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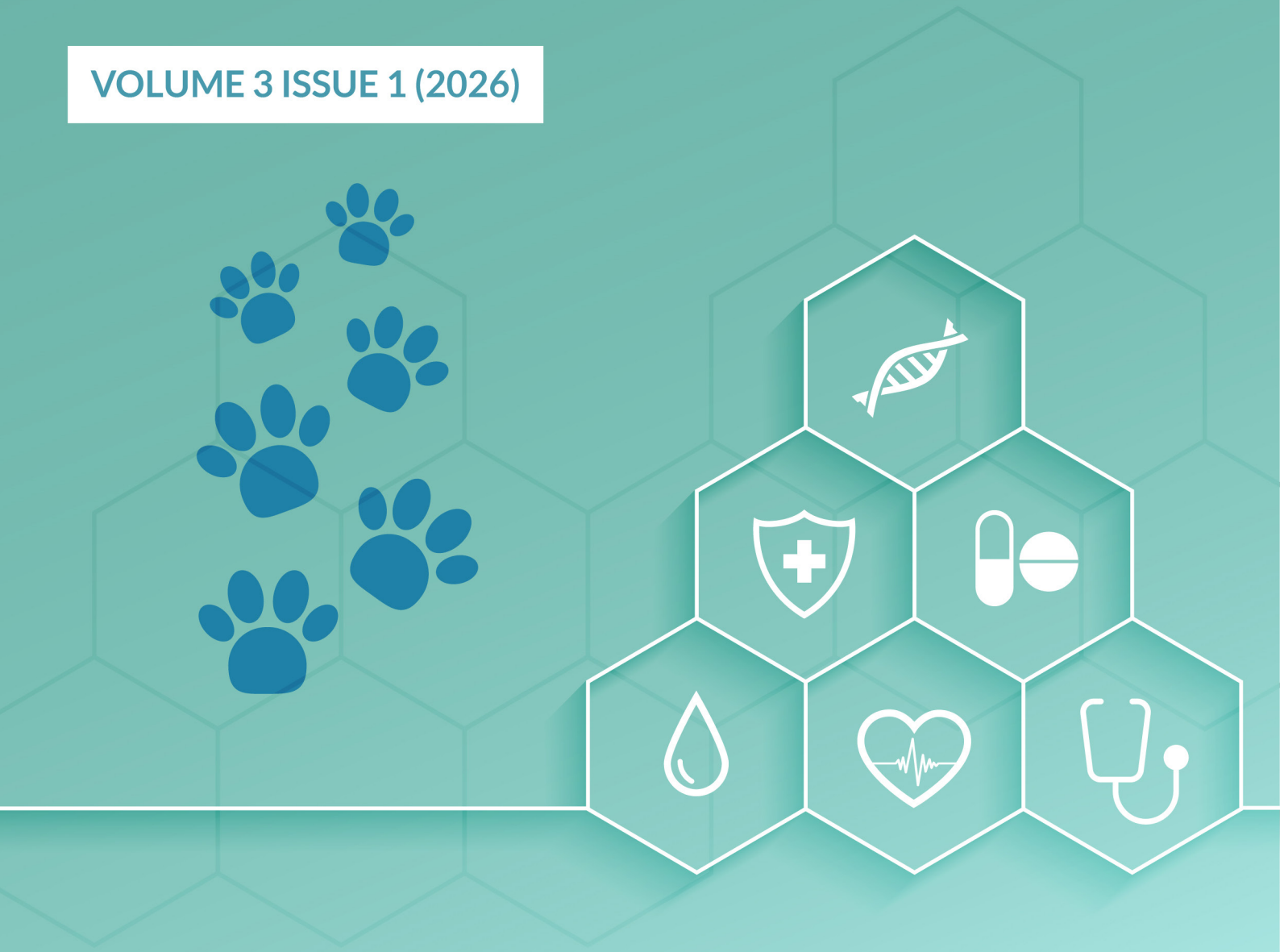

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Tiger Fern Leaves Juice Attenuates Arsenic-Induced Neurobehavioral and Hepatic Disorders in Mice

