Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4, 3146-3158
2025
Publisher: Learning Gate
DOI: 10.55214/25768484.v9i4.6745

The prophylactic effect of aqueous chamomile extract on sulfur dioxide exposed mice

Maryam Kbaysi[1], Ruzanna Petrosyan[2], Jamilah Borjac[1*]
[1]Beirut Arab University, Department of Biological Sciences, Faculty of Sciences, Debbieh Campus, Lebanon; j.borjac@bau.edu.lb (J.B.).
[2]Lebanese American University, Gilbert and Rose-Marie Chagoury School of Medicine, Beirut, Lebanon.

Abstract: Sulfur Dioxide (SO_2) is an atmospheric contaminant known for its harmful effects on various organs. Chamomile, traditionally used for several years, has shown efficacy in treating and alleviating various disorders due to its antioxidant properties. This study aims to examine the prophylactic effect of Chamomile extract on mice exposed to sulfur dioxide. Four groups of Balb/c mice (n=9 per group) were used. Group 1 received no treatment (control); Group 2 was given Chamomile extract orally at 200 mg/kg body weight for 14 days; Group 3 was exposed to SO_2 (sodium sulfite and sodium bisulfite, 3:1 ratio) via intraperitoneal injection at 690 mg/kg for 7 days; Group 4 received Chamomile for 7 days before co-administration with SO_2 for an additional 7 days. After treatment, blood was collected and the liver, brain, and lungs were harvested for histological and biochemical analyses. SO_2 exposure elevated liver enzymes and malondialdehyde levels, reduced SOD and CAT activity, and caused lung histological damage without brain alterations. Chamomile extract significantly alleviated these effects. Chamomile shows preventive effects against SO_2-induced damage, supporting its use as a protective agent against environmental contaminants.

Keywords: *Antioxidant, Chamomile, Liver damage and lung damage, Oxidative stress, Sulfur Dioxide.*

1. Introduction

Over the past decade, industrial growth has led to a proliferation of environmental health concerns due to the rise in emitted pollutants. A significant gaseous pollutant of industrial activity is sulfur dioxide (SO2) [1]. Upon inhalation of SO_2, it converts into sulfurous acid upon contact with moisture in the respiratory system. This acid further dissociates to produce bisulfite and sulfite ions, which maintain a ratio of 1:3 under neutral circumstances [2]. Inhalation of these compounds might lead to respiratory distress and other detrimental effects. In addition to the presence of SO2 in the atmosphere, sulfiting chemicals are widespread in food, beverages, and medications as preservatives.

The detrimental consequences of sulfur dioxide originate from its ability to diffuse throughout the bloodstream, thereby affecting all bodily tissues and organs and resulting in systemic health complications [3, 4]. Respiratory issues may arise from the release of sulfite ions from sulfur dioxide and various sulfiting agents, such as sodium sulfite, sodium bisulfite, sodium metabisulfite, and potassium metabisulfite [5]. The release of these ions can induce the formation of free radicals, resulting in oxidative stress and an inflammatory response. Moreover, they may induce allergy or hypersensitive reactions, especially in those sensitive to sulfites [6, 7]. These reactions may vary from dermatological problems to severe systemic symptoms, including anaphylaxis. In addition to the aforementioned detrimental effects, SO2 may exhibit carcinogenic properties and promote cardiovascular issues [8].

History: Received: 12 February 2025; Revised: 13 March 2025; Accepted: 17 March 2025; Published: 30 April
* Correspondence: j.borjac@bau.edu.lb

Treatment using herbal extracts, especially those rich in powerful antioxidants, has garnered significant interest for mitigating the detrimental effects of pollutants. Chamomile, a plant widely distributed across Asia and Europe, has been utilized for medicinal purposes by our ancestors. The extract is utilized for the treatment of several illnesses, including cancer and mental health difficulties. Chamomile is renowned for its calming and soothing effects and is commonly prepared as tea to alleviate anxiety, enhance sleep, and relieve digestive issues [9]. The therapeutic efficacy of chamomile is attributed to its phytochemical components, including flavonoids (such as luteolin and apigenin), terpenoids (notably chamazulene), and phenolic acids. These phytochemicals possess significant antioxidant capabilities, contributing to the mitigation of oxidative stress. Antioxidants are essential for enhancing the body's detoxifying abilities and alleviating the harmful impacts of oxidative stress, resulting from an imbalance between reactive oxygen species (ROS) and antioxidant levels. Antioxidants work on neutralizing free radicals, thereby mitigating their deleterious effects on proteins, lipids, and DNA. Chamomile's anti-inflammatory properties, driven by bioactive compounds that can regulate inflammatory pathways, may reduce inflammation caused by sulfite exposure. The aforementioned qualities may act as a mitigating factor against oxidative damage caused by sulfur dioxide [10, 11].

This study aims to investigate whether prophylactic administration of chamomile extract can provide protection against sulfur dioxide-induced damage in mice.

2. Materials and Methods

2.1. Preparation of Chamomile Extract

Chamomile was purchased from a local herb store located in Tyre, South Lebanon. The identification was conducted using taxonomic keys of the flora as outlined by [12]. A voucher has been deposited at the Research Center for Environment and Development located at the University. A 5% extract was prepared by infusing 5 g of dried chamomile flowers in 100 ml of boiling water for 40 minutes, followed by filtration. The filtrate was subsequently dried in an oven at 40 °C. The pellet was subsequently dissolved in distilled water to achieve a final concentration of 20 mg/ml and stored in aliquots at -20°C until further use.

2.2. Animals

Female Balb/c mice, aged 6 to 8 weeks, were obtained from the Animal House. The subjects were maintained in a controlled laboratory environment at a temperature of 22 ± 2°C, with a 12-hour light/dark cycle and access to food and water. All animal experiments adhered to the ethical standards approved by the Institutional Review Board, under code number: 2023-A-0054-S-M-0555.

The mice were assigned randomly to four groups, each consisting of nine mice:

- Group 1 (Control): Mice received no treatment throughout the study.
- Group 2 (Extract Treatment): Mice were administered 200 mg/kg body weight of chamomile extract daily through oral gavage for a duration of 14 days.
- Group 3 (SO_2): Mice were administered a mixture of sodium sulfite and sodium bisulfite at a 3:1 molar ratio, with a dosage of 690 mg/kg delivered via intraperitoneal injections over a period of 7 days.
- Group 4 (SO_2 + Extract Treatment**):** Mice received a pre-treatment of chamomile extract at a dosage of 200 mg/kg for a duration of 7 days, followed by a co-administration of the chamomile extract and a combination of sodium sulfite and sodium bisulfite, as outlined in Group 3, for an additional 7 days.

At the end of the experiment, the mice were anesthetized using isoflorane drop jar method, and blood samples were obtained. Organs were subsequently dissected for histological and biochemical analysis.

Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4: 3146-3158, 2025
DOI: 10.55214/25768484.v9i4.6745

2.3. Biochemical Analysis

Blood samples were obtained through cardiac puncture under anaesthesia and sent to the Professional Healthcare Diagnostic Laboratory in Saida, Lebanon, for analysis. The samples underwent the following analyses:

- Complete Blood Count (CBC): Performed using a Sysmex XP-300 automated hematology analyzer.
- Kidney and Liver Function Tests: Serum levels of blood urea nitrogen (BUN), creatinine, alanine aminotransferase (ALT), and aspartate aminotransferase (AST) were measured using a Roche Cobas c111 automated chemistry analyzer.
- Glucose Levels: Blood glucose levels were also determined using the Roche Cobas c111 automated chemistry analyzer.

2.4. Histological Analysis

Liver, brain, and lung tissues were harvested from anesthetized mice, fixed in 10% formalin, and dispatched to the Institute National de Pathologie in Beirut, Lebanon, for histological analysis. Upon arrival, the specimens were dehydrated, cleared, and embedded in paraffin blocks. The paraffin-embedded tissues were sliced into 5 µm thick sections utilizing a microtome. The tissue sections were stained with hematoxylin and eosin (H&E) to evaluate structural changes and were observed under a microscope at 10X and 40X magnifications.

2.5. Tissue Homogenization

Liver and brain tissues were homogenized in phosphate-buffered saline (PBS) with a pH of 7.4, containing 1 mM phenylmethylsulfonyl fluoride (PMSF) as a protease inhibitor to prevent protein degradation. The tissue-to-buffer ratio was established at 1 gram of tissue per 5 mL of solution. Homogenization was conducted manually on ice for a duration of 15 minutes. The homogenates underwent centrifugation at 10,000 rpm for 15 minutes at 4°C. The resulting supernatants, which contained the extracted proteins, were collected and stored at -80°C for subsequent analysis.

2.6. Protein Quantification

The protein concentration in each tissue homogenate was assessed using the Bradford assay Bradford [13] a colorimetric technique reliant on dye-protein interactions. Bovine serum albumin (BSA) served as a standard for the development of a calibration curve. The assay consisted of incubating 50 µl of each homogenate with 200 µl of 1X Bradford reagent for 30 minutes at room temperature in the absence of light. Absorbance was recorded at 595 nm utilizing a UV-Visible Spectrophotometer, employing PBS and Bradford reagent as the blank reference. Protein concentrations were determined by comparing the absorbance values of samples to the BSA standard curve, which ranged from 0 to 10 µg/mL.

2.7. Oxidative Stress Markers:

2.7.1. Superoxide Dismutase (SOD) Assay

The activity of Superoxide Dismutase (SOD) was assessed in each tissue homogenate by a spectrophotometric approach based on the suppression of nitroblue tetrazolium (NBT) reduction by superoxide radicals [14]. The results were quantified in units per milligram of protein (U/mg protein).

2.7.2. Catalase (CAT) Assay

Catalase (CAT) activity was assessed in each tissue homogenate by quantifying the decomposition of hydrogen peroxide (H_2O_2) at 240 nm [15]. The results were expressed in units per milligram of protein (U/mg protein).

Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4: 3146-3158, 2025
DOI: 10.55214/25768484.v9i4.6745

2.7.3. Malondialdehyde (MDA) Assay

Malondialdehyde (MDA) content, a marker of lipid peroxidation, was determined using the thiobarbituric acid reactive substances (TBARS) assay [16]. MDA concentrations were calculated and expressed as nmol MDA per mg of protein.

2.8. Statistical Analysis

All statistical analyses and graphs were conducted using GraphPad Prism software, with data expressed as means ± standard deviations. Statistical significance was assessed utilizing the Tukey test subsequent to a one-way ANOVA. Significance levels denoted as follows: **** $P < 0.0001$, *** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$.

3. Results

3.1. Haematological Parameters

Table 1 illustrates the results obtain on haematological parameters across all study groups. Haematocrit levels (HCT) in the SO_2 group increased significantly by 29% ($p<0.05$) compared to the Control group. The chamomile treatment in SO_2+Extract group did not restore HCT levels back to normal.

SO_2 exposure significantly raised Mean Corpuscular Haemoglobin (MCV) by 14% ($p<0.05$) in SO_2 group compared to the Control group and by 10% ($p<0.05$) compared to the Extract treatment group. Chamomile extract treatment did not normalize MCV values in SO_2+ Extract group.

The platelet counts (PLT) in the Extract treatment group, SO_2 group, and SO_2+ Extract group dropped significantly compared to the Control by 57% ($p<0.01$), 34% ($p<0.05$), and 43% ($p<0.01$), respectively. No statistically significant variations were detected among the other haematological markers.

Table 1.
Hematological analysis of all groups.

Parameters	Group 1 (Control)	Group 2 (Extract Treatment)	Group 3 (SO_2)	Group 4 (SO_2 + Extract)
RBCs (10^6/µL)	5.98 ± 0.86	6.62 ± 0.43	7.28 ± 0.71	7.04 ± 0.61
HGB, (g/dL)	12.53 ± 0.81	14.07 ± 0.90	14.57 ± 1.22	13.70 ± 1.22
HCT, (%)	34.67 ± 5.23	39.50 ± 1.74	48.60 ± 6.65 [a]	43.10 ± 2.69
MCV	57.90 ± 0.73	59.90 ± 2.62	66.6± 0.84 [ab]	62.45 ± 0.31
MCH	21.20 ± 1.98	21.23 ± 0.40	20.03 ± 1.90	19.77 ± 0.83
MCHC	36.63 ± 3.70	35.57 ± 2.00	30.20 ± 3.43	31.70 ± 1.37
PLT, (10^3/µL)	808.67 ± 107.57	348.00 ± 38.27 [a]	533.00 ± 110.24 [a]	458.33 ± 64.35 [a]
WBC (10^9/L)	5.97± 0.82	5.19± 0.42	4.74 ± 2.98	1.83 ± 0.96
NEU, (%)	6.33± 0.47	4.33 ± 0.94	11.33 ± 8.51	11.66 ± 5.86
LYM, (%)	74.00 ± 1.41	76.00 ± 14.85	79.67 ± 12.58	68.67 ±13.32
MON, (%)	19.33 ± 1.25	19.00 ± 14.90	7.67 ± 4.16	18.33 ± 9.24
EOS, (%)	0.33 ± 0.47	0.67 ± 0.47	1.33 ± 0.58	1.33 ± 0.58
BASO, (%)	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
RDW	17.83 ± 1.50	19.00 ± 1.13	18.43 ± 0.16	19.70 ± 1.39

Note: Data are presented as mean ± SD. [a]$p<0.05$ vs. the Control group, [b]$p<0.05$ vs. the Extract treatment group.

3.2. Fasting Blood Glucose

No statistically significant variations in fasting blood glucose levels were detected among the experimental groups.

