxidation Analysis

According to Heath and Backer's description, lipid peroxidation was assessed as the generation of malondialdehyde (MDA) (Heath and Backer1968). On the sixteenth day, the animals were decapitated and sacrificed. After dissection, the kidney was placed in ice-cold saline to prevent blood contamination. Following the collected sample's removal process, the plant part was first washed in distilled water. After that, they were weighed, compressed onto blotting paper, and homogenized in 1.5 percent KCl using a Teflon homogenizer. Following the addition of 2.5 mL of 20% TCA to 1 mL homogenate, centrifugation was performed at 3500 rpm for 10 minutes. The pellet was dissolved in 2.5 ml of 0.5 M H_2SO_4, thiobarbituric acid was added (3 ml), and incubated at 37^0C for 30 minutes. Spectrophotometry at 530 nm was used to quantify the absorbance after extraction of the substances into 4 milliliters of n-butanol.

Enzymatic Antioxidant Analysis

The Kakkar et al. (1984) method was used to test the renal tissue supernatant for SOD. (1984) method. The Sinha (1972) method was employed to quantify the renal tissue supernatant's CAT activity. applying Rotruck et al.'s (1973) method. GPx activity in the renal tissue supernatant was assessed in 1973.

Histopathological Analysis

Following animal sacrifice, kidney samples from the treatment and control groups were removed and cleaned with regular saline. Before being incorporated in paraffin wax, they were fixed for a whole day in 10% buffered formalin. Haematoxylin-eosin dye was used to stain cross sections of kidney tissue that were five to six millimeters thick (Bartelink et al. 2002).

Statistical Analysis

The results were displayed as the standard deviation of the mean value. Analysis of vari-

ance using the ANOVA test was used to conduct within-group comparisons. To ascertain whether there was a significant difference between the experimental and normal control groups, the student's t test was employed. A probability level of less than five percent ($P < 0.05$) was considered significant.

RESULTS

Impact of Agaricus bisporus on Renal Enzymatic Antioxidants in Nephrotoxic Rats Induced by Cisplatin

Tables 1 and 2 display the enzymatic antioxidant activity in the kidney homogenates of each animal group. Renal antioxidant enzyme levels, including GPx, CAT and SOD, were considerably lower in the cisplatin-treated group II than in the normal control group I. Group IV treated with *A. bisporus* had considerably ($p<0.05$) higher levels of antioxidant enzymes such as CAT, SOD, and GPx than group II treated with cisplatin. Antioxidant enzyme levels were also significantly ($p<0.05$) higher in group III treated with silymarin compared to group II treated with cisplatin.

Agaricus bisporus's Effect on Cisplatin-Induced Renal Lipid Peroxidation in Nephrotoxic Rats

Tables 1 and 2 demonstrate the quantity of MDA in the kidney homogenates of animals in groups I, II, III, and IV. The cisplatin-treated group II showed significantly higher levels of lipid peroxidation than the normal control group I. Treatment with *A. bisporus* (Group IV) significantly (pd"0.05) reduced the quantity of lipid peroxidation in renal tissue when compared to group II, which received cisplatin. In addition, the kidney tissue lipid peroxidation level was considerably ($p<0.05$) less in group III treated with silymarin than in group II treated with cisplatin.

Agaricus bisporus Impact on Histopathological Analysis in Rats with Nephrotoxicity Caused by Cisplatin

Group I

The glomeruli (G) are numerous and morphologically typical in the cortex of a normal control rat kidney, according to sections of that kidney. Cells with copious amounts of brilliant eosinophilic cytoplasm line the tubules (T), which are normal (arrows). Additionally, the interstitium and blood vessels are normal (Fig. 1 A).

Group II

Sections of a rat kidney treated with cisplatin demonstrate the abundance and aberrant morphology of glomeruli (G) in the cortex. The lining epithelium sloughs off of the tubules (T),

Table 1: Observed anti-oxidant values at different concentrations

Concentration (Mg)	*SOD (OD values)*	*CAT (OD values)*	*GPX (OD values)*	*MDA (OD values)*
0	8.50	3.80	6.00	6.50
50	8.70	4.00	6.20	6.20
100	8.90	4.20	6.30	5.80
150	9.10	4.30	6.40	5.50
200	9.30	4.50	6.50	5.10

Table 2: *Agaricus bisporus's* impact on MDA and renal enzymatic antioxidants in rats with cisplatin-induced nephrotoxicity

Groups	*SOD*	*CAT*	*GPX*	*MDA*
I	9.07±0.63	4.61±0.32	7.86±0.55	4.44±0.31.
II	6.79±0.47	3.47±0.24	5.86±0.41	7.75±0.54
III	8.82±0.61	4.65±0.32	6.93±0.48	4.79±0.33
IV	9.20±0.64	4.22±0.29	6.53±0.45	5.14±0.35

indicating pathogenic alterations (arrows). Additionally, the interstitium and blood vessels are normal (Fig. 1 B).

