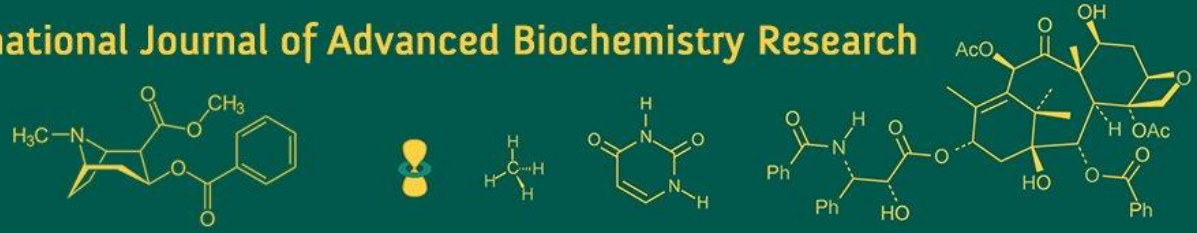
International Journal of Advanced Biochemistry Research



ISSN Print: 2617-4693
ISSN Online: 2617-4707
NAAS Rating (2025): 5.29
IJABR 2025; 9(12): 637-645
www.biochemjournal.com
Received: 10-11-2025
Accepted: 13-12-2025

Ramya DH
Masters Scholar, Department of Veterinary Pathology, Veterinary College, Hassan, Karnataka, India

Dr. Anjan Kumar KR
Associate professor, Department of Veterinary Pathology, Veterinary College, Hebbal, Bengaluru, Karnataka, India

Dr. Girish BC
Associate professor, Department of Veterinary Pathology, Veterinary College, Hassan, Karnataka, India

Dr. Shilpa V T
Assistant professor, Department of Veterinary Pathology, Veterinary College, Hassan, Karnataka, India

Dr. Ravikumar C
Associate professor and Head, Department of Veterinary Pharmacology and Toxicology, Veterinary College, Hassan, Karnataka, India

Dr. Sanganagouda Koppad
Associate professor, Department of Veterinary Microbiology, Veterinary College, Gadag, Karnataka, India

Ramya DH
Masters Scholar, Department of Veterinary Pathology, Veterinary College, Hassan, Karnataka, India

Golla Harsha
Masters in Veterinary Pathology Veterinary College Hassan, Karnataka, India

Corresponding Author:
Ramya DH
Masters Scholar, Department of Veterinary Pathology, Veterinary College, Hassan, Karnataka, India

Protective efficacy of resveratrol against arsenic induced sub-chronic toxicity: A pathomorphological study in wistar rats

Ramya DH, Anjan Kumar KR, Girish BC, Shilpa VT, Ravikumar C, Sanganagouda Koppad and Golla Harsha

DOI: https://www.doi.org/10.33545/26174693.2025.v9.i12h.6600

Abstract
The present investigation was undertaken to evaluate the pathomorphological alterations associated with arsenic-induced sub-chronic toxicity in Wistar rats and its amelioration with resveratrol. Thirty-six adult male Wistar rats were randomly divided into six groups (n=6). Group I served as normal control, Group II as disease control ($NaAsO_2$ @ 10 mg/kg b.wt. orally for 60 days), Group III received treatment same as Group II along with telmisartan (10 mg/kg b.wt. orally for 60 days) as a reference control group and Groups IV-VI received treatment same as Group II along with resveratrol at graded doses (4, 8, and 16 mg/kg b.wt. orally for 60 days). The disease control group exhibited a significant reduction (p<0.05) in body weight, hematological parameters, and alterations in biochemical indices including increased ALT, ALP, BUN, and creatinine with decreased total protein levels. Antioxidant assays revealed significant elevation in lipid peroxidation and depletion of SOD and catalase activities in the disease control group. Treatment with resveratrol and telmisartan ameliorated these changes in a dose-dependent manner. Histopathological examination of the aorta revealed lesions consistent with oxidative stress and inflammation. Special staining of the aorta further demonstrated vascular alterations, which were markedly reduced in the resveratrol (16 mg/kg) and telmisartan-treated groups. The study concludes that resveratrol exhibits potent antioxidant and cytoprotective efficacy against sub-chronic arsenic-induced vascular and systemic toxicity, comparable to telmisartan.

Keywords: Arsenic-induced toxicity, Resveratrol, Oxidative stress, Vascular pathology, Wistar rats

Introduction
Metals are natural constituents of the Earth's crust and enter the environment through erosion and weathering. However, anthropogenic activities such as mining, electroplating, tanning, and chemical manufacturing have greatly increased the release of heavy metals, resulting in serious ecological and health hazards (Jaishankar *et al.*, 2014; Yadav *et al.*, 2017) [15, 51]. Among these, arsenic is one of the most widespread and toxic environmental contaminants. It occurs naturally in both organic and inorganic forms, with the inorganic species arsenite (As^{3+}) and arsenate (As^{5+}) being highly toxic due to their reactivity with thiol groups in biological systems (Souza *et al.*, 2020) [40].

Chronic arsenic exposure through contaminated water is a global public health concern affecting over 200 million people across regions including India, Bangladesh, and China (Turk *et al.*, 2019) [42]. The World Health Organization (WHO, 2011) and US-EPA (2001) recommend a maximum limit of 0.01 ppm arsenic in drinking water, while higher concentrations up.

to 14.2 ppm have been reported in parts of India (Mazumder and Dasgupta, 2011). Prolonged exposure leads to multisystem toxicity, affecting hepatic, renal, cardiovascular, reproductive, and nervous systems, and is linked to carcinogenesis (Bhowmick *et al.*, 2018) [6].

Experimental evidence indicates that arsenic induces oxidative stress, inflammation, and vascular dysfunction, contributing to cardiovascular diseases and hypertension (Waghe *et al.*, 2015; Khatun *et al.*, 2024) [17, 47].

Maintaining vascular integrity is essential for cardiovascular health, and disruption of endothelial homeostasis leads to pathological consequences

Recently, natural phytochemicals have gained significant attention as safer therapeutic alternatives to synthetic drugs. Resveratrol (trans-3,4,5-trihydroxystilbene), a polyphenolic compound found in grapes, peanuts, and red wine, exhibits potent antioxidant, anti-inflammatory, and cytoprotective properties (Rizvi *et al.*, 2010) [31]. It scavenges reactive oxygen species and modulates redox-sensitive signaling pathways, thereby reducing oxidative DNA damage and lipid peroxidation (Mognetti *et al.*, 2024) [22].