3.3. Liver Function (ALT and AST)

The impact of chamomile extract treatment on serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) levels was illustrated in Figure 1. The Control and Extract treatment groups

Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4: 3146-3158, 2025
DOI: 10.55214/25768484.v9i4.6745

exhibited comparable outcomes. AST levels markedly elevated in the SO_2 group relative to the Control and Extract treatment groups by 90% (P <0.01) and 83% (P <0.01) respectively. The prophylactic administration of chamomile extract in SO_2+Extract group significantly diminished the SO_2-induced increase in AST by 77% (P <0.01).

Similarly, the ALT levels were comparable across the Control and Extract treatment groups. ALT levels markedly elevated in SO_2 group in comparison to the Control and Extract Treatment groups by 95% (P <0.01) and 96% (P <0.01), respectively. The prophylactic administration of chamomile extract in SO_2+ Extract group resulted in a considerable 90% reduction in the SO_2-induced elevation of ALT (P <0.01).

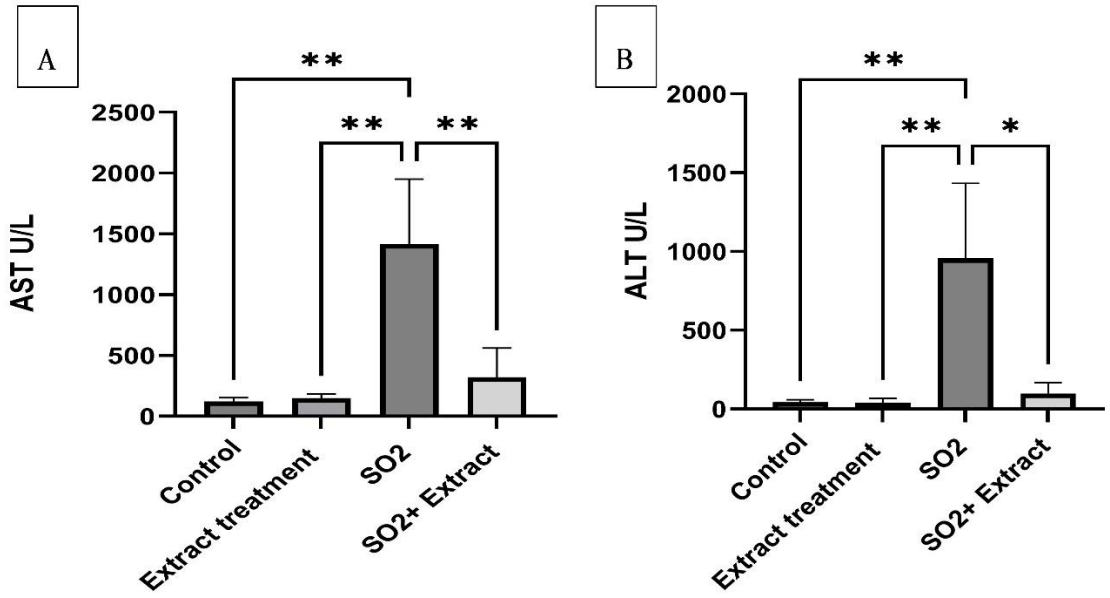

Figure 1.
AST and ALT Serum Levels of all study groups. (A) AST and (B) AST.
Note: mean±SD.*p < 0.05, **p < 0.01, ***p < 0.001.

3.4. Kidney Function (Creatinine, Urea and BUN)

Kidney function parameters (creatinine, Urea, and BUN) were determined across all study groups as illustrated in Figure 2.

Creatinine levels did not exhibit any substantial differences between the study groups (Figure 2A). Although the SO_2 group exhibited higher Urea and BUN levels than the Control group, the results were not statistically significant. On the other hand, the SO_2 group exhibited a statistically significant increase in Urea and BUN when compared to the Extract treatment group by 48% (p <0.01). chamomile treatment in SO_2 + Extract group resulted in a 40% decrease in BUN and Urea results (p <0.01) when compared to the SO_2 group. (figure 2B and 2C)

Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4: 3146-3158, 2025
DOI: 10.55214/25768484.v9i4.6745

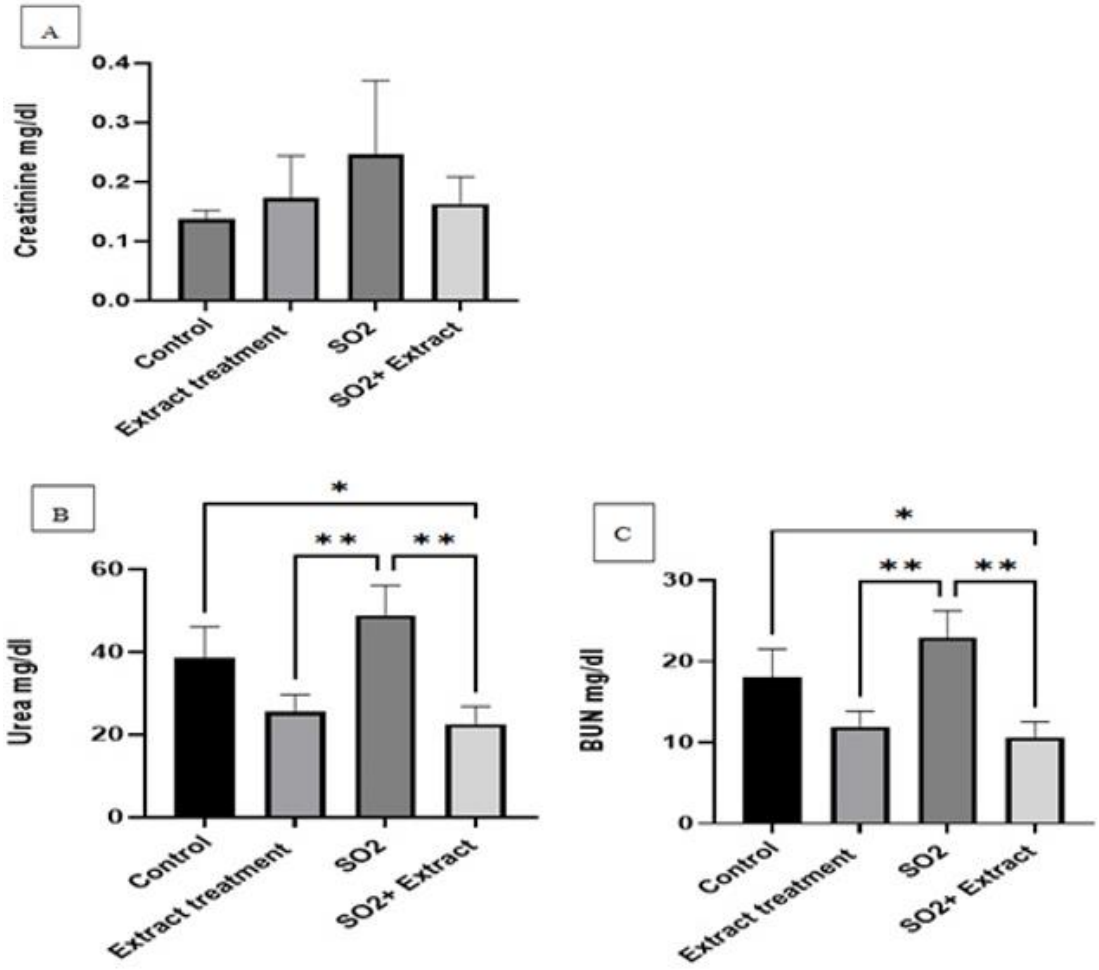

Figure 2.
Creatinine, Urea and BUN serum levels of all study groups. (A) Creatinine, (B) Urea and (C) BUN.
Note: mean±SD.*$p < 0.05$, **$p < 0.01$, ***$p < 0.001$.