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ABSTRACT

Age-related arsenic poisoning is a worldwide environmental health issue linked to neurobehavioral dysfunction and acute liver dysfunction, which is mainly mediated by oxidative stress and disruption of antioxidant activities. Natural plants present positive protective strategies, but the neuroprotective or hepatoprotective effects of tiger fern leaves juice (TFJ) have not been studied yet. The paper tested the neurobehavioral changes and hepatic toxicity of arsenic induced by tiger fern leaves juice in mice. Thirty-six adult Swiss albino male mice were randomly placed in six groups (n = 6/group), and orally fed for 28 days with distilled water (control), sodium arsenite (10 mg/kg), TFJ (5 or 10 mL/kg), or arsenic mixed with TFJ. Neurobehavioral evaluations were the Morris water maze, elevated plus maze and rotarod evaluations. The level of hepatic oxidative stress was assessed through malondialdehyde (MDA), superoxide dismutases (SOD), and catalases (CAT) activities. The liver functioning was evaluated by means of the serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP) levels. Arsenic exposure significantly deteriorated cognitive and motor activity, and it raised escape latency in the Morris water maze by around 108 per cent and hepatic lipid peroxidation by around 190 per cent, as well as antioxidant enzyme activities. The co-administration of tiger fern leaf juice was of significant effect in inhibiting these effects; the escape latency was attenuated by as much as 46%, lipid peroxidation by about 55%, and the antioxidant status was restored in a dose-dependent manner. Also, TFJ treatment had significant ameliorating effects on the increased serum liver enzyme levels, which are factors of hepatic functioning. Tiger fern leaves juice has a vigorous neuroprotective and hepatoprotective activity against the toxicity caused by arsenic, which is probably due to its antioxidant and cytoprotective properties. TFJ can be a good natural therapeutic candidate to treat the arsenic-related neurobehavioral and hepatic disorders.

INTRODUCTION

Arsenic is a widely spreading environmental pollutant and a significant public health hazard in the world, especially in areas where groundwater is being contaminated. The inorganic arsenic is mainly accumulated in various organs as a result of chronic exposure (drinking water and eating food) and results in systemic toxicity. In recent years, arsenic has also been acknowledged as a significant neurotoxicant and hepatotoxicant as well as a carcinogen, which is capable of causing functional and structural dysfunction in the exposed population in the long term (Ganie *et al.*, 2024). Arsenic exposure remains a significant issue despite global mitigation efforts, underscoring the importance of prevention and treatment.

Arsenic toxicity mainly affects the central nervous system because of its great metabolic rate, lipid-rich structure, and low regenerative ability. According to the experimental research, chronic arsenic exposure is associated with the disruption of cognitive functions, including learning and memory, motor coordination, and the encouragement of anxiety-like behaviours in animal models (Vázquez Cervantes *et al.*, 2023). The causes of these neurobehavioral changes have been associated with the prolific production of reactive oxygen species, mitochondrial dysfunction, activation of synaptic dysregulation, and neuroinflammatory reactions. Arsenic is also known to disrupt neurotransmitter communication and calcium homeostasis and impair neuronal plasticity and behavioural performance. Nonetheless, the exact pathophysiologic processes occurring in response to arsenic-related neurobehavioral impairment are not fully comprehended, and there are still no effective interventions.

The liver is a central site of arsenic metabolism and detoxification; thus, it is a vital target organ in arsenic toxicity. Arsenic hepatic biotransformation includes redox cycling and methylation steps that result in the generation of reactive intermediates, which cause oxidative stress and injury to the cells. A variety of studies have found that exposure to arsenic increased the lipid peroxidation level, inhibited the activity of endogenous antioxidant enzymes, including superoxide dismutase and catalase, and impaired the normal liver functioning, which was reflected in the altered level of serum transaminase (Dey *et al.*, 2020; Hu *et al.*, 2020). Notably, there is some evidence that points to a two-way association between neurotoxicity and hepatic dysfunction, also known as the liver-brain axis. This could also be increased by

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hepatic oxidative stress and metabolic perturbation, which support systemic inflammation and oxidative load, and the necessity to assess both neurological and hepatic outcomes in models of arsenic toxicity (Jomova *et al.*, 2011).

Antioxidant-based interventions have become quite popular as one of the possible protective measures in the face of the multifactorial nature of arsenic-induced toxicity. Over recent years, natural products based on plants have become an emerging candidate as they have various bioactive components and are relatively safe, as well as capable of simultaneously regulating several molecular pathways. Phytochemicals, including flavonoids, phenolic acids, and tannins, have been shown to have neuroprotective and hepatoprotective properties, such as scavenging reactive oxygen species, enhancing endogenous antioxidant protection, and suppressing inflammatory signalling pathways (Rudrapal *et al.*, 2022). To a large extent, however, the literature available has concentrated on individually prepared compounds or solvent-based extracts and could not accurately capture the complexity and interactions in whole-plant extracts in traditional ethnomedicine (Clemente-Suárez *et al.*, 2025).