Materials and methods

Chemicals

Sodium meta arsenite A.R was obtained from Jai Maruthi Scientific Bangalore, manufactured by M/s NICE Chemicals Private Limited, Kochi. and Resveratrol (M. W: 228.24 g/mol and CAS number 501-36-0) was procured from M/s Dhamtech pharmaceutical limited, Navi Mumbai. Telmisartan (M. W: 514.617 g/mol and CAS number 144701-48-4) was used as standard drug for reference control. It was procured from Intas pharmaceutical limited, Ahmedabad, India.

Experimental Animals

Male Wistar rats (N = 36) of 6-8 weeks of age and weighing ~ 150-200 g was obtained from an authorized vendor (Chromed Biosciences Private limited Labs Plot No.C- 38, KIADB, Industrial area, Hirehalli, Mydala, Tumkur, Karnataka-572168 (Reg. No. 2171/PO/RcBiBt/S/22/CCSEA). They were housed in Small Animal House facility, Veterinary College Hassan, in polypropylene cages and maintained under standard management practices including light: dark cycle of 12 h each. Experimental rats were fed with standard rodent chow (Amrut®, Ms. Pranav Agro Industries Ltd, M.H, India, supplied by a local vendor) and given free access to water ad libitum. Prior approval of the Institutional Animal Ethics Committee (IAEC) was obtained (Approval No: HVC/IAEC/12/2024, dated 13-05-2025) to carry out the current investigation as per the guidelines of the Committee for the Control and Supervision of Experiments on Animals (CCSEA) New Delhi.

Experimental Design

After 1 week of acclimatization, the experimental rats were randomly divided into six groups as detailed below: group-I (n = 6): Served as control group and received 0.5 per cent (W/V) aqueous methylcellulose containing 0.2 per cent (W/V) tween 80. Group-II (n = 6): received sodium (meta) arsenite (Na_2AsO_3) @ 10 mg/kg for a period of 60 days. Group-III (n = 6) received telmisartan (1% aqueous solution made by using Tween- 80 as a vehicle) @ 10 mg/kg; per os) daily 1 h after the administration of arsenic as a reference control along with Na_2AsO_3) @ 10 mg/kg orally for 60 days. Groups IV, V and VI were treated with Resveratrol 1 h before the administration of sodium meta arsenite, at doses of 4 mg/kg, 8 mg/kg and 16 mg/kg body weight, respectively, orally for 60 days with 0.5 per cent (W/V) aqueous methylcellulose as a vehicle.

General Observation and Body Weight

All the animals were observed for general clinical signs if any, during the course of the study period twice daily. The animals were weighed individually on a digital scale for every seven days to adjust the dosage of compounds during the experiment and to evaluate the effect of treatments on their body weights till the end of the experiment.

After end of experiment overnight fasting was done (but had free access to drinking water ad libtum), and they were sacrificed [i.e., day 61 (morning hours)] under anesthesia overdose (Ketamine hydrochloride @ 40 mg/kg; i.p and Xylazine hydrochloride @ 10 mg/kg; i.p) and subjected to further studies.

Hematobiochemical analysis

On the 61st day of the experiment, blood samples were collected for hematological and biochemical analyses. Hematological parameters including Hb, TEC, TLC, PCV, MCV, MCH, and MCHC were estimated using a Mindray BC-2800VET auto hematology analyzer at the Department of Teaching Veterinary Clinical Complex, Veterinary College, Hassan. For biochemical analysis, blood collected in serum vacutainers was centrifuged at 3000 rpm for 15 minutes, and the separated serum stored at −20°C was analyzed for ALT, ALP, total protein, BUN, and creatinine using an Artos Elita semi-automatic biochemistry analyzer with Swemed Biomedicals Pvt. Ltd. kits.

Preparation of Aorta Tissue Homogenate

The abdominal aorta was rapidly excised, rinsed in phosphate buffer, blotted dry, and stored at −80°C for antioxidant enzyme analysis. For tissue homogenate preparation, 500 mg of the abdominal aorta from each rat was minced, transferred to a 10 ml Borosil® glass homogenizer, and homogenized with 5 ml of ice-cold 50 mM potassium phosphate buffer (pH 7.4) at 3-5°C (Camp *et al.*, 2003). The homogenate was centrifuged at 10,000 × g for 10 minutes, and the supernatant was stored at −80°C for estimation of superoxide dismutase, catalase, and thiobarbituric acid reactive substances (MDA concentration).

Determination of Superoxide Dismutase (SOD) Activity

The SOD activity in the aorta was determined by the procedure of Madesh and Balasubramanian [20]. Briefly, the reaction mixture contained 0.65 ml of PBS (pH 7.4), 30 µl of 3-(4-5 dimethyl thiazol 2-yl) 2,5-diphenyl tetrazolium bromide (1.25 mM), 75 µl of pyrogallol (100 µM) and 10 µl supernatant of aorta homogenate (10%). The mixture was incubated at room temperature for 5 min. and the reaction was stopped by adding 0.75 ml of dimethyl sulfoxide. The absorbance was read at 570 nm, and the activity was expressed as unit/mg protein.

Determination of Catalase Activity

The CAT activity in the aorta was assayed by the spectrophotometric method of Aebi [2]. In brief, 1.99 ml of phosphate buffer (50 mM, pH 7.0) and 10 µl supernatant of homogenate (10%) were taken in a cuvette. The reaction was started by adding 1 ml of H_2O_2 (10 mM), and the absorbance was recorded at every 30 s for 3 min at 240 nm against a water blank. The activity was expressed as mmol H_2O_2 utilized/min/mg protein.

Assessment of Peroxidative Damage

Peroxidative damage of the aorta was assessed by evaluating lipid peroxidation (LPO) in terms of malondialdehyde (MDA) production as described by Paula *et al.,* [29] In brief, 1 ml of the aorta homogenate was mixed with 1 ml of 2% thiobarbituric acid, 1 ml of 25% HCl and 90 µl butylated hydroxytoluene. The mixture was kept in a boiling water bath for 10 min at 95 °C and then centrifuged. The supernatant was transferred to a quartz cuvette, and the absorbance was read at 535 nm. Results have been expressed as nmol MDA formed/g of aorta.

Gross and histopathology

A detailed post-mortem examination was conducted on all the sacrificed rats in all the experimental groups. The gross lesions were recorded and representative tissue samples of abdominal aorta was collected and preserved in 10% neutral buffered formalin for histopathological studies. Fixed tissues were processed by routine paraffin embedding technique. Sections of 5-6 (µ) thickness was cut and stained with routine Hematoxylin and Eosin method (H&E). Similarly, the standard procedure of Masson's trichrome staining was employed to stain histopathological sections of the aorta.

