Research Article

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Insights about the deleterious impact of a carbamate pesticide on some metabolic immune and antioxidant functions and a focus on the protective ability of a Saharan shrub and its anti-edematous property

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Abstract: In the Algerian desert, individuals often enjoy a soothing cup of herbal tea made from *Ephedra alata* Decne before bedtime; this aids in their relaxation. Our previous and earlier investigations demonstrated the richness of the shrub in terms of polyphenols and flavonoids and their effective medicinal properties. In the light of that fact, our interest has been aroused to check for other types of metabolites and for the protective ability of the shrub crude extract (SCE) regarding chemically induced edema and subacute toxicity (following a formalin-induced paw edema model and using a carbamate pesticide "pirimicarb" as a toxic agent, in Wistar male rats, respectively). Evidently, the SCE was used as a preventive agent. Swelling of formalin-injected foot was measured, and the anti-edematous ability was expressed as a percent of paw edema. At the end of induced subacute toxicity procedure, many investigations were carried, namely, checking for biochemical several parameters (hepatic, renal), hematological parameters, oxidative stress status (OSS), and histological examining of liver, spleen, and kidney tissues. The results revealed a remarkable anti-edematous effect. Furthermore, the association of the SCE effect has clearly minimized the OSS, the tissue aberrations, and the disturbance of the other metabolic parameters, along with a reduced immunodepression that was provoked under the effect of pirimicarb. In conclusion, pirimicarb has an important deleterious impact on metabolic and immune functions, and the studied shrub has exhibited an amazing protective potential against chemically induced inflammation and toxicity.

Keywords: hepato-nephrotoxicity, immunodepression, inflammation, oxidative stress, pirimicarb, phytochemical screening, rat Wistar, shrub

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1 Introduction

Bioactive compounds, encompassing both essential and nonessential elements such as vitamins and polyphenols, are naturally occurring substances integral to the food chain, demonstrating significant impacts on human health. Coined as nutraceuticals by Stephan DeFelice in 1979, this term underscores their presence in human dietary intake and their biological virtues. In the past decade, our understanding of the role of phytochemicals, like polyphenols, in specific pathologies has rapidly advanced. Various classes of phytochemicals, such as phytoestrogens, have been

identified for their preventive effects on specific diseases, particularly in the early stages of development, notably in relation to cancer. The prevalence of these compounds in vegetables aligns with epidemiological evidence suggesting that elevated vegetable consumption is associated with a reduced risk of various types of cancer [1]. Free radicals naturally accumulate as byproducts in metabolic pathways, causing oxidative stress within the body. External sources, including pollution, cigarette smoke, radiation, and certain medications, can also contribute to this stress, resulting in cellular damage. Oxidative stress (OS) is implicated in the onset of enduring and serious health conditions, for instance, cancer, senescent ailments, autoimmune disorders, ocular opacity, rheumatoid arthritis, neurological disorders, and heart-related issues. Fortunately, antioxidants could play the role of regulators toward this mechanism, which are either naturally produced *in situ* or supplied externally through foods and herbal supplements. Elevated amounts of antioxidants correlate with increased resilience against a range of pathologies, highlighting their crucial role in maintaining overall health [2]. Ephedraceae belong among the earliest known plants with medicinal properties, encompassing a total of 69 species. These species are predominantly found in semi-arid regions across the Palearctic and Nearctic areas. They are thriving in subtropical and temperate territories spanning China, Bhutan, India, Afghanistan, Pakistan, North America, and North Africa. Members of the Ephedra genus are characterized by their evergreen nature and small perennial shrubs, which exhibit resistance to frost and aridity. The diverse range of species from Ephedra highlights their adaptability to various environmental conditions, contributing to their historical significance in traditional medicine and their continued relevance in contemporary contexts [3,4]. Ephedra, also known as mahuang, is a Chinese shrub with a rich history dating back at least 5,000 years. In the late sixteenth century, Li Shih-Chen documented its properties in the renowned pharmacopeia, the Pents'ao Kang Mu. Ephedra was recognized for its utility as a circulatory stimulant, sudorific, and fever-reducer. Additionally, it has a value in treating coughs, leading to its incorporation into various cough suppressant preparations. Toward the end of the sixteenth century, ephedra stems found their way to Japan through commerce. This trade played a pivotal role in sparking interest among Japanese physicians and chemists, laying the groundwork for their engagement with the plant three centuries later [5]. According to the Chinese Pharmacopoeia 15th edition, *Ephedrae Herba* is described as the stem of *Ephedra intermedia* Schrenket C. A. Meyer, *Ephedra equisetina* Bunge, or *Ephedra sinica* Stapf. It has traditional applications in addressing rheum, bronchial asthma, delirium, cough, flu, migraine, edema, and hypersensitivity. Notably, it serves as a natural source for compounds like ephedrine and pseudoephedrine. In some Western countries, it is employed as a dietary supplement, aiding in weight loss by promoting sweating, increasing basal metabolism rate, and boosting the function of the brain and the spinal cord [6].