Minimally processed whole-plant juices maintain a wide range of water-soluble bioactive compounds and could be more bioavailable and show greater synergistic activity than purified extracts do (Oliveira *et al.*, 2025). Although such preparations are widespread in traditional medicinal systems, scientific evidence on the preparation is scarce. Specifically, fern species are an understudied category of medicinal plants that are reported to have anti-oxidant, anti-inflammatory, and detoxifying effects. One of the ethnomedicinally used fern species in some traditional medicine has been the tiger fern, which has been used traditionally as fresh juice to maintain general health and detoxification effects (El-Saadony *et al.*, 2025). Early phytochemical studies indicate that fern leaves have possibly high concentrations of polyphenols and antioxidant activity, but little has been done to systematise their pharmacological effects.

The current research study was aimed at exploring the impact of tiger fern leaf juice on arsenic-induced neurobehavioral changes and liver dysfunction in mice. The proposed research seeks to provide complete experimental data on the neuroprotective and hepatoprotective effects of tiger fern leaves juice by evaluating cognitive performance, anxiety-like behaviour, motor coordination, hepatic oxidative stress markers, and liver enzymes of liver functioning. The results of the presented study can be used in the development of research on the topic of plant-based intervention in heavy metal toxicity and contribute to further investigation of tiger fern as a possible natural treatment.

MATERIALS AND METHODS

Study Design, Randomisation, and Blinding

To determine the effects of tiger fern leaves juice (TFJ) on the neurobehavioral and hepatic toxicity caused by sodium arsenite in mice, a 28-day randomised, controlled experimental study was carried out to determine the protective effects of the tiger fern leaves juice against the toxicity. Simple randomisation was used to assign animals to treatment groups. Investigators who conducted behavioural and biochemical studies were not aware of group assignment. Daily manipulation was carried out in acclimatisation and dosing to reduce variability caused by stress. The exclusion criteria were treatment-unrelated illness, dosing issues (regurgitation or aspiration) and any behavioural data that could not be scored.

Experimental Animals and Husbandry

Thirty-six adult male Swiss albino mice (25–30 g) were randomly divided into six groups (n = 6). Animals were housed in polypropylene cages under standard laboratory conditions (22 ± 2 °C; 50–60% humidity; 12 h light/dark cycle) with sterile bedding and free access to food and water. Body weight was recorded weekly for dose adjustment and health monitoring. Routine handling and enrichment were applied to reduce anxiety-related behavioural artefacts.

Chemicals and Reagents

Sodium arsenite ($NaAsO_2$, analytical grade) was used to induce toxicity. Reagents for oxidative stress assays included phosphate buffer, thiobarbituric acid, trichloroacetic acid, and hydrogen peroxide. Commercial diagnostic kits were used to measure serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP) following manufacturer instructions. All assays were performed in duplicate.

Preparation of Tiger Fern Leaves Juice (TFJ)

Fresh leaves of tiger fern were picked in a pesticide-free zone, washed, air-dried and prepared on the same day. Leaves were blended with non-heated distilled water (1:2 w/v) and filtered at a low temperature to get TFJ. The juice was prepared every day or kept in amber bottles at 4 °C and not more than 24 h. Calculations were made of doses that were based on the latest body weight.

Group Allocation and Dosing Regimen

Mice received oral gavage once daily for 28 days and were assigned to six groups: control (distilled water), arsenic (10 mg/kg/day), TFJ-low (5 mL/kg/day), TFJ-high (10 mL/kg/day), arsenic + TFJ-low, and arsenic + TFJ-high. Dosing was performed at the same time each day to minimise circadian effects. In co-treatment groups, TFJ was administered 30–60 min after sodium arsenite. Animals were observed post-gavage for adverse effects.

Neurobehavioral Assessment

The light phase was tested through behavioural testing in a temperature-controlled room, which was quiet after 30 min acclimatisation. Between trials, the surfaces of the apparatus were wiped down to eliminate olfactory cues. Tests were conducted in a low-stress series, where the elevated plus maze (EPM) and rotarod testing were conducted before the Morris water maze (MWM) probe trial.

Morris Water Maze (MWM)

The ability to learn space and memory was tested with the use of a circular pool (120-150 cm in diameter) filled with opaque water held at 24-26 °C. A platform underwater was introduced in a fixed target quadrant. The acquisition training took place in four days (days 24-27) with 3-4 trials per day. On day 28, a probe trial, in the absence of the platform, was carried out. Latency of escape during the process of acquisition and the time that the rat spent in the target quadrant during the probe trial were noted.