3.5. Oxidative Stress Parameters in Liver Homogenate

MDA content, SOD, and CAT activity were assessed in liver homogenates of all groups as shown in figure 3A, 3B, and 3C, respectively.

Exposure to SO_2 led to a significant increase in MDA levels by 51% ($p < 0.05$) and 59% ($p < 0.01$) compared to the Control and Extract treatment groups, respectively. Fortunately, treating the SO_2 group with the Chamomile extract minimizes its damaging effect as seen by the significant decrease in MDA levels (38%, $p < 0.05$) in the SO_2+ Extract group.

Exposure to SO_2 significantly decreased SOD activity levels by 62% ($p < 0.01$) and 59% ($p < 0.01$) compared to the Control and Extract treatment groups, respectively. Administration of chamomile extract treatment minimizes the harmful effect of SO_2 by increasing SOD activity levels (49%, $p<0.05$) in the SO_2+ Extract group compared to the SO_2 group.

SO_2 exposure reduced CAT levels by 77% ($p < 0.001$) and 74% ($p < 0.001$) when compared to the control and Extract treatment groups respectively.

Chamomile extract treatment increased CAT activity in SO_2 + Extract group by 65% ($p < 0.05$) when compared to the SO_2 group.

Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4: 3146-3158, 2025
DOI: 10.55214/25768484.v9i4.6745

Although the SO2 + Extract group had higher levels of CAT than the SO2 group, they still had significantly lower levels than the control and chamomile groups ($p < 0.05$), indicating that chamomile treatment increased CAT activity but did not restore CAT levels to those of the control and chamomile groups.

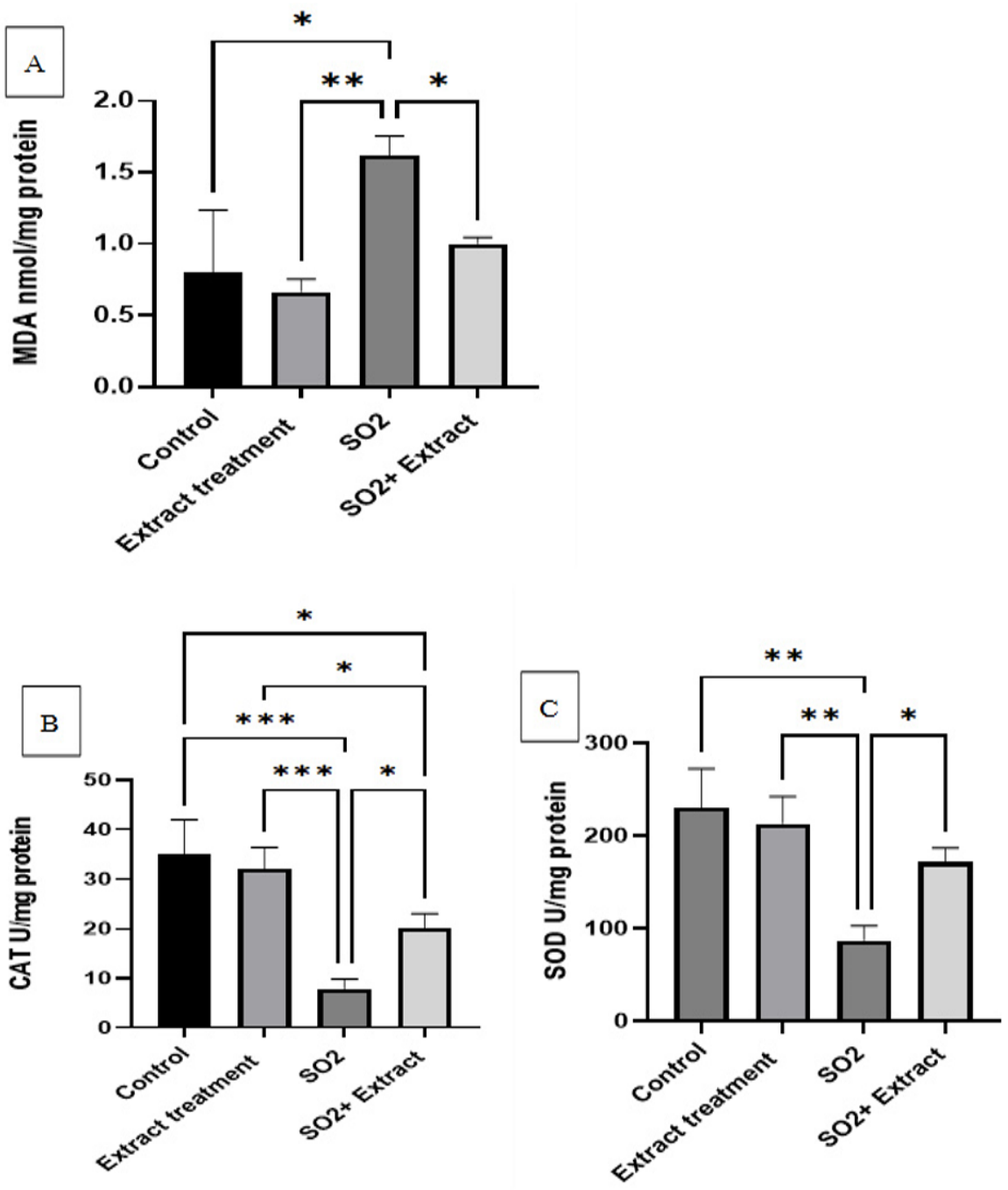

Figure 3.
MDA, CAT and SOD Liver homogenate levels of all study groups (A) MDA, (B) CAT and (C) SOD.
Note: mean±SD.*$p < 0.05$, **$p < 0.01$, ***$p < 0.001$.

Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4: 3146-3158, 2025
DOI: 10.55214/25768484.v9i4.6745

3.6. Oxidative Stress Parameters in Brain Homogenate

The MDA content, CAT and SOD activity were assessed in the brain homogenates of all groups. No statistically significant changes were observed in these measured parameters among all groups.