Statistical Analysis

The values obtained from the various experiments were expressed as mean±S.E with 'n' equal to number of animals or samples. Data obtained were statistically subjected to one-way analysis of variance (ANOVA) followed by Duncan's post hoc multiple comparison test using SPSS statistics software (IBM® SPSS® statistics software, Version 21.0.0, 2012, Armonk, NY, USA). The difference was considered significant at $p<0.05$ or lower. Graphical presentation of the data was carried out by using GraphPad Prism software programme (GraphPad® software Inc., Version 8.4.3; San Diego, CA, USA).

Results

The present study was undertaken to investigate the ameliorative effect of resveratrol against arsenic induced sub-chronic toxicity in Wistar rats.

Sub-chronic exposure to 'As' did not show any acute signs of illness or mortality in any of the experimental groups. However, upon close clinical examination, normal control rats (Group I) remained healthy and active throughout the experimental period. Disease control rats (Group II) exhibited a minor degree of weakness, reduced feed intake, ruffled hair and dehydration after two weeks of arsenic exposure. Rats in Groups III to VI showed similar signs but with markedly reduced intensity and frequency.

Body weight

Throughout the experimental period, Group I (normal control) rats showed a steady and consistent increase in body weight. In contrast, Group II (disease control) rats exhibited a marked reduction in body weight gain from the 3rd week onward following arsenic administration, showing a significant difference ($p<0.05$) compared to Group I. Rats in Group III (reference control) showed gradual improvement in body weight from the 3rd week, indicating partial recovery. Among the resveratrol-treated groups (Groups IV-VI), a dose-dependent improvement in body weight was observed, with Group VI showing body weight changes comparable to normal controls and Group V approaching normal values from the 4th week onward presented in Fig 1:. These findings indicate that resveratrol supplementation effectively mitigated arsenic-induced weight loss.

Haematology

On the 61st day, hematological parameters showed significant alterations among the experimental groups (Fig 2:). The mean Hb level in Group I was 14.63±0.41 g/dl, while Group II exhibited a significant ($p<0.05$) decrease to 10.36±0.45 g/dl. Treatment groups III, IV, V, and VI showed improved Hb values of 12.64±0.47, 11.74±0.32, 12.99±0.45, and 14.09±0.30 g/dl, respectively, with Group VI comparable to controls. TEC decreased significantly ($p<0.05$) in Group II (6.12±0.29 × 10^6/µl) compared to Group I (7.33±0.16 × 10^6/µl), while Groups III-VI showed restoration (7.02±0.10 to 7.24±0.13 × 10^6/µl). The total leucocyte count was markedly reduced in Group II (5.30±0.38 × 10^3/µl) relative to Group I (8.35±0.08 × 10^3/µl), whereas treatment groups (7.01±0.32 to 7.85±0.31 × 10^3/µl) showed significant recovery. Similarly, PCV declined in Group II (30.38±0.74 %) compared to Group I (41.88±0.71 %), while Groups III-VI (36.31±0.90 % to 40.66±1.33 %) exhibited significant improvement. MCV and MCH were significantly elevated in Group II (63.75±1.64 fL and 25.13±1.44 pg, respectively) compared to controls (51.51±1.36 fL and 17.01±0.53 pg), but treatment with resveratrol and telmisartan, normalized these values (MCV: 51.76±0.75-54.54±0.89 fL; MCH: 18.67±0.69-23.12±0.82 pg). The MCHC decreased significantly ($p<0.05$) in Group II (26.20±0.88 %) compared to Group I (32.76±0.83 %), whereas Groups III-VI (30.16±0.52 % to 33.06±0.66 %) demonstrated marked restoration, with Group VI showing near-normal values. Overall, arsenic exposure induced anaemia and haematological disturbances, which were effectively ameliorated by resveratrol.

Biochemistry

Arsenic treated (Group II) rats showed a significant ($p<0.05$) rise in serum ALP (131.04±2.49 IU/L), ALT (71.22±1.16 IU/L), creatinine (0.84±0.04 mg/dl), and BUN (25.76±1.37 mg/dl), along with a marked decrease in total protein (5.71±0.25 g/dl), compared to Group I rats, which recorded 107.54±1.08 IU/L, 41.88±0.42 IU/L, 0.55±0.01 mg/dl, 16.37±0.99 mg/dl, and 8.58±0.27 g/dl, respectively. The telmisartan-treated group (Group III) showed marked improvement with ALP 109.93±1.86 IU/L, ALT 48.56±1.51 IU/L, total protein 7.44±0.19 g/dl, creatinine 0.67±0.02 mg/dl, and BUN 20.53±0.72 mg/dl, all significantly ($p<0.05$) better than Group II. The resveratrol-treated groups (Groups IV-VI) also exhibited dose-dependent amelioration, with ALP values of 124.03±1.12, 110.99±0.97, and 108.08±1.16 IU/L; ALT values of 56.85±1.62, 48.43±1.52, and 45.43±1.19 IU/L; total protein levels of 6.74±0.10, 7.35±0.18, and 8.05±0.07 g/dl; creatinine levels of 0.80±0.03, 0.68±0.06, and 0.63±0.06 mg/dl; and BUN levels of 23.32±0.21, 20.07±0.49, and 18.37±0.50 mg/dl, respectively. All treatment groups demonstrated significant ($p<0.05$) restoration of hepatic and renal biochemical parameters compared to Group II, with Group VI (high-dose resveratrol) showing values most comparable to the normal control group (Fig 3:).

Oxidative stress biomarkers

On the 61st day of the experiment (Fig 4:), oxidative stress biomarkers showed marked alterations in the disease control group (Group II) compared to the normal control (Group I).

Lipid peroxidation (LPO):

The mean aortic LPO level was significantly (p<0.05) elevated in Group II (27.90±0.61 nmol MDA/g tissue) compared to Group I (7.07±0.11). The telmisartan-treated group (Group III) and resveratrol-treated groups (Groups IV-VI) showed a considerable reduction in LPO levels to 7.59±0.35, 15.86±0.34, 10.91±0.38, and 7.10±0.30, respectively. Among these, Groups III and VI exhibited values comparable to the normal control, indicating effective protection against lipid peroxidation.

Catalase (CAT)

A significant (p<0.05) depletion in aortic catalase activity was observed in Group II (31.52±0.48 mmol H_2O_2 utilized/min/mg protein) relative to Group I (47.07±0.11). The telmisartan-treated (Group III) and resveratrol-treated (Groups IV-VI) rats exhibited increased CAT activities of 42.59±0.52, 41.50±0.84, 43.43±0.93, and 46.83±1.26, respectively. All treatment groups showed significant improvement over Group II, with Group VI nearing normal values.

Superoxide dismutase (SOD)

The mean aortic SOD level was significantly (p<0.05) reduced in Group II (6.02±0.28 U/mg protein) compared to Group I (7.90±0.31). Treatment with telmisartan (7.63±0.36) and resveratrol at different doses (7.10±0.25, 7.64±0.17, and 7.96±0.23 for Groups IV-VI, respectively) led to a significant (p<0.05) increase in SOD activity compared to Group II, with Groups III and VI showing values nearly identical to normal controls.