Group III

Rat kidney slices treated with silymarin demonstrate that the glomeruli (G) are numerous and anatomically normal in the cortex. Some tubules (T) exhibit cells with a lot of brilliant eosinophilic cytoplasm (right arrows), whereas others display pathological alterations marked by shedding the lining epithelium (down arrows). Additionally, the interstitium and blood vessels are normal. Therefore, the tubular pathological alterations brought on by cisplatin seem to be partially reversed (Fig. 1 C).

Group IV

Rat kidney sections treated with *A. bisporus* show that the cortex's glomeruli (G) are abundant and anatomically normal. The lining epithelium (arrows) of every tubule (T) has cells with eosinophilic, normal, and copious cytoplasm. The interstitium, limbs, and blood vessels are all in normal condition. As a result, Agaricus bisporus completely reversed the tubular pathological alterations brought on by cisplatin (Fig. 1 D).

DISCUSSION

It is commonly known that oxidative stress plays a key role in cisplatin nephrotoxicity. Cisplatin increases the generation of reactive oxygen species (ROS), including hydrogen peroxide and superoxide anion radicals, exceeding renal antioxidant mechanisms and causing cellular damage (Zhang et al. 2022). The initial line of defense against ROS consists of SOD, CAT, and GPx. Superoxide radicals are dismutated by SOD into hydrogen peroxide, which is then broken

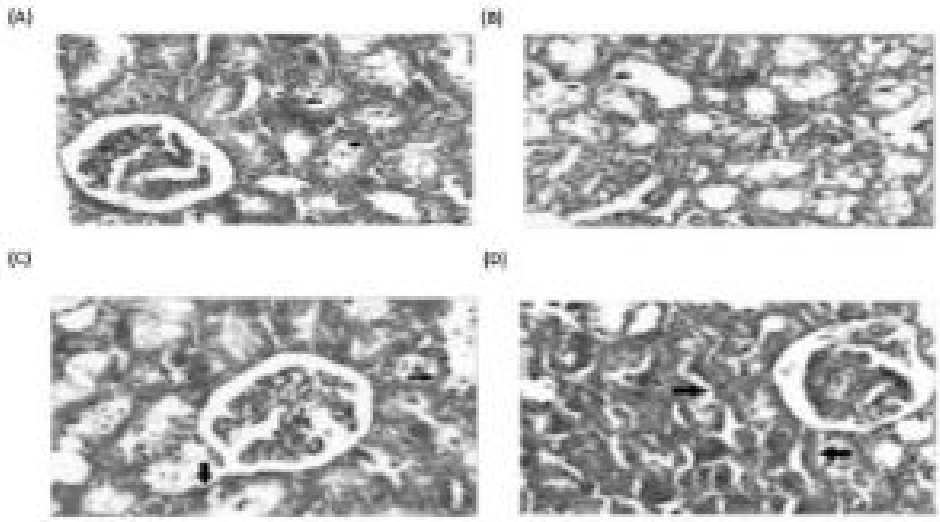

Fig. 1. *Agaricus bisporus's* impact on histopathological analysis in rats with cisplatin-induced nephrotoxicity
(A) Tubular epithelial cells and glomeruli are arranged normally in a section of a normal rat kidney (Group I)
(B) Tubular epithelial cells and glomeruli are severely necrotic in a section of a cisplatin-treated rat kidney (Group II)
(C) Tubular epithelial cells and glomeruli in a portion of a rat kidney treated with silymarin (Group III) exhibit regenerative alterations
(D) Tubular epithelial cells and glomeruli in a portion of a rat kidney treated with Agaricus bisporus (Group IV) exhibit full regenerative alterations

down into water by CAT and GPx. When these enzymes are compromised, as seen in cisplatin toxicity, ROS accumulation facilitates lipid peroxidation, DNA damage, and protein oxidation, leading to tubular cell injury (Deng et al. 2023). The observed reduction in antioxidant enzyme activity in group II (cisplatin alone) reflects this well-established pathological cascade.

Multiple studies corroborate these results. In cisplatin-treated rodent models, significant declines in renal SOD activity were consistently reported, often accompanied by reductions in CAT and GPx activities (Atessahin et al. 2005; Li et al. 2022). These declines are attributed to oxidative depletion of enzyme cofactors and direct inhibition by ROS, which impairs the catalytic functions of antioxidant proteins. Furthermore, cisplatin has been shown to reduce renal glutathione (GSH), an essential cofactor for GPx function, further exacerbating oxidative vulnerability (Peres and da Cunha 2013). Thus, diminished antioxidant enzyme activities are characteristic biochemical hallmarks of cisplatin nephrotoxicity, and reversal of these changes represents a meaningful marker of nephroprotection.