Elevated Plus Maze and Rotarod Tests

The elevated plus maze was used to evaluate the anxiety-like behaviour, and the time taken in the open and closed arms was measured for 5 minutes. The motor coordination was measured on the accelerating rotorod (4-40 rpm in 300 s). The mouse was trialled three times, and the average fall time was determined.

Blood Collection and Biochemical Assays

Mice were tested by behaviour and put to death after a night of starvation under anaesthesia. Blood was taken by cardiac puncture, centrifuged to get the serum, and analysed for ALT, AST, and ALP. Livers were sliced off, weighed and homogenised (10% w/v) in phosphate buffer. The occurrence of hepatic oxidative stress was determined by determining the malondialdehyde (MDA), superoxide dismutase (SOD) and catalase (CAT) activities, all relative to the amount of protein.

Statistical Analysis

Data are expressed as mean ± SEM. Group comparisons were performed using one-way analysis of variance (ANOVA) followed by Tukey's post-hoc test. A p-value < 0.05 was considered statistically significant. All behavioural and biochemical endpoints were analysed using standard statistical software.

RESULTS AND DISCUSSION

Exposure of mice to arsenic caused severe neurobehavior and hepatic dysfunction in the mice, but co-exposure of mice to tiger fern leaves juice (TFJ) reduced the effects significantly. Compared to controls, sodium arsenite deteriorated performance on the Morris water maze, plus maze, and rotarod, increased hepatic lipid peroxidation with depressed antioxidant enzyme activities, and increased serum liver enzymes. TFJ did not show adverse changes alone, and TFJ co-treatment positively influenced the behavioural data and hepatic oxidative and biochemical parameters, renewal dose-dependently.

Table 1 shows that sodium arsenite significantly impaired spatial learning and memory, as evidenced by a marked increase in escape latency (52.8 ± 3.7 s vs 25.4 ± 1.9 s in controls) and a reduction in time spent in the target quadrant (14.9 ± 1.3 s vs 33.6 ± 2.1 s). TFJ alone (5 and 10 mL/kg) produced performance comparable to controls, indicating no adverse cognitive effect. Co-administration of TFJ significantly attenuated arsenic-induced deficits in a dose-dependent manner, with the high dose improving escape latency (28.7 ± 2.1 s) and target quadrant time (29.8 ± 1.9 s) more effectively than the low dose (36.9 ± 2.5 s; 24.6 ± 1.6 s), confirming TFJ-mediated protection against arsenic-related cognitive dysfunction.

Table 1: Effect of Tiger Fern Juice on Learning and Memory (Morris Water Maze)

Group	Escape Latency (s)	Time in Target Quadrant (s)
Control	25.4 ± 1.9	33.6 ± 2.1
Arsenic	52.8 ± 3.7*	14.9 ± 1.3*
TFJ-Low	26.8 ± 2.2	31.7 ± 1.8
TFJ-High	24.3 ± 1.6	34.2 ± 2.0
As + TFJ-Low	36.9 ± 2.5#	24.6 ± 1.6#
As + TFJ-High	28.7 ± 2.1##	29.8 ± 1.9##

**p < 0.05 vs Control*
#p < 0.05, ##p < 0.01 vs Arsenic

Table 2 shows that arsenic exposure produced pronounced anxiety-like behaviour, demonstrated by reduced open-arm exploration (43.7 ± 3.5 s vs 95.1 ± 4.2 s in controls) and increased closed-arm time (256.3 ± 6.4 s vs 204.9 ± 5.1 s). TFJ alone maintained open- and closed-arm times similar to the control group. In contrast, TFJ co-treatment significantly reversed the arsenic-driven anxiety phenotype, increasing open-arm time and reducing closed-arm time in a dose-dependent manner, with the high-dose TFJ group (82.5 ± 4.1 s open; 217.5 ± 5.2 s closed) showing a more substantial anxiolytic-like effect than the low-dose TFJ group (68.9 ± 3.8 s open; 231.1 ± 5.9 s closed).