Histological evaluation

Histopathological analysis of the abdominal aorta showed intact vascular layers with normal architecture in Group I. Arsenic treated group (Group II) exhibited severe damage, including disrupted tunica adventitia, thickened tunica media, vacuolated intima, oedema, disorganized elastic laminae, and collagen deposition. Telmisartan treatment (Group III) improved vascular integrity with reduced medial thickening and restored elastic structure (Plates 17-19). Resveratrol-treated groups (IV-VI) showed dose-dependent recovery with progressive normalization of smooth muscle and elastic fibers and reduced collagen content, with Group VI displaying near-normal architecture.

Discussion

In the present study sub-chronic exposure to sodium (meta) arsenite ('As') (group-II) resulted in a significant (p<0.05) decrease in body weight (b.wt.) compared to other groups, aligning with previous reports (Hou *et al.*, 2007; Alenzi *et al.*, 2010; Yang *et al.*, 2019; Zhang *et al.*, 2021) [3, 14, 50, 52]. The reduction was likely due to arsenic-induced anorexia, decreased feed intake, and disrupted glucose metabolism (Garcia *et al.*, 2006; Paul *et al.*, 2007; Lu *et al.*, 2011) [19, 26]. Telmisartan-treated rats (Group III) showed marked improvement, attributed to its antioxidant and anti-inflammatory actions, suppression of lipid peroxidation, and activation of PPAR-γ, which enhances glucose utilization and metabolic balance (Fouad *et al.*, 2012; He *et al.*, 2010; Sharma *et al.*, 2021) [10]. Among resveratrol-treated groups, Group VI showed near-normal weight, indicating dose-dependent protection through enhanced glucose utilization, lipid metabolism, and antioxidant defense (Ray *et al.*, 1999; Oak *et al.*, 2005; Baur & Sinclair, 2006) [4, 24, 30].

In the present study, normal control rats (Group I) maintained stable hematological parameters, confirming physiological homeostasis, whereas arsenic-exposed rats (Group II) showed a marked decline in TEC, Hb, PCV, TLC, MCH, and MCHC with an increase in MCV, indicating macrocytic anemia caused by arsenic-induced oxidative stress, disruption of erythropoiesis, inhibition of heme synthesis, and interference with folate and vitamin B_{12} metabolism (Flora *et al.*, 2005; Acharyya *et al.*, 2015) [1]. Telmisartan-treated rats (Group III) exhibited significant improvement due to its antioxidant, anti-inflammatory, and PPAR-γ agonistic effects, which enhanced bone marrow microenvironment, promoted erythropoiesis, reduced lipid peroxidation, and restored erythrocyte membrane stability (Schupp & Janke, 2006; Sharma *et al.*, 2016). Resveratrol-treated groups (IV-VI) showed dose-dependent normalization of hematological parameters by activating the Nrf2-ARE and SIRT1-PGC-1α pathways, which increased antioxidant enzyme activity, reduced oxidative stress and inflammation, improved mitochondrial energy metabolism, and protected bone marrow cells from apoptosis, resulting in restoration of red cell production and function, with Group VI values comparable to the control group (Pandey *et al.*, 2010; Wang *et al.*, 2017) [25, 48].

In the present study, normal control rats (Group I) showed stable serum biochemical parameters, whereas arsenic-exposed rats (Group II) exhibited a significant rise in ALP, ALT, BUN, and creatinine with a decrease in total protein, indicating hepatic and renal injury. These alterations were mainly due to arsenic-induced oxidative stress, lipid peroxidation, mitochondrial damage, and depletion of antioxidants (SOD, CAT), leading to hepatocellular membrane leakage and impaired protein synthesis (Pi *et al.*, 2003; Valko *et al.*, 2005; Nandi *et al.*, 2006) [27, 45]. Accumulation of methylated arsenic metabolites (MMA, DMA) in renal tubules further aggravated nephrotoxicity and elevated serum urea and creatinine (Vahter, 2002; Tseng, 2009) [41, 44]. Telmisartan-treated rats (Group III) showed significant restoration of biochemical values through its dual action as an AT_1 receptor blocker and partial PPAR-γ agonist, reducing oxidative stress and inflammation, stabilizing hepatocyte membranes, and improving antioxidant enzyme activity (Schupp *et al.*, 2004; Sharma *et al.*, 2010) [35, 38]. Resveratrol-treated groups (IV-VI), particularly Group VI, displayed values comparable to normal controls due to its Nrf2-mediated antioxidant activation, which enhanced SOD, CAT, and GPx, reduced lipid peroxidation, and prevented renal tubular degeneration and hepatic enzyme leakage (Robb *et al.*, 2008; Xia *et al.*, 2017; Sonmez *et al.*, 2016) [32, 39, 49].

Arsenic-exposed rats (Group II) displayed a sharp rise in LPO and a fall in SOD and CAT, indicating severe oxidative stress from excess ROS generation, mitochondrial dysfunction, and inhibition of the Nrf2-ARE antioxidant pathway (Pi *et al.*, 2003; Valko *et al.*, 2005) [27, 45]. Telmisartan-treated rats (Group III) exhibited restored antioxidant enzyme activity and reduced MDA levels due to AT_1 receptor blockade and PPAR-γ activation, which

improved redox balance and antioxidant gene expression (Benson *et al.*, 2004; Schupp *et al.*, 2004) [5, 35]. Resveratrol-treated groups (IV-VI), especially Group VI, showed near-normal enzyme levels by activating Nrf2-ARE and PGC-1α pathways, enhancing mitochondrial stability, and suppressing inflammatory mediators like NF-κB and AP-1 (Robb *et al.*, 2008; Park *et al.*, 2012) [28, 32]. Both telmisartan and resveratrol effectively mitigated arsenic-induced oxidative stress by restoring antioxidant defenses and preventing cellular damage.

Histopathological examination of the Group I (normal control) aorta showed intact vascular layers and normal architecture with orderly elastic laminae. In contrast, Group II (arsenic-exposed) rats exhibited vascular degeneration characterized by disrupted tunica adventitia, thickened tunica media, vacuolation, edema, and irregular elastic laminae with collagen deposition, consistent with previous findings (Saad *et al.*, 2006; Sinha *et al.*, 2008; *et al.*, 2022) [33, 34]. These alterations arise from arsenic-induced oxidative stress, where excessive ROS generated via NADPH oxidase and mitochondrial dysfunction promotes lipid peroxidation, smooth muscle proliferation, and endothelial injury (Jalaludeen *et al.*, 2015; Nirwane *et al.*, 2015; Cheng *et al.*, 2022) [7, 16, 23]. However, resveratrol-treated groups showed restoration of vascular integrity with normal tunica layers, reduced collagen deposition, and preserved elastic laminae, suggesting its protective effect through antioxidant, anti-inflammatory, and endothelial-stabilizing actions (Das and Das, 2007; Robb *et al.*, 2008; Ungvari *et al.*, 2010; Li *et al.*, 2019) [18, 32, 43].