The observed increase in SOD, CAT, and GPx activities in *A. bisporus*-treated rats (group IV) suggests that this fungus has a capacity to enhance the renal antioxidant defense system in relation to oxidative damage brought on by cisplatin. This effect could be explained by the rich bioactive profile of *A. bisporus*, which is known to contain high levels of phenolic compounds, flavonoids, ergothioneine,β-glucans, and other antioxidants (Kalaras et al. 2017). Ergothioneine, a thiol antioxidant highly concentrated in mushrooms, has unique ROS-scavenging properties and accumulates preferentially in tissues prone to oxidative damage (Cheah and Halliwell 2012). Ergothioneine has been shown to protect mitochondrial and nuclear DNA from oxidative harm, which is particularly relevant in kidneys, where cisplatin disrupts mitochondrial function, leading to ROS overproduction (Yang et al. 2023).

Following *A. bisporus* treatment, increased gene expression of antioxidant enzymes mediated by the nuclear factor erythroid 2-related factor 2 (Nrf2) pathway may also lead to increases in CAT and GPx activity. Nrf2 is a master transcriptional regulator of endogenous antioxidant defense and phase II detoxification enzymes. Several dietary antioxidants and phytochemicals are known to activate Nrf2, leading to upregulation of SOD, CAT, and GPx gene expression. Recent research indicates that bioactive mushroom extracts can stimulate Nrf2 signaling, thereby promoting antioxidant enzyme synthesis and strengthening cellular resilience to oxidants (Li et al. 2023; Rahman et al. 2022). Although direct measurement of Nrf2 activation was not performed in the present study, the elevation of these enzymatic levels suggests that *A. bisporus* may exert modulatory effects at both transcriptional and post-transcriptional levels.

The protective effect of *A. bisporus* was comparable to that of silymarin in group III, a well-studied antioxidant known for its ability to ameliorate oxidative tissue injury. Silybum marianum is the source of the flavonolignan complex silymarin, has been shown to attenuate cisplatin nephrotoxicity by restoring antioxidant enzyme activities, reducing lipid peroxidation, and suppressing pro-inflammatory signalling pathways (Atessahin et al. 2005). The fact that both *A. bisporus* and silymarin significantly enhanced antioxidant enzyme levels underscores that the underlying protective mechanism in this model is largely dependent on attenuating oxidative stress.

Comparison with other natural antioxidants further validates the potential therapeutic relevance of *A. bisporus*. Numerous substances derived from plants, including as quercetin, resveratrol, and curcumin, have been demonstrated to lessen kidney damage caused by cisplatin by upregulating SOD, CAT, and GPx activities and reducing ROS generation (Ozyilmaz et al. 2021; Sharma et al. 2020). The consistent pattern across these diverse antioxidants suggests that enhancing enzymatic antioxidant capacity is a fundamental strategy to counteract cisplatin nephrotoxicity. It further highlights the significance of dietary bioactives, like those found in *A. bisporus*, as accessible and low-toxicity adjuncts in chemoprotective regimens.

Another important consideration is that enhanced antioxidant enzyme activities in *A. bisporus*-treated rats likely contributed to improvements in other biochemical and histopathological markers of renal function (data not shown here but often reported in similar studies). Restoration of SOD, CAT and GPx not only reduces

ROS burden but also prevents subsequent lipid peroxidation and membrane destabilisation, preserving renal cell integrity. Other nephroprotection studies using mushroom extracts have shown that this biochemical improvement can result in decreased serum indicators of kidney injury, such as creatinine and blood urea nitrogen (BUN) (Zhang et al. 2022).

However, while the findings of the present study support a robust antioxidant effect of *A. bisporus*, several limitations and future directions merit discussion. First, measurement of gene expression levels of antioxidant enzymes and transcription factors like Nrf2 would provide deeper insights into the molecular mechanisms. Second, identification and quantification of specific bioactive compounds in the *A. bisporus* extract would allow correlation between compound potency and biological efficacy. Finally, expanding research to include other models of renal injury (for example, ischemia-reperfusion, diabetic nephropathy) would establish broader nephroprotective applicability.

The observed increase in SOD, CAT and GPx activities in *A. bisporus*-treated rats (group IV) suggests that this fungus has a capacity to enhance the renal antioxidant defense system in relation to oxidative damage brought on by cisplatin. This effect could be explained by the rich bioactive profile of *A. bisporus*, which is known to contain high levels of phenolic compounds, flavonoids, ergothioneine, â-glucans, and other antioxidants (Kalaras et al. 2017). Ergothioneine, a thiol antioxidant highly concentrated in mushrooms, has unique ROS-scavenging properties and accumulates preferentially in tissues prone to oxidative damage (Cheah and Halliwell 2012). Ergothioneine has been shown to protect mitochondrial and nuclear DNA from oxidative harm, which is particularly relevant in kidneys, where cisplatin disrupts mitochondrial function, leading to ROS overproduction (Yang et al. 2023).