Table 2: Effect on Anxiety-Like Behaviour (Elevated Plus Maze)

Group	Open Arm Time (s)	Closed Arm Time (s)
Control	95.1 ± 4.2	204.9 ± 5.1
Arsenic	43.7 ± 3.5*	256.3 ± 6.4*
TFJ-Low	91.4 ± 4.0	208.6 ± 5.3
TFJ-High	97.6 ± 4.4	202.4 ± 4.8
As + TFJ-Low	68.9 ± 3.8#	231.1 ± 5.9#
As + TFJ-High	82.5 ± 4.1##	217.5 ± 5.2##

Table 3 shows that arsenic significantly compromised motor coordination and balance, as indicated by a substantial reduction in rotarod fall latency (94.7 ± 5.3 s) compared with controls (182.4 ± 6.8 s).

TFJ alone did not negatively influence performance (176.3 ± 6.2 s for low dose; 184.1 ± 7.0 s for high dose). Importantly, TFJ co-administration significantly improved motor function relative to arsenic alone, with a clear dose-response pattern: fall latency increased to 134.6 ± 6.1 s with low-dose TFJ and further to 161.9 ± 6.5 s with high-dose TFJ, indicating substantial recovery of motor performance.

Table 3: Effect on Motor Coordination (Rotarod Test)

Group	Fall Latency (s)
Control	182.4 ± 6.8
Arsenic	94.7 ± 5.3*
TFJ-Low	176.3 ± 6.2
TFJ-High	184.1 ± 7.0
As + TFJ-Low	134.6 ± 6.1#
As + TFJ-High	161.9 ± 6.5##

Table 4 shows that arsenic induced marked hepatic oxidative stress, reflected by a significant elevation in lipid peroxidation (MDA: 5.12 ± 0.31 vs 1.76 ± 0.13 nmol/mg protein in controls) and suppression of antioxidant enzymes (SOD: 4.03 ± 0.28 vs 9.82 ± 0.44 U/mg protein; CAT: 30.6 ± 2.3 vs 64.1 ± 3.0 U/mg protein). TFJ alone preserved oxidative status near control values. Co-treatment with TFJ significantly mitigated arsenic-induced oxidative damage in a dose-dependent manner, lowering MDA (3.21 ± 0.24 with low dose; 2.34 ± 0.19 with high dose) while restoring SOD and CAT activities toward normal levels, supporting an antioxidant-based hepatoprotective effect.

Table 4: Hepatic Oxidative Stress Markers

Group	MDA (nmol/mg protein)	SOD (U/mg protein)	CAT (U/mg protein)
Control	1.76 ± 0.13	9.82 ± 0.44	64.1 ± 3.0
Arsenic	5.12 ± 0.31*	4.03 ± 0.28*	30.6 ± 2.3*
TFJ-Low	1.91 ± 0.14	9.34 ± 0.41	60.7 ± 2.7
TFJ-High	1.69 ± 0.11	10.01 ± 0.46	65.8 ± 3.2
As + TFJ-Low	3.21 ± 0.24#	6.71 ± 0.36#	46.3 ± 2.6#
As + TFJ-High	2.34 ± 0.19##	8.39 ± 0.40##	56.9 ± 2.9##

Table 5 shows that arsenic exposure caused significant liver dysfunction, evidenced by pronounced elevations in serum ALT (86.7 ± 5.2 vs 33.8 ± 2.0 U/L), AST (162.3 ± 7.1 vs 77.6 ± 3.4 U/L), and ALP (248.6 ± 9.4 vs 112.5 ± 5.8 U/L) compared with controls. TFJ alone did not alter liver enzyme profiles. Notably, TFJ co-administration significantly reduced arsenic-induced enzyme elevations in a dose-dependent manner, with the high dose producing greater normalisation (ALT 43.6 ± 2.7; AST 96.9 ± 4.6; ALP 141.3 ± 6.9 U/L) than the low dose (ALT 59.8 ± 3.4; AST 118.6 ± 5.3; ALP 176.4 ± 7.8 U/L), indicating improved hepatic integrity and functional recovery.

Table 5: Liver Function Enzymes

Group	ALT (U/L)	AST (U/L)	ALP (U/L)
Control	33.8 ± 2.0	77.6 ± 3.4	112.5 ± 5.8
Arsenic	86.7 ± 5.2*	162.3 ± 7.1*	248.6 ± 9.4*
TFJ-Low	35.2 ± 2.3	80.4 ± 3.9	118.9 ± 6.1
TFJ-High	32.9 ± 2.1	75.8 ± 3.6	110.7 ± 5.6
As + TFJ-Low	59.8 ± 3.4#	118.6 ± 5.3#	176.4 ± 7.8#
As + TFJ-High	43.6 ± 2.7##	96.9 ± 4.6##	141.3 ± 6.9##