Carbamate compounds, esters of carbamic acid commonly known as *N*-methylcarbamates, serve as insecticides. When used appropriately, carbamate pesticides play a vital role in protecting and enhancing agricultural production and guarding against insect-borne diseases that affect human and animal health. However, exposing excessively to these pesticides could have serious consequences, leading to poisoning incidents. The toxicity of *N*-methylcarbamate insecticides stems from their ability to inhibit the acetylcholinesterase enzyme, resulting in elevated cholinergic activity and toxic manifestations. Additionally, the carbamate could induce excitotoxicity that involves the overstimulation of *N*-methyl-D-aspartate receptors [7]. Pirimicarb, known chemically as 2-dimethylamino-5,6-dimethylpyrimidin-4-yldimethylcarbamate, finds application in agriculture as an insecticide. In mammals, its catabolism includes the breakdown of the carbamate part through hydrolysis, followed by demethylation at the dimethylamino group linked to the heterocyclic part. This process yields major metabolites excreted in the urine, namely, 2-dimethylamino-5,6-dimethyl-4-hydroxypyrimidine, 2-methylamino-5,6-dimethyl-4-hydroxypyrimidine, and 2-amino-5,6-dimethyl-4-hydroxypyrimidine. These compounds were consistently found in micturition samples collected from a group of farmers who had been involved in the application of pirimicarb [8]. In the Algerian desert, individuals often enjoy a soothing cup of herbal tea made from *Ephedra alata Decne* before bedtime to promote relaxation. Our previous investigations have highlighted this shrub plant as a potent and promising natural source of abundant metabolites (polyphenols and flavonoids); these latter showed impressive *in vitro* antioxidant and anti-inflammatory potentials [9]. In addition to its fertilizing potentials that enhanced secretion of androgenic hormones, spermatogenesis, and euphoric ability that dealt with stress prevent anxiety and depression provoked under the effects of pirimicarb [10]. Therefore, we have planned to reveal the presence of other metabolites and estimate the ability of the plant to prevent inflammation, proceeding by an *in vivo* approach. Second, we aimed to assess the ability of the plant to protect itself from toxicological events provoked by pirimicarb interfering with the function of liver, kidney, and immune tissues. The selection of these target tissues was determined by two main factors; Firstly, we chose tissues known for their vital roles in detoxification and immune response. Secondly, this investigation serves as a natural extension of our prior research, which focused on studying the brain and testis [10].

2 Materials and methods

2.1 Requirements in chemical products

The list of chemicals used is provided in Table 1.

2.2 Shrub crude extract (SCE) preparation

The stems, leaves, and flowers of the shrub "*ephedra alata* Decne" were collected and prepared as a crude extract, as described in our previous works [9,11].

2.3 Phytochemical screening

The presence of various metabolites in the SCE solution was examined through specific chemical tests. For alkaloids, 1,000 µL of SCE solution was reacted with 1 mL of Mayer's reagent, and the development of a soft yellow color showed the presence of alkaloids [12]. In regards to tannins, 1,000 µL of $FeCl_3$ (1%) was reacted with 1,000 µL of SCE solution; then, the formation of a greenish or blackish-blue color confirmed the presence of tannins [13]. Concerning saponins, 200 µL of the extract solution was mixed with 5 mL of deionized water and stirred rapidly for a period of 5 min. The formation of foam indicated a positive result [14]. Regarding coumarins, their presence is revealed by an intense fluorescence under UV at 365 nm after reacting 1,000 µL of the SCE solution with 500 µL of NH_4OH (25%) [15]. To determine the presence of steroids and terpenoids, we have followed the tests described by Pathak and Shrivastav [16]. Finally, carbohydrates were detected according to Katoch [17].