Increases in CAT and GPx activities following *A. bisporus* treatment may also result from enhanced gene expression of antioxidant enzymes mediated by the nuclear factor erythroid 2-related factor 2 (Nrf2) pathway. Nrf2 is a master transcriptional regulator of endogenous antioxidant defense and phase II detoxification enzymes. Several dietary antioxidants and phytochemicals are known to activate Nrf2, leading to upregulation of SOD, CAT, and GPx gene expression. Recent research indicates that bioactive mushroom extracts can stimulate Nrf2 signalling, thereby promoting antioxidant enzyme synthesis and strengthening cellular resilience to oxidants (Li et al. 2023; Rahman et al. 2022). Although direct measurement of Nrf2 activation was not performed in the present study, the elevation of these enzymatic levels suggests that *A. bisporus* may exert modulatory effects at both transcriptional and post-transcriptional levels.

The protective effect of *A. bisporus* was comparable to that of silymarin in group III, a well-studied antioxidant known for its ability to ameliorate oxidative tissue injury. Silybum marianum is the source of the flavonolignan complex silymarin, has been shown to attenuate cisplatin nephrotoxicity by restoring antioxidant enzyme activities, reducing lipid peroxidation, and suppressing pro-inflammatory signalling pathways (Atessahin et al. 2005). The fact that both *A. bisporus* and silymarin significantly enhanced antioxidant enzyme levels underscores that the underlying protective mechanism in this model is largely dependent on attenuating oxidative stress.

Comparison with other natural antioxidants further validates the potential therapeutic relevance of *A. bisporus*. By increasing SOD, CAT and GPx activities and lowering ROS production, a variety of plant-derived substances, such as curcumin, resveratrol, and quercetin, have been demonstrated to lessen cisplatin-induced kidney damage (Ozyilmaz et al. 2021; Sharma et al. 2020). The consistent pattern across these diverse antioxidants suggests that enhancing enzymatic antioxidant capacity is a fundamental strategy to counteract cisplatin nephrotoxicity. It further highlights the significance of dietary bioactives, like those found in *A. bisporus*, as accessible and low-toxicity adjuncts in chemoprotective regimens.

Another important consideration is that enhanced antioxidant enzyme activities in *A. bisporus*-treated rats likely contributed to improvements in other biochemical and histopathological markers of renal function (data not shown here but often reported in similar studies). Restoration of SOD, CAT and GPx not only reduces ROS burden but also prevents subsequent lipid peroxidation and membrane destabilisation,

preserving renal cell integrity. Other nephroprotection studies using mushroom extracts have shown that this biochemical improvement can result in decreased serum indicators of kidney injury, such as creatinine and blood urea nitrogen (BUN) (Zhang et al. 2022).

However, while the findings of the present study support a robust antioxidant effect of *A. bisporus*, several limitations and future directions merit discussion. First, measurement of gene expression levels of antioxidant enzymes and transcription factors like Nrf2 would provide deeper insights into the molecular mechanisms. Second, identification and quantification of specific bioactive compounds in the *A. bisporus* extract would allow correlation between compound potency and biological efficacy. Finally, expanding research to include other models of renal injury (for example, ischemia-reperfusion, diabetic nephropathy) would establish broader nephroprotective applicability.

Additionally, the study's findings showed that cisplatin significantly reduced GSH, raised MDA generation in the renal tissue, and reduced the activity of the antioxidant enzymes SOD and CAT. These results align with the findings of Yadav et al. (1997), and Fouad et al. (2012). Cisplatin nephrotoxicity is evidently brought on by oxidative stress and increased formation of superoxide anion, hydrogen peroxide, and hydroxyl radicals due to enhanced activity of NADPH oxidase, xanthine oxidase, and adenosine deaminase (Lalila et al. 2001). These free radicals cause enzymatic inactivation by denaturing and peroxiding the lipid components of the cell membrane (El Arem et al. 2014).

Additionally, Cisplatin-induced lipid peroxidation in the kidney of rats was considerably reduced by Berne date extract, as seen by increased GSH content, decreased MDA content, and improved CAT and SOD enzymatic activity. The study's findings are consistent with those of El Arem et al. (2014), who discovered that date fruit aqueous extract had a nephroprotective impact on rats exposed to oxidative stress brought on by trichloroacetic acid, as evidenced by a drop in MDA levels and an increase in antioxidant enzyme activity. Similar findings were reported by Saafi-Been Salah et al. (2012), who demonstrated the date palm fruit extract's antioxidant efficacy against oxidative stress and dimethoate-induced nephrotoxicity in rats.