3.7. Histology Observations

Histopathological analysis of the lungs of SO_2 exposed mice showed signs of tissue damage indicated by perivascular inflammatory cellular infiltrate, septal edema, congestions, with increased extravasation of red blood cells into the lung parenchyma and thickening of the alveolar wall with collapse of terminal bronchioles (Figure 4C) compared to the Control and Extract treatment groups (Figure 4A and 4B). The harmful effect of SO_2 was mitigated upon treatment with chamomile extract as shown in Figure 4D.

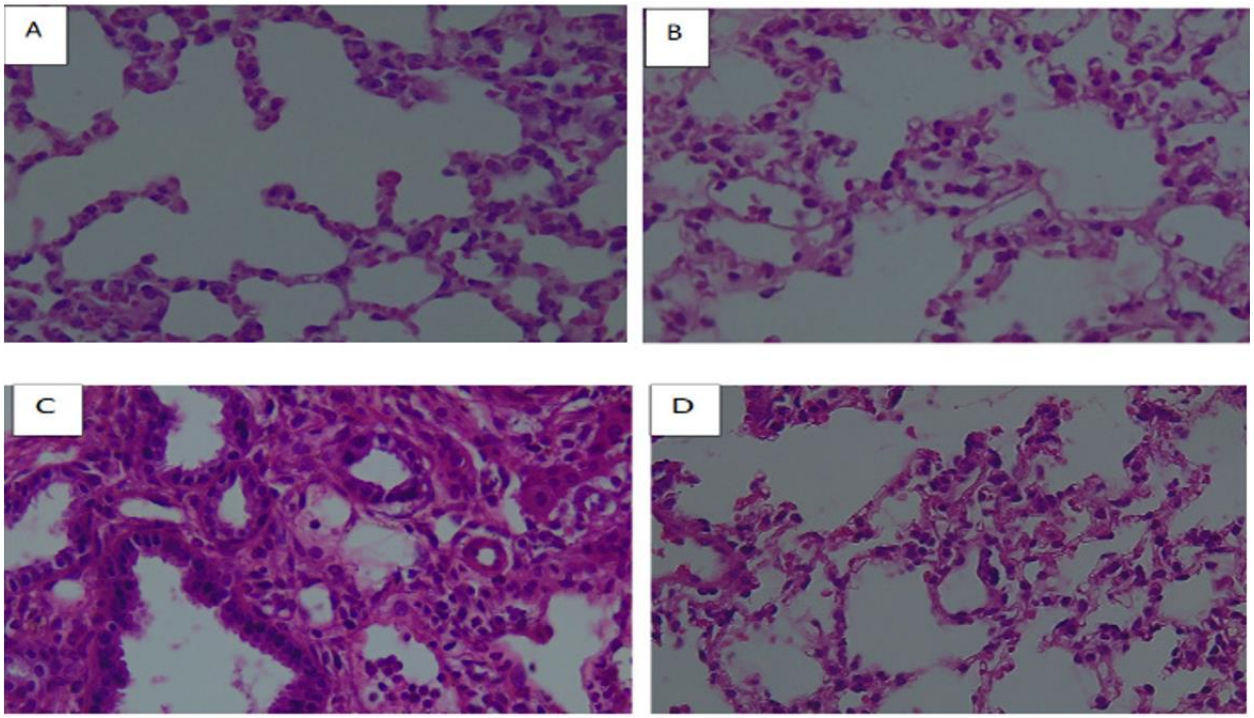

Figure 4.
Histology of lung sections of all study groups.
(A) Control, (B) Extract Treatment, (C) SO_2 group and (D) SO_2+ Extract group. Slides were stained with H& E staining and examined under 10X magnification.

Similarly, SO_2 exposure induced hepatocytes damage as indicated by micro-ballooning degeneration that is characterized by enlarged and swollen hepatocytes, and loss of their usual polygonal shape with granular material in the cytoplasm representing a collapsed cytoskeleton. Some hepatocytes contain condensed pyknotic nuclei, while others contain glycogenic nuclei. Areas of disorganized hepatic architecture are shown in Figure 5C that were restored to normal upon treatment with the extract (Figure 5D). Normal histology of liver tissue was observed in Control and Extract treatment groups (Figure 5A and 5B).

Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4: 3146-3158, 2025
DOI: 10.55214/25768484.v9i4.6745

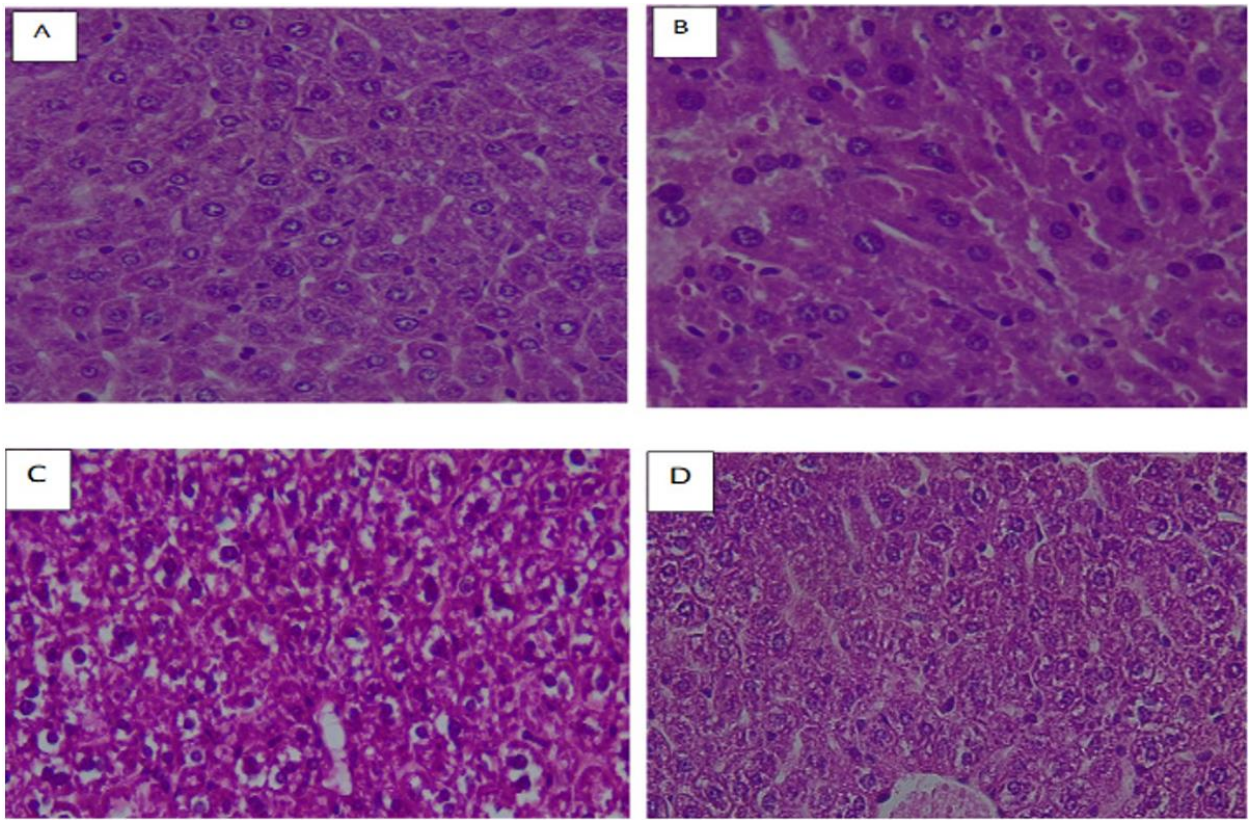

Figure 5.
Histology of liver sections of all study groups. (A) Control, (B) Extract Treatment, (C) SO_2 group and (D) SO_2+ Extract group. Slides were stained with H& E staining and examined under 10X magnification.