Fig 1: Graphical representation of mean body weight (g) values of rats of different groups at weekly intervals

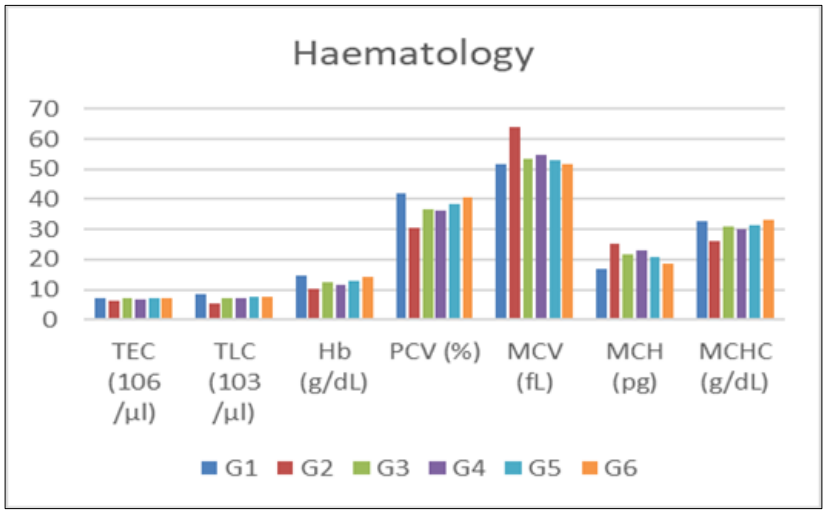


Fig 2: Graphical representation of hematological parameters values of rats of different groups (Mean±SE) on final day (61st day) of the study.

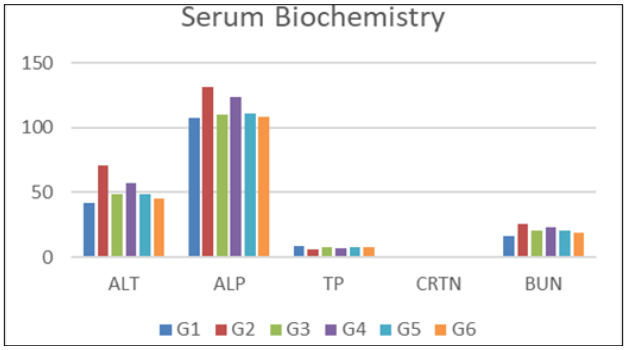


Fig 3: Graphical representation of serum biochemical parameters values of rats of different groups (Mean±SE) on final day (61st day) of the study.

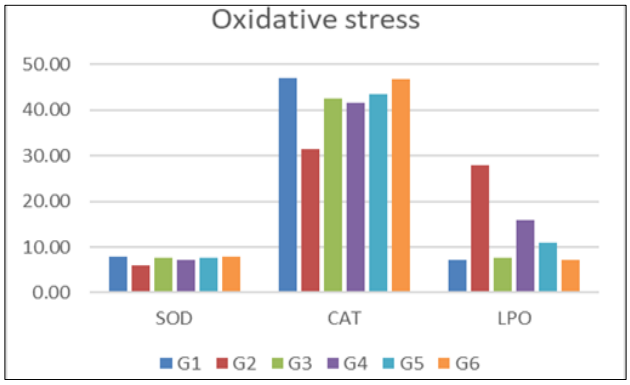


Fig 4: Graphical representation of aortic tissue oxidative stress parameters values of rats of different groups (Mean±SE) on final day (61st day) of the study.

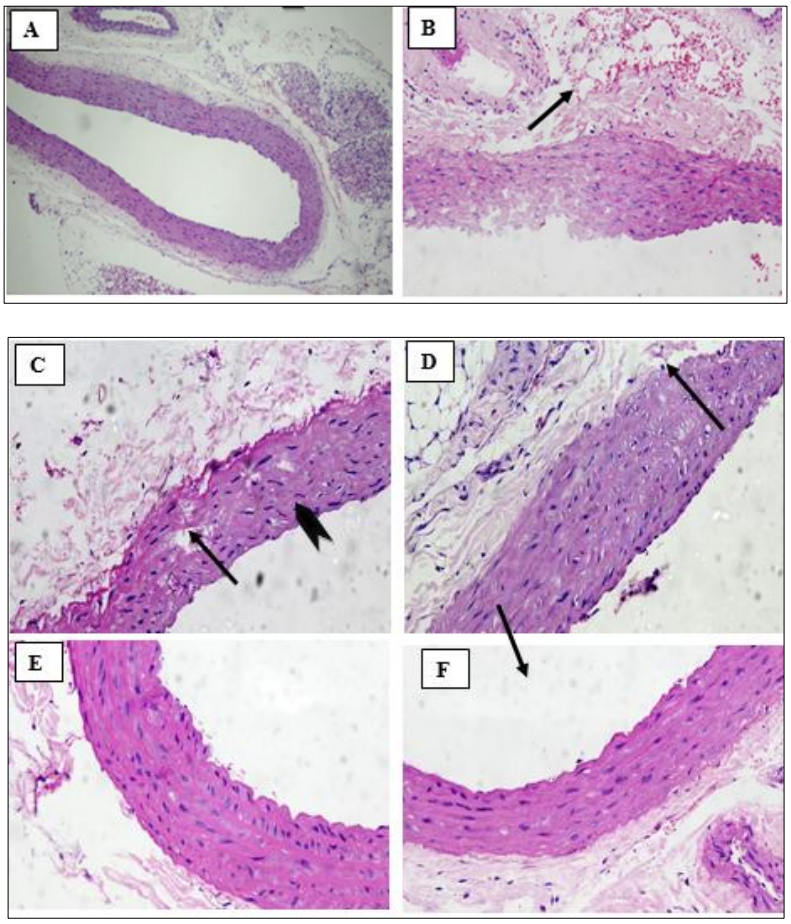


Fig 5: Light microscopy of aorta stained with H & E stain, showing normal architecture (A, 100X), arsenic exposed group showed focal thickening of T. media, (→ arrow) with irregular arrangement of elastic fibres and disruption of T. adventitia (➨ chevron) (B, 400X), telmisartan treated group showed focal disruption of smooth muscle elastic laminae (→ arrow) with mild derangement of elastic fibers (➨ chevron) (C, 400X).