In this work, the researchers tried to find out how a methanolic extract of Agaricus bisporus affected the nephrotoxicity that cisplatin caused in rats. Cisplatin-induced rats had higher levels of reactive oxygen species (ROS), free radicals like superoxide anion, hydrogen peroxide, and hydroxyl radical, as well as a decrease in the synthesis of antioxidant enzymes because of the accumulation of cisplatin in the proximal and distal nephrons (Elberry et al. 2011). Superoxide is converted to hydrogen peroxide, a considerably less reactive chemical that acts as the first line of defense against damage caused by ROS, by the primary ROS generated by metalloenzymes called SOD (Bartelink et al. 2002). Chronic renal failure is mostly caused by oxidative stress. Due to the kidneys' loss of copper and zinc, which are necessary for enzyme function, animals treated with cisplatin may have reduced tissue SOD levels. In contrast to the cisplatin-induced group, silymarin and Agaricus bisporus therapy significantly increased SOD levels in kidney tissue in the current study. The breakdown of hydrogen p is catalysed by the ubiquitous enzyme catalase (CAT). CAT is very good in preventing different ROS-mediated damage and may shield the kidney from nephrotoxicity brought on by cisplatin (Vaziri et al. 2003). The tissue CAT level is lower in the cisplatin-induced animals than in the cisplatin-induced group. However, silymarin and Agaricus bisporus therapy markedly raised the CAT level in kidney tissue, indicating its antioxidant activity during nephrotoxicity. The most significant antioxidant enzyme in humans is glutathione peroxidase (GPx), it has a high expression level in the kidney. It shields the body from oxidative damage by scavenging and inactivating lipid and hydrogen peroxides, as well as by eliminating peroxides and peroxynitrite that may harm the kidneys (Chirino et al. 2004). Agaricus bisporus and silymarin therapy dramatically raised kidney tissue GPx levels, demonstrating their antioxidant effectiveness against cisplatin- induced oxidative stress.

Lipid peroxidation (LPO), which is mostly brought on by a range of ROS, such as hydroxyl and hydrogen peroxide radicals, is produced in trace amounts by the body naturally. An increase in the concentration of LPO end products indicates the role of free radicals in human disease

(Ma et al. 2007). Proteins and DNA are more important targets of oxidative stress damage than lipids and LPO, which frequently happen later in the injury process (De Haan et al. 2005). The LPO level in kidney tissue is reportedly elevated by the production of free radicals from cisplatin-mediated renal tissue injury. This is directly related to elevated LPO levels in nephrotoxic conditions. By significantly reducing the kidney LPO level and halting the production of free radicals brought on by cisplatin-induced nephrotoxicity, *A. bisporus* treatment proved its protective role in preventing renal damage. The popular drug silymarin also has a similar effect during nephrotoxicity. The findings from this investigation also made it abundantly clear that there was a noticeable enlargement of the proximal convoluted tubules and that the tubular epithelium was desquamated, resulting in the sloughing off of nearly all of the epithelium. Increased tissue in the interstitium and cellular debris in the tubular lumen are other signs of renal necrosis brought on by cisplatin (Sener et al. 2006). The glomerulus and renal tubular necrosis are shielded from the effects of cisplatin-treated group rats by *A. bisporus* therapy.

Recent studies have further reinforced the central role of oxidative stress in cisplatin-induced nephrotoxicity and highlighted the therapeutic potential of natural antioxidants in mitigating renal injury. For example, Zhang et al. (2023) reported that antioxidant-rich natural products significantly restored renal antioxidant enzyme activities while reducing lipid peroxidation and inflammatory responses in cisplatin-treated experimental animals. Similarly, Rahman et al. (2024) demonstrated that dietary bioactive compounds capable of activating endogenous antioxidant pathways effectively attenuated cisplatin-mediated oxidative damage and preserved renal architecture. These findings are in agreement with the present study, where *A. bisporus* treatment significantly enhanced SOD, CAT, and GPx activities while reducing oxidative stress-associated renal damage.

The observed nephroprotective effects of *A. bisporus* are also consistent with growing evidence supporting the medicinal value of edible mushrooms. Recent investigations have shown that mushroom-derived bioactive compounds, including ergothioneine, polysaccharides, phenolic acids, and flavonoids, possess potent antioxidant and cytoprotective properties (Kalaras et al. 2017; Yang et al. 2023). In a recent review, Sharma and Kaur (2024) highlighted that edible mushrooms can modulate oxidative stress, inflammatory pathways, and mitochondrial dysfunction, thereby providing protection against various forms of organ toxicity. The improvement in antioxidant enzyme activities observed in the current study may therefore be attributed to the synergistic action of multiple bioactive constituents present in *A. bisporus*.