Concerning brain histology, all groups showed normal architecture, indicating no effect of SO_2 on the brain tissue in this study.

4. Discussion

SO_2 is one of the prominent atmospheric air pollutants in highly polluted regions resulting from diverse industrial activity. It is recognized to be linked with numerous health issues in exposed populations [1, 4]. Chamomile has been extensively utilized as a natural medicine owing to its abundant flavonoid content (antioxidants) [9]. This study has assessed the impact of chamomile extract in alleviating the detrimental effects of SO_2 exposure.

The count of red blood cells and hemoglobin concentrations are essential metrics of erythrocyte viability and oxygen-carrying capacity. The findings of this study indicate that RBC counts and hemoglobin concentrations were consistent throughout the groups. The findings suggest that SO_2 exposure did not markedly affect the examined erythrocyte parameters; however, a cross-sectional study conducted over 1 to 2 years revealed a statistically significant reduction in both RBC count and hematocrit in the SO_2 group, indicating that prolonged exposure may result in anemia and diminished hemoglobin levels. The short duration of the study may account for the differences in the results [17].

SO_2 exposure resulted in elevated levels of HCT and MCV. This phenomenon can be attributed to the malabsorption of vitamin B12 and folate in the intestines, resulting from oxidative stress-induced damage to the intestinal lining [18]. Deficiencies in vitamin B12 and folate are marked by the presence of macrocytic cells [19]. Oxidative stress can induce protein damage Patlolla, et al. [20] potentially

Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4: 3146-3158, 2025
DOI: 10.55214/25768484.v9i4.6745

impairing the function of various enzymes, including methionine synthase and dihydrofolate reductase, which are crucial for the metabolism of Vitamin B12 and Folate [21, 22].

Following the administration of chamomile extract, the platelet count was dramatically decreased in comparison to the control group. Prior research has shown that chamomile plant extract possesses anti-platelet effects. It inhibits the activation and aggregation of platelets [23]. Despite this, previous studies did not discover any harmful effects of chamomile extract [10]. Therefore, additional research is required to ascertain whether the low platelet count is attributable to the extract's possible toxicity at the examined dosage.

The SO_2 group and the SO_2 + chamomile extract treatment group exhibited a significant reduction in platelet count. This may be elucidated by the capacity of SO_2 and its derivatives to induce DNA methylation in platelets [24]. Furthermore, oxidative stress elicited by SO_2 can activate caspases and precipitate mitochondrial dysfunction in platelets, as it has been previously demonstrated to induce apoptosis in alveolar cells [25]. Additional research is required to establish the connection.

Serum alanine aminotransferase and aspartate aminotransferase levels have long been utilized as measures of hepatic health, with elevated levels signifying liver disease [26]. This study demonstrated that exposure to SO_2 resulted in a statistically significant elevation of both ALT and AST levels compared to the control group, indicating its detrimental impact, which was alleviated by chamomile treatment. This study's findings are consistent with prior studies on the hepatotoxic effects of SO_2, indicating that sodium sulfite, a product of SO_2, induced apoptosis in hepatocytes Liu, et al. [27]. Meng, et al. [18] proposed that exposure to sulfur dioxide derivatives, specifically sodium sulfite and sodium bisulfite, resulted in DNA damage across multiple tissues, including liver tissue. Our investigation indicates that chamomile extract prophylactic therapy is promising in alleviating the detrimental effects of SO_2 on hepatocytes. This aligns with other studies indicating the hepatoprotective properties of chamomile extract against carcinogenesis produced by 1,2-Dimethylhydrazine in murine models [28]. The hepatoprotective effects of chamomile extract are probably due to its recognized antioxidant and anti-inflammatory characteristics. Research indicates that chamomile extract can diminish oxidative stress by neutralizing free radicals and altering the function of antioxidant enzymes [29]. Furthermore, chamomile's anti-inflammatory properties may protect the liver from injury by inhibiting leukocyte infiltration and the production of pro-inflammatory cytokines [30].

This study evaluated the impact of SO_2 on renal function by measuring the markers of kidney function, specifically creatinine, BUN, and urea levels [31]. Creatinine levels remained unaffected by SO_2 exposure or chamomile extract. A prior study revealed conflicting findings, demonstrating that SO_2 exposure resulted in a diminished glomerular filtration rate relative to unexposed individuals, suggesting renal impairment associated with prolonged SO_2 exposure [32]. This implies that an extended duration is required for the effects on creatinine levels to become evident.

Chamomile extract treatment, in comparison to the SO_2 group, diminished BUN and urea levels in the SO_2+Extract group. The substantial reduction in urea and blood urea nitrogen (BUN) levels in the chamomile-treated groups may be attributed to the extract's diuretic properties, which enhance urine production and inhibit urea accumulation in the bloodstream, given that urea is a byproduct of amino acid catabolism and is excreted by the kidneys [31, 33]. The results indicate that chamomile extract may enhance kidney function. The limited duration of the trial is believed to have been inadequate for SO_2 exposure to induce nephrotoxicity in mice. Further research is necessary to evaluate the long-term renal impairment.

Exposure to SO_2 led to increased levels of malondialdehyde in the liver, signifying oxidative stress due to lipid peroxidation [34]. This indicates that SO_2 causes oxidative damage in the liver. Chamomile extract may safeguard against this harm. The protective effect of chamomile extract aligns with its established antioxidant properties, as noted in previous research [30].

Moreover, SO_2 therapy markedly impaired the activity of superoxide dismutase and catalase, crucial antioxidant enzymes that neutralize free radicals within cells. Administration of chamomile extract to

Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4: 3146-3158, 2025
DOI: 10.55214/25768484.v9i4.6745

SO_2-exposed mice markedly enhanced SOD and CAT activity; nevertheless, CAT activity remained below compared to that of the control and chamomile groups. This indicates that SO_2 exposure could exceed the liver's inherent antioxidant defense mechanisms by producing free radicals, potentially disrupting antioxidant enzymes and leading to their inactivation [35]. Chamomile extract seems to provide protection by neutralizing free radicals, reducing their harmful effects on antioxidant enzyme inactivation, and enhancing the synthesis of antioxidant enzymes [9]. Additional research is necessary to ascertain whether an elevated dosage of chamomile extract is essential for the complete restoration of antioxidant enzyme levels or if extending the duration of extract administration suffices to normalize enzyme levels.