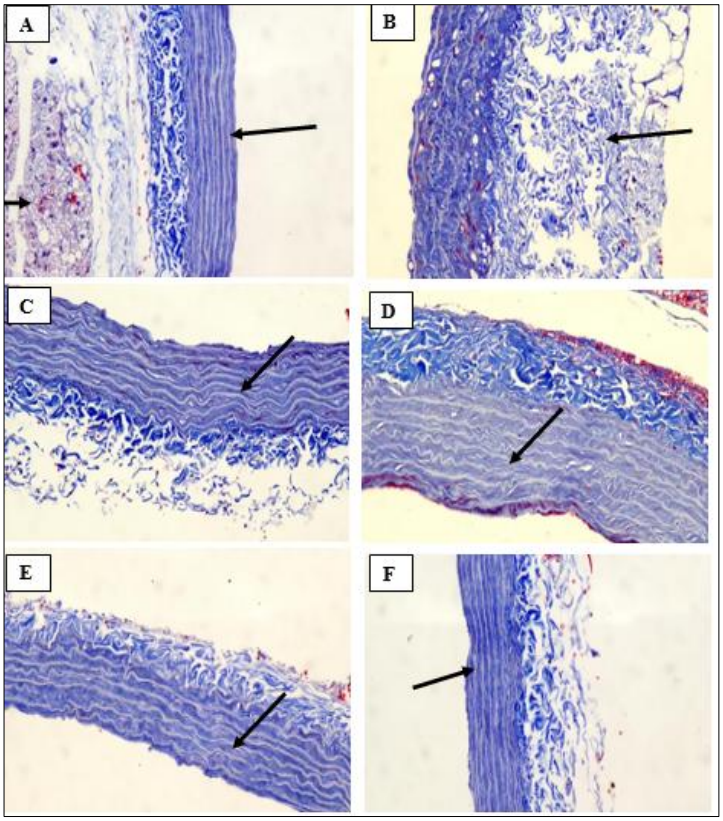


Fig 6: Light microscopy of aorta stained with Masson's Trichrome showing normal architecture with linear arrangement of elastic laminae (➞ arrow) (A, 200X). arsenic treated group showed disruption of T. adventitia (➞ arrow) disorderly arrangement of elastic laminae in the T. media layer with mild to moderate deposition of collagen (B, 400X). telmisartan treated group showed disruption of T. adventitia with derangement of elastic fibers (➞ arrow) (C, 400X). Resveratrol treated group preserved dose dependent aortic architecture, 4mg showed mild disruption of T. adventitia, and derangement of elastic laminae (➞ arrow) (D, 400X). 8mg showed mild derangement of elastic laminae (➞ arrow) (E, 400X). 16mg showed linear arrangement of elastic lamiane (➞ arrow) (F, 200X).

Table 1: The mean (± SE) body weight (g) values of rats of different experimental groups in the study at weekly interval

Groups	Day 0	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8
Group I	170.83±2.83^{a}	181.66±2.23^{a}	200.16±3.08^{a}	223.16±2.08^{d}	245.33±3.12^{de}	268.5±3.86^{d}	276.33±3.79^{c}	282.83±4.04^{bc}	294.5±3.35^{c}
Group II	172.66±3.91^{a}	179.16±3.71^{a}	191.33±2.51^{a}	202±2.9^{a}	212.83±3.3^{a}	221.16±3.13^{a}	228.83±4.62^{a}	240.83±4.43^{a}	250.33±2.98^{a}
Group III	171.16±4.61^{a}	180.5±4.57^{a}	193.16±3.84^{a}	212.33±3.26^{bc}	233.33±1.78^{c}	255.5±1.45^{bc}	264.83±1.92^{b}	273.83±1.07^{b}	277.66±1.14^{b}
Group IV	169.16±1.81^{a}	179.5±0.84^{a}	192.33±0.76^{a}	210.16±0.94^{b}	226.66±2.44^{b}	253.16±1.53^{b}	264.83±1.22^{b}	275.83±1.79^{b}	281.66±2.13^{b}
Group V	171±3.53^{a}	181.33±2.2^{a}	195.66±2.12^{a}	215.16±2.93^{bc}	239.5±0.67^{cd}	261.33±1.33^{cd}	271.16±1.97^{bc}	280.66±2.18^{bc}	289.16±1.95^{c}
Group VI	172.33±3.07^{a}	182.16±1.66^{a}	200.16±2.56^{a}	219.16±2.84^{cd}	247.16±0.94^{e}	267.5±3.49^{d}	277.83±3.26^{c}	286.83±3.61^{c}	294.83±2.05^{c}

Note: Mean±SE bearing different superscripts differ statistically significant at p <0.05 (n=6).

Table 2: The mean (± SE) values of various hematological parameters of rats in different groups on final day (61st day) of the study

Groups	Group I	Group II	Group III	Group IV	Group V	Group VI
Hb (g/dl)	14.63±0.41^{d}	10.36±0.45^{a}	12.64±0.47^{b}	11.74±0.32^{b}	12.99±0.45^{bc}	14.09±0.3^{cd}
TEC (10^{6}/µl)	7.33±0.16^{b}	6.12±0.29^{a}	7.02±0.1^{b}	6.8±0.13^{b}	7.14±0.14^{b}	7.24±0.13^{b}
TLC (10^{3}/µl)	8.35±0.08^{d}	5.3±0.38^{a}	7.01±0.32^{b}	7.32±0.14^{b}	7.64±0.22^{bc}	7.85±0.31^{cd}
PCV (%)	41.88±0.71^{d}	30.38±0.74^{a}	36.71±1.58^{bc}	36.31±0.9^{b}	38.38±2.23^{bcd}	40.66±1.33^{cd}
MCV (fL)	51.51±1.36^{a}	63.75±1.64^{b}	53.25±1.69^{a}	54.54±0.89^{a}	52.75±1.08^{a}	51.76±0.75^{a}
MCH (pg)	17.01±0.53^{a}	25.13±1.44^{d}	21.62±1.08^{bc}	23.12±0.82^{cd}	20.92±1.14^{bc}	18.67±0.69^{ab}
MCHC (%)	32.76±0.83^{d}	26.2±0.88^{a}	31.04±0.72^{bc}	30.16±0.52^{b}	31.26±0.85^{bc}	33.06±0.66^{d}

Note: Mean±SE bearing different superscripts differ statistically significant at *p* <0.05 (n=6).