Furthermore, increasing attention has been directed toward the role of the Nrf2 signalling pathway in protecting against cisplatin-induced renal injury. Activation of Nrf2 promotes the transcription of several antioxidant and detoxifying enzymes, thereby enhancing cellular resistance to oxidative stress. Recent studies have demonstrated that mushroom-derived phytochemicals can activate Nrf2-mediated antioxidant responses and reduce renal oxidative damage (Li et al. 2023; Rahman et al. 2022). Although Nrf2 expression was not evaluated in the present investigation, the significant restoration of antioxidant enzyme activities suggests that this pathway may contribute to the nephroprotective effects of *A. bisporus* and warrants further investigation.

The present findings also compare favourably with studies evaluating other natural nephroprotective agents. Curcumin, quercetin, resveratrol, and several plant-derived polyphenols have been reported to attenuate cisplatin-induced nephrotoxicity through restoration of antioxidant enzyme activities and suppression of lipid peroxidation (Ozyilmaz et al. 2021; Sharma et al. 2020). The magnitude of antioxidant improvement observed with *A. bisporus* in the current study suggests that edible mushrooms may represent an additional promising source of renoprotectivebioactives. Importantly, unlike many purified phytochemicals, *A. bisporus* is widely consumed as a dietary component, which may facilitate its future translational application as a complementary nutritional intervention.

Despite substantial progress in understanding cisplatin nephrotoxicity, important knowledge gaps remain. Most previous studies have focused primarily on synthetic antioxidants or isolated phytochemicals, while relatively few investigations have examined the nephroprotec-

tive potential of commonly consumed edible mushrooms. Moreover, mechanistic studies exploring the relationship between mushroom bioactives, oxidative stress modulation, and renal tissue preservation remain limited. Therefore, the present study contributes to the growing body of evidence by demonstrating that methanolic extract of *A. bisporus* effectively ameliorates cisplatin-induced oxidative stress, restores endogenous antioxidant defenses, reduces lipid peroxidation, and improves renal histopathological alterations. These findings provide further support for the development of mushroom-based functional foods and nutraceuticals as potential adjunctive strategies for reducing chemotherapy-associated renal toxicity.

Future investigations should focus on characterizing the specific bioactive compounds responsible for the observed nephroprotective effects, evaluating their molecular targets, and examining their influence on signalling pathways involved in oxidative stress, inflammation, apoptosis, and mitochondrial dysfunction. Additionally, studies involving gene-expression profiling, protein-level analyses, and clinical translation will be necessary to establish the therapeutic relevance of *A. bisporus* in the prevention of cisplatin-associated kidney injury.

CONCLUSION

In an experimental model system, the current study discovered that Agaricus bisporus methanolic extract showed potential antioxidant activity. The findings show that the antioxidant, anti-inflammatory, and antiapoptotic properties of Berne date extract shield kidney tissue from cisplatin-induced nephrotoxicity in rats. Consequently, kidney impairment, a significant and dose-limiting issue when using cisplatin, can be avoided with Berne date extract. The crude extract's phenols, flavonoids like quercetin, and other phytochemical components may be responsible for the antioxidant activity. Future clinical treatments for patients with renal failure may be significantly impacted by additional research into Agaricus bisporus's mode of action.

RECOMMENDATIONS

Future clinical treatments for individuals with renal failure may be significantly impacted by additional research into the mechanism of action of *Agaricus bisporus*.

REFERENCES

Al-Hashem FH 2009. Camel's milk protects against aluminum chloride-induced toxicity in the liver and kidney of white albino rats. *American Journal of Biochemistry and Biotechnology,* 5(3): 98-108. https://doi.org/10.3844/ajbbsp.2009.98.108

Atessahin A, Yilmaz S, Karahan I, Ceribasi AO, Sakin F 2005. Lycopene counteracts cisplatin-induced nephrotoxicity and oxidative stress in rats. *Toxicology,* 212(2-3): 116-123. https://doi.org/10.1016/j.tox.2005.04.003

Bartelink IH, Rademaker CM, Schobben AF, van den Anker JN 2002. Guidelines on paediatric dosing on the basis of developmental physiology and pharmacokinetic considerations. *Clinical Pharmacokinetics,* 41(9): 705-718. https://doi.org/10.2165/00003088-200241090-00005

Cheah IK, Halliwell B 2012. Ergothioneine; antioxidant potential, physiological function and role in disease. *Biochimica et Biophysica Acta (BBA) – Molecular Basis of Disease*, 1822(5): 784-793. https://doi.org/10.1016/j.bbadis.2011.09.017