Table 1: Employed chemical products

Products	Chemical formula/ abbreviation	Brand
5,5-Dithio-bis-(2-nitrobenzoic acid)	DTNB	Sigma Aldrich
Ammonium hydroxide	NH_4OH	
Bradford reagent		
Bovine serum albumin	BSA	
Certistain		Merck
Disodium phosphate disodium phosphate	Na_2HPO_4	Sigma Aldrich
Ethanol 96%		PanReac AppliChem
Ethylene diamine tetraacetic acid	EDTA	Sigma Aldrich
Formaldehyde 37–38%	/	PanReac AppliChem
Formic acid 98–100%	/	Sigma Aldrich
Hydrogen chloride	HCl	
Hydrogen peroxide 30%	H_2O_2	
Iron chloride	$FeCl_3$	
Mayer's hematoxylin	/	Specilab
Monosodium phosphate	NaH_2PO_4	Sigma Aldrich
Neoxylene	/	Eukitt.
Pirimor 50 DG	/	Syngenta
Potassium phosphate monobasic	KH_2PO_4	Sigma Aldrich
Pyrogallol	/	
Sodium chloride	NaCl	
Thiobarbituric acid	TBA	
Trichloroacetic acid	TCA	
Trizma	Tris	
Xylen	/	PanReac AppliChem

3 Anti-Paw edema

3.1 Animals

Thirty-five male albino Wistar rats, obtained from Algiers Pasteur Institute (IPA), weighing between 210 and 235 g, underwent a 15-day acclimatization period before starting the experimental procedure. The rats were maintained in conditions of 22 ± 2°C temperature and 50–60% humidity, following a 12-h light/dark cycle. In the UMC1 animal laboratory, they had unrestricted access to clean tap water and commercial pelleted feed (provided by "ONAB" Guelma, Algeria).

3.2 Experiment procedure

Five groups (n = 5) were randomly selected from among the rats. Sodium diclofenac, a reference anti-inflammatory drug, was administered intraperitoneally to the second group, which served as a positive control. The third, fourth, and fifth groups were given varying doses of sodium codafat (25, 200, and 400 mg/kg bw, respectively). The first group, acting as a negative control, was given normal saline (5 mL/kg bw). The rat was given a subplantar injection of 0.1 mL of 1% formalin 30 min after the drug was administered to cause edema on the right hind paw. A water displacement plethysmometer was used to measure the swelling of the formalin-injected foot hourly from the first to the fifth hour following the injection [18,19]. The following formula was used to express the anti-edematous effect's capacity to reduce paw inflammation as a percentage of paw edema:

$$\text{Edema\%} = \frac{\text{VF} - \text{VI}}{\text{VI}} \times 100, \quad (1)$$

where VI is the intimal volume of edema (before injection of formalin) and VF is the final volume of edema (after injection of formalin).

4 Pesticide induced toxicity

4.1 Animals

Twenty-four male albino Wistar rats, ranging in weight from 200 to 225 g, were procured from IPA. Before the initiation of the experiment, the rats underwent 15 days as acclimatization period. They were kept in a 12-h light/dark cycle with a temperature of 22 ± 2°C and a humidity level of 50–60%. The rats at the CRBt animal laboratory had unlimited access to clean tap water and commercial pelleted feed (provided by "ONAB" Guelma, Algeria).

4.2 Experiment procedure

To induce pesticide toxicity, we followed the protocol that we designed in our previous work [10]. The study involved the categorization of animals into four groups (Gs), each consisting of six rats. These groups were subjected to distinct oral gavage treatments as follows: C received deionized water, SCE received 200 mg/kg of SCE, P 15 mg of pirimicarb, and P + SCE received simultaneously treatments of SCE and P (the SCE administered 1 h before pirimicarb). These daily doses were administered over a 28-day period. Afterward, euthanasia was performed via cervical dislocation subsequent to anesthesia. Organs (liver, spleen, and kidney) as well as blood were collected for further investigations. All procedures adhered to laboratory guidelines for animal care and were reviewed and approved by the institutional ethic committee of the CRBt (ethical approval reference: N07KH-2021/2023/CCE).

4.3 Food and water intake

The intake of food and water of each experimental group was tracked daily, the weakly consumption was recorded, and the variation was compared between groups.

4.4 Weight gain (WG) and relative weight organ (RWO)

The weight of rats was checked daily during the period of the experiment, and the WG was assessed at the end and expressed by the variation among the fourth groups. Similarly, the weights of the studied organs (liver, spleen, and kidney) were taken after sacrifice in order to estimate the RWO of each of them.

5 Hepatic and renal biochemical parameters

5.1 Hepatic

To check the liver biochemical function, we analyzed from serum samples three parameters: aspartate and alanine transaminases (AST and ALT) and alkaline phosphatase (ALP), using COBAS INTEGRA 400 plus and a specific kit of each parameter.

5.2 Kidney

To check the kidney biochemical function, we analyzed from serum samples two parameters: urea and creatinine, using COBAS INTEGRA 400 plus and a specific kit of each parameter.