Table 3: The mean (± SE) values of various serum biochemical parameters of rats in different groups on final day (61st day) of the study

Groups	Group I	Group II	Group III	Group IV	Group V	Group VI
ALP IU/L	107.54±1.08^{a}	131.04±2.49^{c}	109.93±1.86^{a}	124.03±1.12^{b}	110.99±0.97^{a}	108.08±1.16^{a}
ALT IU/L	41.88±0.42^{a}	71.22±1.16^{d}	48.56±1.51^{b}	56.85±1.62^{c}	48.43±1.52^{b}	45.43±1.19^{ab}
TP g/dl	8.58±0.27^{d}	5.71±0.25^{a}	7.44±0.19^{c}	6.74±0.1^{b}	7.35±0.18^{c}	8.05±0.07^{d}
CRT mg/dl	0.55±0.01^{a}	0.84±0.04^{b}	0.67±0.02^{a}	0.80±0.03^{b}	0.68±0.06^{a}	0.63±0.06^{a}
BUN mg/dl	16.37±0.99^{a}	25.76±1.37^{d}	20.53±0.72^{b}	23.32±0.21^{c}	20.07±0.49^{b}	18.37±0.5^{ab}

Note: Mean±SE bearing different superscripts differ statistically significant at *p* <0.05 (n=6).

Table 4: The mean (± SE) values of various serum oxidative stress parameters of rats in different groups on final day (61st day) of the study

Groups	Group I	Group II	Group III	Group IV	Group V	Group VI
LPO (nmol MDA/g of tissue)	7.07±0.11[a]	27.90±0.61[d]	7.59±0.35[a]	15.86±0.34[c]	10.91±0.38[b]	7.10±0.30[a]
CAT ('mmol' H2O2 utilized/min/mg protien)	47.07±0.11[c]	31.52±0.48[a]	42.59±0.52[b]	41.50±0.84[b]	43.43±0.93[b]	46.83±1.26[c]
SOD (U/mg of protein)	7.90±0.31[b]	6.02±0.28[a]	7.63±0.36[b]	7.10±0.25[b]	7.64±0.17[b]	7.96±0.23[b]

Note: Mean±SE bearing different superscripts differ statistically significant at $p < 0.05$ (n=6).

Conclusion

The present study confirmed that oral administration of sodium arsenite (10 mg/kg b.wt.) for 60 days effectively induced systemic toxicity in male Wistar rats, as shown by altered hematobiochemical values, oxidative stress, lipid peroxidation, upregulated pro-inflammatory cytokines, NOX-2 gene expression, and vascular lesions. Telmisartan (10 mg/kg b.wt.) showed marked protective effects by restoring antioxidant balance, reducing oxidative stress and inflammation, and maintaining vascular integrity, serving as the standard antioxidant agent. Resveratrol produced dose-dependent protection, with higher doses (8 mg/kg and 16 mg/kg b.wt.) antioxidant, and histopathological parameters compared to the lower dose (4 mg/kg b.wt.). Overall, resveratrol exhibited strong vasculoprotective and antioxidative effects, highlighting its potential as a preventive and adjunct therapy against arsenic toxicity, though further mechanistic and clinical validation is required.

References

1. Acharyya N, Deb B, Chattopadhyay S, Maiti S. Arsenic-induced antioxidant depletion, oxidative DNA breakage, and tissue damages are prevented by the combined action of folate and vitamin B12. Biol Trace Elem Res. 2015;168(1):122-32.
2. Aebi H. Catalase. In: Methods of Enzymatic Analysis. 2nd ed. Academic Press; 1983. p. 673-84.
3. Alenzi FQ, Salem ML, Alenazi WS, Alotaibi MR. Oxidative stress and hepatotoxicity: the role of alkaline phosphatase. J Appl Toxicol. 2010;30(7):600-8.
4. Baur JA, Sinclair DA. Therapeutic potential of resveratrol: the *in vivo* evidence. Nat Rev Drug Discov. 2006;5(6):493-506.
5. Benson SC, Pershadsingh HA, Ho CI, Chittiboyina A, Desai P, Pravenec M, *et al*. Identification of telmisartan as a unique angiotensin II receptor antagonist with selective PPAR-gamma-modulating activity. Hypertension. 2004;43:993-1002.
6. Bhowmick S, Pramanik S, Singh P, Mondal P, Chatterjee D, Nriagu J. Arsenic in groundwater of West Bengal, India: a review of human health risks and assessment of possible intervention options. Sci Total Environ. 2018;612:148-69.
7. Cheng XM, Hu YY, Yang T, Wu N, Wang XN. Reactive oxygen species and oxidative stress in vascular-related diseases. Oxid Med Cell Longev. 2022;2022:70-91.
8. Das AK, Bag S, Gangopadhyay M, Sinha MK, Dewanjee S. Arsenic-induced myocardial injury: protective role of Corchorus olitorius leaves. Food Chem Toxicol. 2012;50(10):3451-7.
9. Ellinsworth DC, Shukla D, Fleming SM, Kelly KE. Vascular toxicity and oxidative stress from arsenic exposure. Toxicol Sci. 2015;145(2):347-57.
10. Fouad AA, Al-Mulhim AS, Jresat I. Telmisartan treatment attenuates arsenic-induced hepatotoxicity in mice. Toxicology. 2012;300(3):149-57.
11. Garcia-Chavez E, Santamaria A, Diaz-Barriga F, Mandeville P, Bertha I, Juarez M, *et al*. Arsenite-induced formation of hydroxyl radical in the striatum of awake rats. Brain Res. 2003;976:82-9.
12. Gowda BR, Prakash N, Santhosh CR, Pavithra BH, Rajashekaraiah R, Sathyanarayana ML, *et al*. Effect of telmisartan on arsenic-induced (sub-chronic) perturbations in redox homeostasis, pro-inflammatory cascade and aortic dysfunction in Wistar rats. Biol Trace Elem Res. 2022;200(4):1776-90.
13. He H, Mostofa M, Ghosh A. Protective antioxidant effect of telmisartan in oxidative stress. J Hypertens. 2017;35(2):345-54.
14. Hou S, Chen H, Yu X, Liu W. Reactive oxygen species mediated hepatotoxicity of arsenic in experimental models. Toxicol Lett. 2007;174(1-3):1-9.
15. Jaishankar M, Tseten T, Anbalagan N, Mathew BB, Beeregowda KN. Toxicity, mechanism and health effects of some heavy metals. Interdiscip Toxicol. 2014;7(2):60-72.
16. Jalaludeen A, Ramesh V, Karthikeyan S. Arsenic-mediated aortic structural changes. Toxicol Mech Methods. 2015;25(4):287-94.
17. Khatun M, Haque N, Siddique AE, Wahed AS, Islam MS, Khan S, *et al*. Arsenic exposure-related hypertension in Bangladesh and reduced circulating nitric oxide bioavailability. Environ Health Perspect. 2024;132(4):47-61.
18. Li H, Xia N, Hasselwander S, Daiber A. Resveratrol and vascular function. Int J Mol Sci. 2019;20(9):21-55.
19. Lu M, Fang Y, Wei Q, Liu L. Renal dysfunction induced by chronic arsenic exposure in rats. Toxicol Mech Methods. 2011;21(2):122-9.
20. Madesh M, Balasubramanian KA. Microtiter plate assay for superoxide dismutase using MTT reduction by superoxide. Indian J Biochem Biophys. 1998;35(3):184-8.
21. Mazumder DN. Chronic arsenic toxicity: clinical features and cellular mechanisms. Indian J Med Res. 2005;122(5):407-16.
22. Mognetti B, Franco F, Castrignano C, Bovolin P, Berta GN. Mechanisms of phytoremediation by resveratrol against cadmium toxicity. Antioxidants. 2024;13(7):782.
23. Nirwane A, Pawar V, Majumdar A. Therapeutic interventions using a combination of telmisartan and omega-3 fatty acids in sodium arsenite-induced vascular endothelial dysfunction in rats. J Complement Integr Med. 2015;12(2):143-51.
24. Oak MH, El Bedoui J, Schini-Kerth VB. Resveratrol protects cardiovascular tissues via antioxidant effects. Br J Pharmacol. 2005;146(3):368-75.