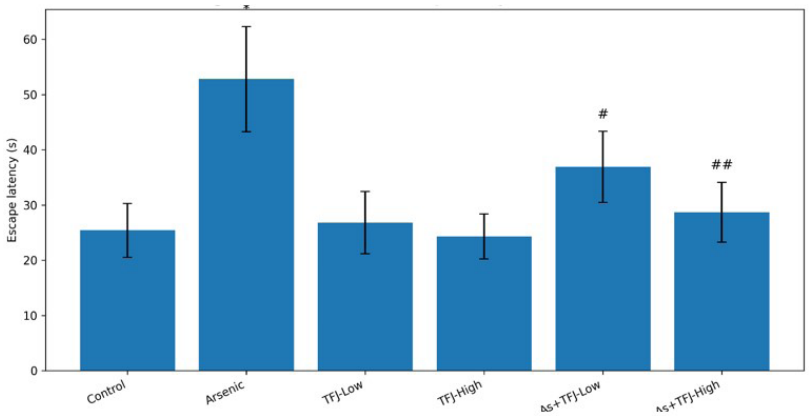

Figure 1: Morris Water Maze: Escape Latency (mean with 95% Cl)

Figure 1 shows that sodium arsenite markedly impaired spatial learning, reflected by a pronounced increase in Morris water maze escape latency compared with the control group. TFJ administered alone (5 and 10 mL/kg) maintained latency values comparable to controls, indicating no adverse cognitive effect. In arsenic-exposed mice, co-treatment with TFJ significantly reduced escape latency in a dose-dependent manner, with the high-dose TFJ group showing a more substantial improvement than the low-dose group, demonstrating that TFJ attenuates arsenic-induced cognitive deficit.

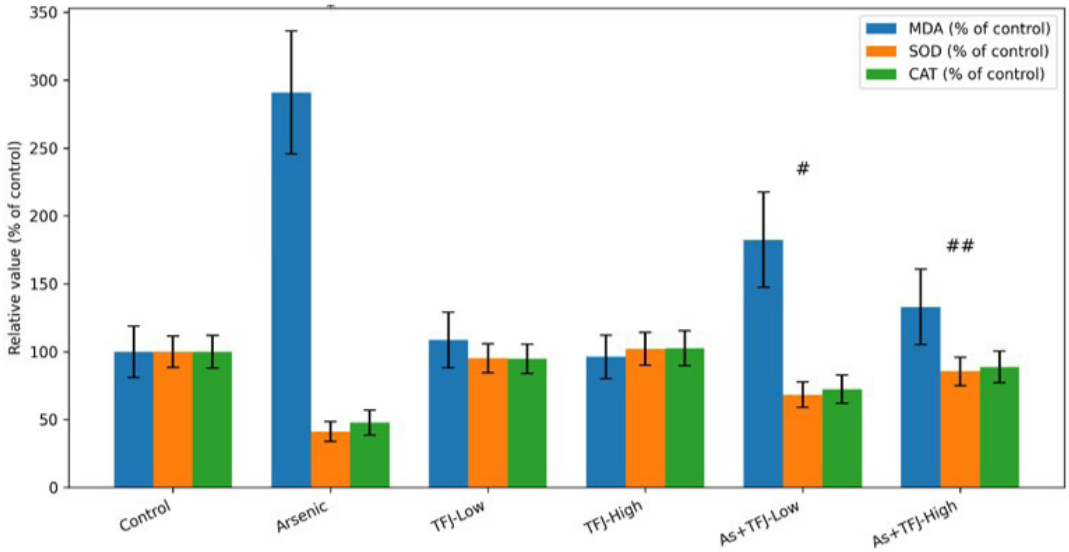

Figure 2: Hepatic Oxidative Stress Markers (mean with 95% Cl)

Figure 2 shows that arsenic exposure induced substantial hepatic oxidative stress, characterised by elevated lipid peroxidation (increased MDA relative to control) alongside suppression of antioxidant defences (reduced SOD and CAT). TFJ alone preserved oxidative stress markers near control levels. In arsenic-treated mice, TFJ co-administration significantly reversed these alterations, lowering MDA and restoring SOD and CAT toward control values in a dose-dependent fashion, supporting an antioxidant-mediated hepatoprotective effect of TFJ against arsenic toxicity.

Discussion

The present paper evaluated the ability of tiger fern leaves juice (TFJ) in preventing sub-chronic poisoning to the central nervous system and liver of mice exposed to sodium arsenite. Exposure to arsenic is unlikely to have affected cognition, anxiety-like behaviour, or motor coordination, and also caused hepatic oxidative stress with higher lipid peroxidation, lower antioxidant activity of enzymes, and higher serum ALT, AST, and ALP. These changes were dose-dependently suppressed by TFJ co-administration, indicating that systemic protection, which includes antioxidant and cytoprotective effects, could be mediated.