6 Oxidative stress status (OSS) evaluation

6.1 Preparation of tissue homogenate

The tissues of liver and kidney were processed according to the method described in our previous investigation [10].

6.2 Quantification of protein in tissue homogenate

Protein quantification in tissue homogenates implicated the spectrophotometric method of Bradford [20]. BSA was serving as the standard for calibration.

6.3 Malondialdehyde (MDA)

The assessment of lipid peroxidation (LPO) titers was achieved by measuring the concentration of MDA in tissue homogenates. MDA interacts with TBA, forming a red-colored complex as a reactive substance. The procedure entailed combining 0.5 mL of tissue homogenate with 1 mL of TCA–TBA–HCl solution (15%, 0.375%, 0.25 N) and thorough mixing. The resulting mixture was then subjected to a 15-min heating process in a boiling water bath. Subsequently, the formed flocculent precipitate was eliminated through centrifugation at 1,000*g* for 10 min, and the absorbance was assessed at 535 nm [21]. The MDA concentration is quantified in nmol/mg of protein and determined using the following equation:

$$\text{MDA(nmol mg protein)} = \frac{\text{OD} \times 10^6}{E \times X \times L \times \text{Fd}}, \quad (2)$$

where OD is the optical density read at 530 nm, E is the molar extinction coefficient of MDA = $1.56 \times 10^5\ \text{M}^{-1}\,\text{cm}^{-1}$, L is the optical path length, Fd is the dilution factor = 0.2083 and X is the protein concentration of the extract (mG/mL).

6.4 Reduced glutathione (GSH)

GSH levels in tissue homogenates were assessed using a colorimetric technique based on the oxidation of GSH by DTNB, yielding a yellow color following the Elman method [22]. In this process, 800 µL of tissue homogenate was combined with 100 µL of sulfosalicylic acid (0.25%) and allowed to stand for 15 min in an ice bath. Following centrifugation at 1,000 rpm for 15 min, 500 µL of the supernatant was collected and mixed with 1 mL of tris–EDTA buffer (0.4 M HCl, 0.02 M EDTA, pH 9.6) and 25 µL of DTNB (0.01 M). After shaking and a 5-min incubation period, the absorbance was recorded at 412 nm. The concentration of GSH is expressed in micromoles per milligram of protein (µmol/mg protein) and calculated using the following equation:

$$\text{GSH(µmol/mg protein)} = \frac{\text{OD} \times L \times 1.525}{13.1 \times 0.8 \times 0.5 \times \text{mg protein}}, \quad (3)$$

where OD is the optical density, L is the optical path length, 1.525 is the total volume reaction mixture used (0.5 mL supernatant + 1 mL Tris–EDTA, 0.025 mL DTNB), 13,100 is the molar extinction coefficient of GSH ($\text{M}^{-1}\,\text{cm}^{-1}$) at 412 nm, and 0.8 is the volume of tissue homogenate.

6.5 Catalase (CAT)

CAT activity was assessed using the procedure developed by Aebi [23]. In summary, 983.5 µL of H_2O_2 (10 mM) in a 50 mM phosphate buffer (KH_2PO_4, Na_2HPO_4) with a pH of 7.2 was reacted with 16.5 µL of tissue homogenate. The reaction relied on the reduction of H_2O_2, and the absorbance decline was observed for 30 s at 240 nm

$$\text{CAT(µmol } H_2O_2\text{/min/mg protein)} = \frac{\Delta\text{OD/min}}{\in \times L \times n}, \quad (4)$$

where ε is the molar extinction coefficient of H_2O_2: 43.6 $\text{M}^{-1}\,\text{cm}^{-1}$, L is the optical path length, and n is the mg of protein present in the sample used volume.

6.6 Hematological parameters

The analysis of blood parameters was conducted using an automated hematology analyzer (Mindray BC-3000 Plus). Rat blood samples were collected in EDTA tubes, with consideration given to the following parameters: white blood cells, erythrocytes, lymphocytes, monocytes, granulocytes, and platelets.

6.7 Histological analysis

No deaths related to the treatment were observed. Rats that were killed underwent a comprehensive necropsy examination. Following a thorough rinse with a NaCl (0.9%) solution, organs were removed and scrutinized for any apparent lesions. Subsequently, they were promptly preserved in a 10% formaldehyde solution. Using an automated tissue processor, tissue samples were processed following standard procedures. The tissues were processed, sectioned, and stained with hematoxylin and eosin (H&E) before being embedded in paraffin. Photos of particular lesions were taken with an optical microscope that had a built-in camera Nikon TS2-S-SM inverted microscope at 100×.