25. Pandey KB, Mehrotra S, Rizvi SI. Resveratrol protects erythrocytes from arsenic-induced oxidative stress. Chem Biol Interact. 2010;185(3):218-24.
26. Paul S, Chatterjee A, Das S. Hepatorenal toxicity of arsenic in experimental animals. Indian J Exp Biol. 2007;45(9):810-5.
27. Pi J, Yamamoto M, Kumagai Y. Reactive oxygen species as a mechanism of arsenic toxicity. Toxicol Appl Pharmacol. 2003;193(3):203-14.
28. Park SJ, Ahmad F, Philp A, Baar K, Williams T, Luo H, *et al*. Resveratrol ameliorates aging-related metabolic phenotypes by inhibiting cAMP phosphodiesterases. Cell. 2012;148(3):421-33.
29. Paula F, Gouvêa CM, Alfredo PP, Salgado I. Protective action of a hexane crude extract of Pterodon emarginatus fruits against oxidative and nitrosative stress induced by acute exercise in rats. BMC Complement Altern Med. 2005;5(1):1-9.
30. Ray PS, Maulik G, Cordis GA, Bertelli AA, Bertelli A, Das DK. The red wine antioxidant resveratrol protects isolated rat hearts from ischemia reperfusion injury. Free Radic Biol Med. 1999;27(2):160-9.
31. Rizvi SI, Pandey KB. Activation of the erythrocyte plasma membrane redox system by resveratrol: a possible mechanism for antioxidant properties. Pharmacol Rep. 2010;62(4):726-32.
32. Robb EL, Stuart JA, Hockenbery DM. Resveratrol protects hepatic and splenic tissue from oxidative damage. Free Radic Biol Med. 2008;44(10):1874-82.
33. Saad SA, Ibrahim AM, Mohamed SA. Arsenic-induced aortic degeneration. Hum Exp Toxicol. 2006;25(3):137-45.
34. Sinha M, Gurusamy N, Lakshmanan AP. Arsenic-induced vascular changes in rats. Toxicol Lett. 2008;178(3):247-54.
35. Schupp N, Jansen F, van Wijk A. Telmisartan protects against oxidative stress by AT1R blockade and PPAR-γ activation. Hypertens Res. 2004;27(12):921-7.
36. Sharma A, Prasad AK, Shankar R. Resveratrol attenuates arsenic-induced hepatic damage. Toxicol Appl Pharmacol. 2007;222(2):222-8.
37. Sharma P, Kumar A, Singh RK. Nephrotoxic effects of chronic arsenic exposure in rats. Toxicol Rep. 2019;6:1234-42.
38. Sharma R, Upadhyay SN, Singh RK. Telmisartan activates antioxidant pathways. Eur J Pharmacol. 2010;627(3):164-70.
39. Sonmez MF, Suzuki K, Kodama M, Watanabe K. Protective role of resveratrol against arsenic-induced nephrotoxicity. Biol Trace Elem Res. 2016;171(1):176-84.
40. Souza ACF, de Paiva Coimbra JL, Ervilha LOG, Bastos DSS, Cossolin JFS, Santos EC. Arsenic induces dose-dependent structural and ultrastructural pathological remodeling in the heart of Wistar rats. Life Sci. 2020;257:132-51.
41. Tseng CH. A review on arsenic methylation, urinary metabolites, and health effects. Toxicol Appl Pharmacol. 2009;235(3):338-50.
42. Turk T, Murillo AM, Velez MP. Global epidemiology of chronic arsenicosis: public health perspectives. Environ Res. 2019;176:538-43.
43. Ungvari Z, Orosz Z, Rivera A, Labinskyy N, Xiangmin Z, Smith K, *et al*. Resveratrol attenuates oxidative stress and inflammation: implications for vascular aging. J Gerontol A Biol Sci Med Sci. 2010;65(8):870-7.
44. Vahter M. Mechanisms of arsenic biotransformation. Toxicology. 2002;181-2:211-7.
45. Valko M, Morris H, Cronin MTD. Metals, toxicity and oxidative stress. Curr Med Chem. 2005;12(10):1161-208.
46. Waghe P, Sarath TS, Gupta P, Kutty HS, Kandasamy K, Mishra SK, *et al*. Sub-chronic arsenic exposure through drinking water alters vascular redox homeostasis and affects physical health in rats. Biol Trace Elem Res. 2014;162(1):234-41.
47. Waghe P, Sarath TS, Gupta P, Kandasamy K, Choudhury S, Kutty HS, *et al*. Arsenic causes aortic dysfunction and systemic hypertension in rats: augmentation of angiotensin II signaling. Chem Biol Interact. 2015;237:104-14.
48. Wang Z, Fleming I, Münzel T, Daiber A. Nrf2 activation and protection against arsenic-induced damage. Toxicol Appl Pharmacol. 2017;336:25-34.
49. Xia N, Daiber A, Habermeier A, Closs EI, Thum T, Spanier G, *et al*. Resveratrol protects against oxidative stress and vascular dysfunction. Br J Pharmacol. 2017;174(14):1771-89.
50. Yang X, Zhang T, Li Y. Arsenic-induced hepatotoxicity and protein synthesis impairment in rats. J Trace Elem Med Biol. 2019;54:68-74.
51. Yadav KK, Gupta N, Kumar V, Singh JK. Bioremediation of heavy metals from contaminated sites using potential species: a review. Indian J Environ Prot. 2017;37(1):65-80.
52. Zhang J, Zhu G, Xu Y, Sun J, Wu H. Clinical and histopathological signs of arsenic toxicity in rats. Environ Pollut. 2021;277:50-68.