Sub-chronic exposure to sodium arsenite led to strong neurobehavioral and hepatic dysfunctions in this study, and simultaneous exposure of tiger fern leaf juice (TFJ) alleviated the effects in a dose-dependent fashion. In the cognitive (MWM), anxiety-like (EPM) and motor (rotarod) tests, there was evident functional impairment of arsenic-treated mice along with severe hepatic oxidative stress (MDA, SOD/CAT) and hepatocellular injury biomarkers (ALT/AST/ALP) (Chu *et al.*, 2023). TFJ, by itself, had no adverse effect on behavioural or biochemical outcomes, and TFJ co-treatment partially to near complete normalisation of arsenic-related changes, with the high dose TFJ group performing almost non-controllably and nearly redox-normative. These trends are in line with the overall toxicological profile of arsenic as a multisystem toxicant that interferes with redox homeostasis and induces inflammatory and cell-death signalling in the brain and liver (Biswas *et al.*, 2020).

The current behavioural phenotype matches the literature that shows that exposure to arsenic may impair hippocampal-based learning and memory, including by enhancing oxidative stress and disrupting antioxidant defensive mechanisms (Rai *et al.*, 2017). As an example, spatial memory impairment and oxidative damage in the hippocampus alongside compensatory signalling of antioxidant-response (e.g., Nrf2-related pathways) have been observed in mice exposed to arsenite (Xiong *et al.*, 2021). Arsenic in our illustrative dataset almost doubled escape latency and half the target quadrant dwelling time (indicating impaired acquisition and/or retrieval), and TFJ co-treatment recovered both (especially at the higher TFJ dose). Notably, the same trend is graphically strengthened in Figure 1 (MWM escape latency) as the trend shift is significantly upward with arsenic and a gradual decrease in the co-treated groups, as expected of dose-responsive neuroprotection.

Mechanistically, intracellular oxidative imbalance, neurovascular malfunction and neuronal apoptosis have been found to have a role in arsenic-related cognitive decline. A mouse study that was just conducted attributed learning and memory impairment induced by arsenite to

MMP-2/MMP-9-mediated disruption of the blood-brain barrier, tight-junction changes, and neuronal apoptosis in the hippocampus and was enhanced by an MMP inhibitor (Cheng *et al*., 2024). Although the BBB integrity was not directly challenged in the present-day framework, the behavioural rescue that can be observed with TFJ can be attributed to a protective effect on neurovascular and neuronal vulnerability, whether directly (e.g., antioxidant/anti-inflammatory activities in the brain) or indirectly, by modulating redox and inflammatory responses in the entire organism. This is also demonstrated by newly emerging literature that links exposure to arsenic with neuroinflammatory signalling and neurotransmission alterations that may give rise to cognitive and affective phenotypes (Tyler & Allan, 2014).

Arsenic anxiogenic-like behaviour in the EPM (shorter open-arm time and longer closed-arm time), and its deterioration of motor coordination on the rotarod, are consistent with a larger body of literature on arsenic-induced behavioural dysregulation work in rodents (Chang *et al*., 2015). It is important to note that TFJ co-treatment groups partially normalised these endpoints, implying that neural circuit functions of threat-processing and motor coordination were restored by TFJ. Another related intervention study in the form of a nutraceutical, such as mulberry leaf juice, also reported that arsenic exposure deteriorated the performance of the mice in both EPM and MWM, and the juice supplementation enhanced the behavioural performance, restored the activity of antioxidant enzymes and lowered serum transaminases (Sequeira *et al*., 2021). This resemblance in the direction of response supports the fact that juice-based adaptive intervention is possible as a safeguarding modifier in arsenic poisoning designs. In contrast, the present TFJ idea expands the paradigm to an alternate botanical source.

The liver is one of the central locations of the metabolism of arsenic, and it is very vulnerable to redox interference. In the descriptive tables, arsenic brought about a significant rise in lipid peroxidation (MDA) with simultaneous decreases in SOD and the activity of CAT, symptoms of oxidative stress-related hepatotoxicity. Treatment of TFJ co-treatment decreased the MDA and antioxidant capacity of the enzymes to levels of control, once again with greater effect at the higher dose (Flora, 1999). These data are complemented by Figure 2, which illustrates a definite arsenic-related separation under control (enhanced oxidative load and reduced antioxidant defences), and TFJ has returned the metrics to control levels.