While SO_2 exposure elevated oxidative stress in the liver, the brain exhibited a distinct response. Levels of MDA, SOD, and CAT in brain homogenate were consistent across all groups, suggesting potential tissue susceptibility to SO_2-induced oxidative stress. This observation may result from the protective function of the blood-brain barrier, which selectively controls the transport of substances from the bloodstream into the brain [36, 37]. Our research indicated that SO_2 exposure inflicted greater harm on the liver compared to the brain. The short duration of our study suggests that neither sulfate nor sulfite ions induced damage on the blood-brain barrier, hence rendering the brain less susceptible to oxidative stress in this study.

5. Conclusion

This study elucidates the adverse impacts of sulfur dioxide (SO_2) exposure on multiple biological parameters, encompassing haematological, hepatic, and renal functions, in addition to oxidative stress indicators. Exposure to SO_2 resulted in substantial changes in haematocrit, mean corpuscular haemoglobin, and platelet count, as well as increased blood indicators of liver damage (AST and ALT). Moreover, SO_2-induced oxidative stress in the liver, indicated by elevated malondialdehyde (MDA) levels and reduced superoxide dismutase (SOD) and catalase (CAT) activities, was mitigated with chamomile extract therapy, highlighting its potential as a protective agent. Histological study validated the preventive benefits of chamomile extract on liver and lung tissues, alleviating the damage caused by SO_2 exposure. However, its influence on brain tissue was negligible, indicating that the brain may exhibit reduced vulnerability to SO_2-induced damage, maybe attributable to the protective function of the blood-brain barrier. Chamomile extract demonstrates considerable antioxidative and hepatoprotective properties, suggesting it as a potential therapeutic strategy to alleviate the detrimental effects of SO_2 exposure.

5.1. Limitations and Recommendations

Additional research is required to validate its long-term efficacy and optimal dosage for complete restoration of affected parameters.

Transparency:

The authors confirm that the manuscript is an honest, accurate, and transparent account of the study; that no vital features of the study have been omitted; and that any discrepancies from the study as planned have been explained. This study followed all ethical practices during writing.

Acknowledgments:

The authors would inform the journal that they acknowledge Beirut Arab University as the institution where the research was conducted.

References

[1] World Health Organization (WHO), *Ambient air pollution: A global assessment of exposure and burden of disease*. World Health Organization, 2016.

[2] R. Shapiro, "Sulfite ion and its chemistry," *Environmental Health Perspectives*, vol. 73, no. 1, pp. 99-105, 1977. https://doi.org/10.1289/ehp.737301

[3] World Health Organization, "Air quality guidelines for particulate matter, ozone, nitrogen dioxide and sulfur dioxide: Global update 2005: Summary of risk assessment," Retrieved: https://www.who.int/news-room/fact-sheets/detail/ambient-(outdoor)-air-quality-and-health, 2005.

[4] United States Environmental Protection Agency, "Sulfur dioxide (SO_2) pollution," Retrieved: https://www.epa.gov/so2-pollution, n.d.

[5] H. Vally and N. L. Misso, "Adverse reactions to the sulphite additives," *Gastroenterology Hepatology Bed Bench*, vol. 5, no. 1, pp. 16-23, 2012.

[6] E. M. Khalaf, M. J. Mohammadi, S. Sulistiyani, A. A. Ramírez-Coronel, F. Kiani, and A. T. Jalil, "Effects of sulfur dioxide inhalation on human health: A review," *Reviews in Environmental Health*, vol. 39, no. 2, pp. 331-337, 2022. https://doi.org/10.1515/reveh-2022-0237

[7] S. L. Taylor, R. K. Bush, and J. C. Selner, "Sensitivity to sulfited foods among sulfite-sensitive subjects with asthma," *Journal of Allergy and Clinical Immunology*, vol. 69, no. 5, pp. 335-338, 1982.

[8] Agency for Toxic Substances and Disease Registry, "Toxicological profile for sulfur dioxide," Retrieved: https://www.atsdr.cdc.gov/toxprofiles/tp116.pdf, 2024.

[9] J. K. Srivastava, E. Shankar, and S. Gupta, "Chamomile: A herbal medicine of the past with a bright future," *Molecular Medicine Reports*, vol. 3, no. 6, pp. 895-901, 2010. https://doi.org/10.3892/mmr.2010.377

[10] A. Ishizaki, A. Miura, and H. Kataoka, "Determination of luteolin and apigenin in herbal teas by online in-tube solid-phase microextraction coupled with LC-MS/MS," *Foods (Basel, Switzerland)*, vol. 13, no. 11, p. 1687, 2024. https://doi.org/10.3390/foods13111687

[11] A. I. Drif, R. Yücer, R. Damiescu, N. T. Ali, T. H. Abu Hagar, and B. Avula, "Anti-inflammatory and cancer-preventive potential of chamomile (Matricaria chamomilla L.): A comprehensive in silico and in vitro study," *Biomedicines*, vol. 12, no. 7, p. 1484, 2024. https://doi.org/10.3390/biomedicines12071484

[12] M. Bou Dagher-Kharrat, H. El Zein, and G. Rouhan, "Setting conservation priorities for Lebanese flora—Identification of important plant areas," *Journal of Natural Conservation*, vol. 43, pp. 85-94, 2018. https://doi.org/10.1016/j.jnc.2017.11.004

[13] M. M. Bradford, "A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding," *Analytical Biochemistry*, vol. 72, pp. 248-254, 1976. https://doi.org/10.1016/0003-2697(76)90527-3

[14] C. C. Winterbourn, R. E. Hawkins, M. Brian, and R. W. Carrell, "The estimation of red cell superoxide dismutase activity," *Journal of Laboratory and Clinical Medicine*, vol. 85, no. 2, pp. 337-341, 1975.