Chirino YI, Hernández-Pando R, Pedraza-Chaverrí J 2004. Peroxynitrite decomposition catalyst ameliorates renal damage and protein nitration in cisplatin-induced nephrotoxicity in rats. *BMC Pharmacology,* 4: 20. https://doi.org/10.1186/1471-2210-4-20

De Haan JB, Bladier C, Lotfi-Miri M, Taylor J, Hutchinson P, Crack PJ, Hertzog P, Kola I 2005. Fibroblasts derived from Gpx1 knockout mice display attenuated upregulation of Nrf2 target genes in response to electrophilic stress. *Free Radical Biology and Medicine,* 38(10): 1400-1408. https://doi.org/10.1016/j.freeradbiomed.2005.01.032

Deng F, Sharma I, Bhatt K, Yang S, Zhou J, Kasinat BS, Bhatt DL 2023. NLRP3 inflammasome activation and ferroptosis are implicated in cisplatin-induced acute kidney injury. *Free Radical Biology and Medicine,* 194: 311-323. https://doi.org/10.1016/j.freeradbiomed.2022.12.004

El Arem A, Ghrairi F, Lahouar L, Thouri A, Zekri M, Ayed A, Zouari N, Ferchichi S, Marzouk B, Achour L 2014. Protective effects of date fruit extract (Phoenix dactylifera L.) on trichloroacetic acid-induced nephrotoxicity in adult rats. *Journal of Functional Foods,* 8: 175-184. https://doi.org/10.1016/j.jff.2014.03.007

Elberry AA, Harraz FM, Ghareib SA, Gabr SA, Nagy AA, Abdel-Sattar E 2011. Methanolic extract of Marrubium vulgare ameliorates cisplatin-induced hepatotoxicity and nephrotoxicity in rats. *Journal of Medicinal Plants Research,* 5(2): 282-289.

Elhusseiny SM, El-Desouky MA, Awad NS, Hassan ME 2023. Agaricusbisporus and Pleurotusostreatus polysaccharides mitigate doxorubicin-induced nephrotoxicity

via anti-oxidative and anti-apoptotic mechanisms in rats. *BMC Complementary Medicine and Therapies,* 23(1): 108. https://doi.org/10.1186/s12906-023-03934-9

Fouad AA, Al-Mulhim AS, Jresat I 2012. Therapeutic effect of coenzyme Q10 against experimentally-induced hepatocellular carcinoma in rats. *Environmental Toxicology and Pharmacology,* 33(2): 265-274. https://doi.org/10.1016/j.etap.2011.12.018

Halliwell B, Aeschbach R, Löliger J, Aruoma O I 1995. The characterization of antioxidants. *Food and Chemical Toxicology,* 33(7): 601-617. https://doi.org/10.1016/0278-6915(95)00024-V

HeathRL, Packer L 1968. Photoperoxidation in isolated chloroplasts: I. Kinetics and stoichiometry of fatty acid peroxidation. *Archives of Biochemistry and Biophysics,* 125(1): 189-198. https://doi.org/10.1016/0003-9861(68)90654-1

Kakkar P, Das B, Viswanathan PN 1984. A modified spectrophotometric assay of superoxide dismutase. *Indian Journal of Biochemistry and Biophysics,* 21(2): 130-132.

KalarasMD, Richie JP, Calcagnotto A, Beelman RB 2017. Mushrooms: A rich source of the antioxidants ergothioneine and glutathione. *Food Chemistry,* 233: 429-433. https://doi.org/10.1016/j.foodchem.2017.04.109

Lalila T, Ahmed M, Mohan G 2001. Role of oxidative stress in cisplatin-induced nephrotoxicity. *Indian Journal of Pharmacology,* 33(3): 183-188.

Li J, Cao F, Yin H, Huang Z, Lin Z, Mao N, Sun B, Wang G 2022. Ferroptosis: Past, present and future. *Cell Death and Disease,* 11(2): 88. https://doi.org/10.1038/s41419-020-2298-2

Li W, Shen X, Liu B, Sun K, Yin S, Zhu X 2023. Polysaccharides from Agaricusbisporus alleviate oxidative stress and inflammation through Nrf2 and NF-êB pathways in LPS-stimulated RAW264.7 macrophages. *Frontiers in Immunology,* 14: 1123895. https://doi.org/10.3389/fimmu.2023.1123895

Li X, Wang Y, Chen Z, et al. 2023. Mushroom-derived bioactive compounds attenuate oxidative stress through Nrf2 activation: Emerging therapeutic perspectives. *International Journal of Biological Macromolecules*, 236: 123945.