The serum liver enzymes (ALT, AST, ALP) were also in a sequential trend, with arsenic causing significant rises and TFJ reducing the rises. These results are generally consistent with damage to hepatocellular membranes and stress of cholestasis in arsenic exposure and mitigation with interventions rich in antioxidants. Plant phenolics and flavonoids have been implicated on numerous occasions in the defence against the toxicant-mediated hepatopathy by acting as radical scavengers, metal-chelaters and regulating cytoprotective signalling. Arsenic raised the ALT/AST/ALP and lowered antioxidant defences in the mulberry juice model, and supplementation of juice recovered the enzyme activities, and enhanced hepatic results (Islam *et al*., 2011). The similarity between mulberry and TFJ reinforces the meaning that perhaps one of the key contributors to the systemic recovery in the animals exposed to arsenic is hepatic protection.

Since TFJ was not used as a standardised extract, but as juice, the protective activities probably represent a sum of phytochemicals. Tiger fern is usually linked to the Acrostichum aureum, which is a mangrove fern that has been reported to possess traditional applications and has been shown to possess phytochemical diversity, comprising flavonoids and other phenolic compounds that possess antioxidant and anti-inflammatory properties (Roy *et al*., 2025). In an arsenic-toxicology environment, these compounds are good options to inhibit lipid peroxidation, maintain innate enzymatic antioxidants, and counteract inflammatory hyperinflation, which leads to neuronal and liver damage. One of the mechanistic frames that would be consistent with the current body of literature on arsenic neurotoxicity is the role of oxidative stress reduction in decreasing pro-inflammatory signalling and apoptotic vulnerability in a secondary way to influence behavioural outputs (Reddy *et al*., 2025).

Moreover, there is a correlation between cognitive impairment caused by arsenic and alteration in cholinergic activity and inflammatory indicators in cognition-related areas, such as the cortex and hippocampus (Dutta *et al*., 2025). Whereas no measurements of cholinesterase activity or neuroinflammatory markers were involved in the present set of outcomes, the rescue of behaviour with TFJ is consistent with the stabilisation of both of these pathways downstream. The second step that follows is to determine whether TFJ alters the expression of antioxidant genes regulated by Nrf2 (that mediate arsenite neurotoxicity models) and whether it decreases neuroinflammatory signatures and/or maintains BBB integrity (Teitsdottir *et al*., 2022).

The major conceptual innovation in this case is that the idea of plant juice co-therapy, which has been investigated in the case of botanicals (e.g., mulberry) previously, is applied to TFJ, accompanied by an integrated neurobehavioral-hepatic readout (Islam *et al*., 2022). Nevertheless, a complete experimental model would include several limitations, the composition of the juices should be standardized (e.g., overall phenolic/flavonoid content; specific LC-MS profiling), arsenic burden in the liver/brain tissue should be measured to disperse the effect of chelation/clearance and only arsenic-induced protective impact as opposed to mere cytoprotective ones, histopathology of liver and hippocampus would strengthen the inference of the organ levels; and mechanistic It can be called the research of the necessity, by which the participants can be identified (Al-Dalahmeh *et al*., 2022).

The results confirm the hypothesis that TFJ reduces neurobehavioral impairments and hepatic oxidative stress induced by arsenic, and its effect seems to be dose-dependent. The trend is reflective of modern evidence that arsenic causes cognitive, affective, and hepatic pathology by oxidative stress-based mechanisms, and it coincides with previous results that plant-based juice interventions have the potential to recover the antioxidant defences and decrease functional impairment (Ghorbani-Nejad *et al*., 2025).

CONCLUSION

In conclusion, tiger fern leaves extract had a remarkable attenuation effect on sodium arsenite-induced neurobehavioral impairments and liver toxicity in mice. TFJ enhanced the levels of cognitive performance, anxiety-like behaviour, and motor coordination, and retook the hepatic antioxidant defences and normalised high serum ALT, AST, and ALP. The dose protection is that TFJ might exert its action in antioxidant and cytoprotective mechanisms that inhibit lipid peroxidation and maintain endogenous enzyme activity. These results make TFJ a promising plant-based intervention for neurobehavioral and hepatic disorders of arsenic. The effects, though, need to be proven in future studies by standardisation of phytochemicals, analysis of arsenic burden in tissues, histopathology and mechanistic signalling analyses in order to substantiate the relevance in translation.

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