[15] H. Aebi, "Catalase in vitro," *Methods in Enzymology*, vol. 105, pp. 121-126, 1984. https://doi.org/10.1016/S0076-6879(84)05016-3

[16] H. Ohkawa, N. Ohishi, and K. Yagi, "Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction," *Analytical Biochemistry*, vol. 95, no. 2, pp. 351-358, 1979. https://doi.org/10.1016/0003-2697(79)90738-3

[17] J. Hwang and H. J. Kim, "Association of ambient air pollution with hemoglobin levels and anemia in the general population of Korean adults," *BMC Public Health*, vol. 24, p. 988, 2024. https://doi.org/10.1186/s12889-024-18492-z

[18] Z. Meng, G. Qin, B. Zhang, and J. Bai, "DNA damaging effects of sulfur dioxide derivatives in cells from various organs of mice," *Mutagenesis*, vol. 19, no. 6, pp. 465-468, 2004. https://doi.org/10.1093/mutage/geh058

[19] O. Stanger, B. Fowler, K. Piertzik, M. Huemer, E. Haschke-Becher, and A. Semmler, "Homocysteine, folate, and vitamin B12 in neuropsychiatric diseases: Review and treatment recommendations," *Expert Review of Neurotherapeutics*, vol. 9, no. 9, pp. 1393-1412, 2009. https://doi.org/10.1586/ern.09.75

[20] A. K. Patlolla, C. Barnes, C. Yedjou, V. R. Velma, and P. B. Tchounwou, "Oxidative stress, DNA damage, and antioxidant enzyme activity induced by hexavalent chromium in Sprague-Dawley rats," *Environmental Toxicology*, vol. 24, no. 1, pp. 66-73, 2009. https://doi.org/10.1002/tox.20395

[21] J. I. Toohey, "Vitamin B12 and methionine synthesis: A critical review," *Is nature's most beautiful cofactor misunderstood? BioFactors*, vol. 26, no. 1, pp. 45-57, 2006. https://doi.org/10.1002/biof.5520260105

[22] R. Green and A. Datta Mitra, "Megaloblastic anemias: Nutritional and other causes," *Medical Clinics of North America*, vol. 101, no. 2, pp. 297-317, 2017. https://doi.org/10.1016/j.mcna.2016.09.013

[23] M. Bijak, J. Saluk, M. Tsirigotis-Maniecka, H. Komorowska, B. Wachowicz, and E. Zaczyńska, "The influence of conjugates isolated from Matricaria chamomilla L. on platelet activity and cytotoxicity," *International Journal of Biological Macromolecules*, vol. 61, pp. 218-229, 2013. https://doi.org/10.1016/j.ijbiomac.2013.06.046

[24] H. Sun, Y. Li, J. Zhang, T. Shi, X. Li, and X. Cao, "Platelet mitochondrial DNA methylation as an epigenetic biomarker of short-term air pollution exposure in healthy subjects," *Frontiers in Molecular Biosciences*, vol. 8, p. 803488, 2022. https://doi.org/10.3389/fmolb.2021.803488

[25] J. Bai and Z. Meng, "Effects of sulfur dioxide on apoptosis-related gene expressions in lungs from rats," *Regulatory Toxicology and Pharmacology*, vol. 43, no. 3, pp. 272-279, 2005. https://doi.org/10.1016/j.yrtph.2005.09.002

[26] T. H. Lee, W. R. Kim, and J. J. Poterucha, "Evaluation of elevated liver enzymes," *Clinical Liver Disease*, vol. 16, no. 2, pp. 183-198, 2012. https://doi.org/10.1016/j.cld.2012.03.006

[27] M. Liu, J. Lu, J. Hu, Y. Chen, X. Deng, and J. Wang, "Sodium sulfite triggered hepatic apoptosis, necroptosis, and pyroptosis by inducing mitochondrial damage in mice and AML-12 cells," *Journal of Hazardous Materials* vol. 467, p. 133719, 2024. https://doi.org/10.1016/j.jhazmat.2024.133719

[28] S. Shebbo, M. El Joumaa, R. Kawach, and J. Borjac, "Hepatoprotective effect of Matricaria chamomilla aqueous extract against 1,2-dimethylhydrazine-induced carcinogenic hepatic damage in mice," *Heliyon*, vol. 6, no. 6, p. e04082, 2020. https://doi.org/10.1016/j.heliyon.2020.e04082

[29] A. El Mihyaoui, J. C. G. Esteves da Silva, S. Charfi, M. E. Candela Castillo, A. Lamarti, and M. B. Arnao, "Chamomile Matricaria chamomilla L.): A review of ethnomedicinal use, phytochemistry, and pharmacological uses," *Life Basel, Switzerland*, vol. 12, no. 4, p. 479, 2022. https://doi.org/10.3390/life12040479

[30] B. Al-Dabbagh, I. A. Elhaty, M. Elhaw, C. Murali, A. Al Mansoori, and B. Awad, "Antioxidant and anticancer activities of chamomile (Matricaria recutita L.)," *BMC Research Notes*, vol. 12, no. 1, p. 3, 2019. https://doi.org/10.1186/s13104-018-3960-y

[31] M. A. Ferguson and S. S. Waikar, "Established and emerging markers of kidney function," *Clinical Chemistry*, vol. 58, no. 4, pp. 680-689, 2012. https://doi.org/10.1373/clinchem.2011.167494

[32] F. Wen, Y. Xie, B. Li, P. Li, H. Qi, and F. Zhang, "Combined effects of ambient air pollution and PM2.5 components on renal function and the potential mediation effects of metabolic risk factors in China," *Ecotoxicology and Environmental Safety*, vol. 259, p. 115039, 2023. https://doi.org/10.1016/j.ecoenv.2023.115039

[33] A. Sah, P. P. Naseef, M. S. Kuruniyan, G. K. Jain, F. Zakir, and G. Aggarwal, "A comprehensive study of therapeutic applications of chamomile," *Pharmaceuticals (Basel, Switzerland)*, vol. 15, no. 10, p. 1284, 2022. https://doi.org/10.3390/ph15101284

[34] D. Del Rio, A. J. Stewart, and N. Pellegrini, "A review of recent studies on malondialdehyde as a toxic molecule and biological marker of oxidative stress," *Nutrition, Metabolism, and Cardiovascular Diseases*, vol. 15, no. 4, pp. 316-328, 2005. https://doi.org/10.1016/j.numecd.2005.05.003

[35] J. M. Matés, "Effects of antioxidant enzymes in the molecular control of reactive oxygen species toxicity," *Toxicology*, vol. 153, no. 1-3, pp. 83-104, 2000. https://doi.org/10.1016/s0300-483x(00)00306-1

[36] E. R. Viscusi and A. R. Viscusi, "Blood-brain barrier: Mechanisms governing permeability and interaction with peripherally acting μ-opioid receptor antagonists," *Regional Anesthesia and Pain Medicine*, vol. 45, no. 9, pp. 688-695, 2020. https://doi.org/10.1136/rapm-2020-101403

[37] M. E. M. Stamp, M. Halwes, D. Nisbet, and D. J. Collins, "Breaking barriers: Exploring mechanisms behind opening the blood-brain barrier," *Fluids and Barriers of the CNS*, vol. 20, no. 1, p. 87, 2023. https://doi.org/10.1186/s12987-023-00489-7

Edelweiss Applied Science and Technology
ISSN: 2576-8484
Vol. 9, No. 4: 3146-3158, 2025
DOI: 10.55214/25768484.v9i4.6745