Ma Q, Cavallin LE, Yan B, Zhu S, Duran EM, Wang H, Bhatt DL, Bhatt D, Plummer P, Bhatt S 2007. Antitumorigenesis of antioxidants in a transgenic Rac1 Model Of Kaposi's Sarcoma. *Proceedings of the National Academy of Sciences of the United States of America,* 106(21): 8683-8688. https://doi.org/10.1073/pnas.0812688106

Ozyilmaz E, Gul M, Gul HH, Erdogan S, Yildirim A, Uz E 2021. Resveratrol attenuates cisplatin-induced renal and liver damage in rats by reducing oxidative stress markers. *Korean Journal ofPhysiology and Pharmacology,* 25(3): 247-255. https://doi.org/10.4196/kjpp.2021.25.3.247

Peres LAB, da Cunha AD 2013. Acute nephrotoxicity of platinum compounds. *Journal of Brasileirodenefrologia,* 35(4): 332-340. https://doi.org/10.5935/0101-2800.20130054

Rahman T, Choudhury MR, Foysal MJ, Rahman MM, Rahman A 2022. Mushroom polysaccharides as potential candidates for alleviating cisplatin-induced nephrotoxicity: A systematic review. *Molecules,* 27(14): 4375. https://doi.org/10.3390/molecules27144375

Rahman MM, Islam MT, Uddin SJ, et al. 2024. Natural antioxidants as promising therapeutic agents against cisplatin-induced nephrotoxicity: Recent advances and future perspectives. *Biomedicine & Pharmacotherapy*, 170: 115921.

Rotruck JT, Pope AL, Ganther HE, Swanson AB, Hafeman DG, Hoekstra WG 1973. Selenium: Biochemical role as a component of glutathione peroxidase. *Science,* 179(4073): 588-590. https://doi.org/10.1126/science.179.4073.588

Saafi-Ben Salah E, Louati H, Soudani N, Hakim A, Zeghal N, Malki A, Boudawara T 2012. Protective effects of date palm fruit extract (Phoenix dactylifera L.) on nephrotoxicity induced by dimethoate in rats. *Food and Chemical Toxicology,* 50(9): 3255-3263. https://doi.org/10.1016/j.fct.2012.06.029

SenerG, Ekþioðlu-Demiralp E, Cetiner M, ErcanF, Yeðen BÇ 2006. â-Glucan ameliorates methotrexate-induced oxidative organ injury via its antioxidant and immunomodulatory effects. *European Journal of Pharmacology,* 542(1-3): 170-178. https://doi.org/10.1016/j.ejphar.2006.05.026

Sharma, N, Dubey A, Grover A 2020. Curcumin and quercetin synergistically attenuate cisplatin-induced nephrotoxicity by modulating oxidative stress and inflammation in rats. *Journal of Functional Foods,* 65: 103727. https://doi.org/10.1016/j.jff.2019.103727

Sharma R, Kaur S 2024. Medicinal and edible mushrooms as sources of antioxidant and nephroprotective bioactive compounds: A comprehensive review. *Food Chemistry*, 446: 138856.

Sinha AK 1972. Colorimetric assay of catalase. *Analytical Biochemistry,* 47(2): 389-394. https://doi.org/10.1016/0003-2697(72)90132-7

Vaziri ND, Dicus M, Ho ND, Boroujerdi-Rad L, Sindhu RK 2003. Oxidative stress and dysregulation of superoxide dismutase and NADPH oxidase in renal insufficiency. *Kidney International,* 63(1): 179-185. https://doi.org/10.1046/j.1523-1755.2003.00702.x

Yadav P, Sarkar S, Bhatnagar D 1997. Action of capparis decidua against alloxan-induced oxidative stress and diabetes in rat tissues. *Pharmacological Research,* 36(3): 221-228. https://doi.org/10.1006/phrs.1997.0222

Yang Z, Wei X, Pan Y, Min J, Li J, Hu J, Xu S, Dong Z 2023. Cisplatin-induced mitochondrial dysfunction and ferroptosis in renal proximal tubular cells. *Free Radical Biology and Medicine,* 194: 107-119. https://doi.org/10.1016/j.freeradbiomed.2022.11.039

Yang J, Liu H, Zhang Q, et al. 2023. Ergothioneine protects against oxidative stress-induced mitochondrial dysfunction and tissue injury. *Antioxidants*, 12(4): 891.

Zhang J, Shi L, Zheng H, Li G, Liu D, Li B, Huang J 2022. Polysaccharides from Agaricusbisporus alleviate cisplatin-induced nephrotoxicity by reducing oxidative stress and inflammation in mice. *Food and Function,* 13(4): 2032-2044. https://doi.org/10.1039/D1FO03438A

Zhang Y, Chen L, Zhao H, et al. 2023. Protective effects of natural antioxidants against cisplatin-induced renal injury: Mechanisms and therapeutic implications. *Frontiers in Pharmacology*, 14: 1265784.

Paper received for publication in October, 2025
Paper accepted for publication in April, 2026