6.8 Statistical analysis

The ANOVA One-Factor and Tukey post hoc tests were used to establish whether there were significant differences between experimental groups concerning food and water intake, liver and kidney parameters, hematological parameters, and anti-

Table 2: Phytochemical screening outcomes

	Alkaloids	Coumarins	Tannins	Saponins	Steroids	Terpenoids	Carbohydrates
SCE	+	–	+	+	+	+	+

(–) absence, (+) presence.

edematous effect of SCE. The significance rate was set at 5% ($P < 0.05$). The statistical assessments were done on IBM SPSS Statistics 23.0 software (IBM SPSS Inc.).

7 Results

7.1 Phytochemical screening

The followed tests have disclosed the countenance of SCE of many substances belonging to alkaloids, saponins, tannins, steroids, terpenoids, and carbohydrates, as shown in Table 2.

7.2 Anti-edematous effect of SCE

The anti-edematous effect was expressed as a percentage of edema. The administration of SCE in parallel with provoked paw edema reduced eminently the percentage of edema comparatively to the diclofenac effect; the effect of SCE before 4H (240 min) was better than the diclofenac effect at 200 and 400 mg/kg, as shown in Figure 1.

8 Pesticide-induced toxicity

8.1 Food and water intake

The daily tracking of the food and water intake of rats per each group is illustrated in Figure 2. Both part labels (Figure 2a and b) showed that G3 has recorded the most important consumption either for food or for water throughout the whole period of treatment.

8.2 Body WG and relative organ weight

The recording of body weight from the beginning of the experiment until its end allowed us to fellow its change

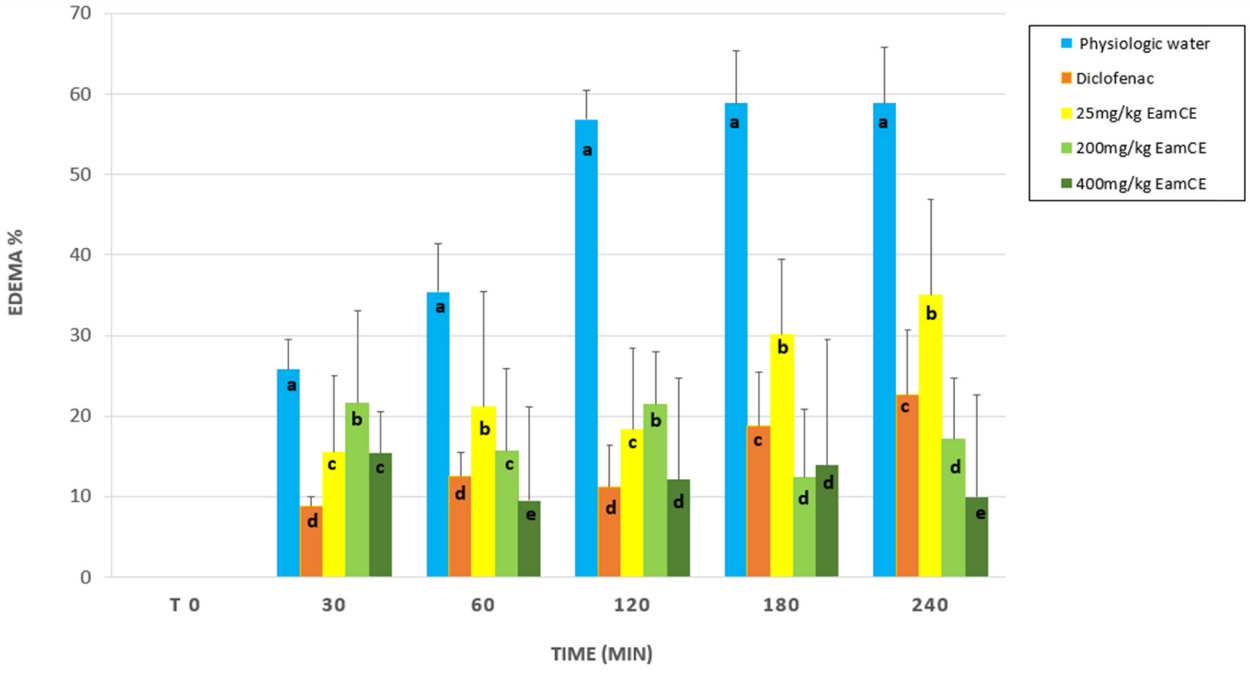

Figure 1: Anti-edematous effect of SCE. Analysis of variance (ANOVA one-factor and Tukey post-hoc tests) revealed statistical difference ($P < 0.05$). Different superscripts (a, b, c, d, and e) for the values in the same time point are statistically different.

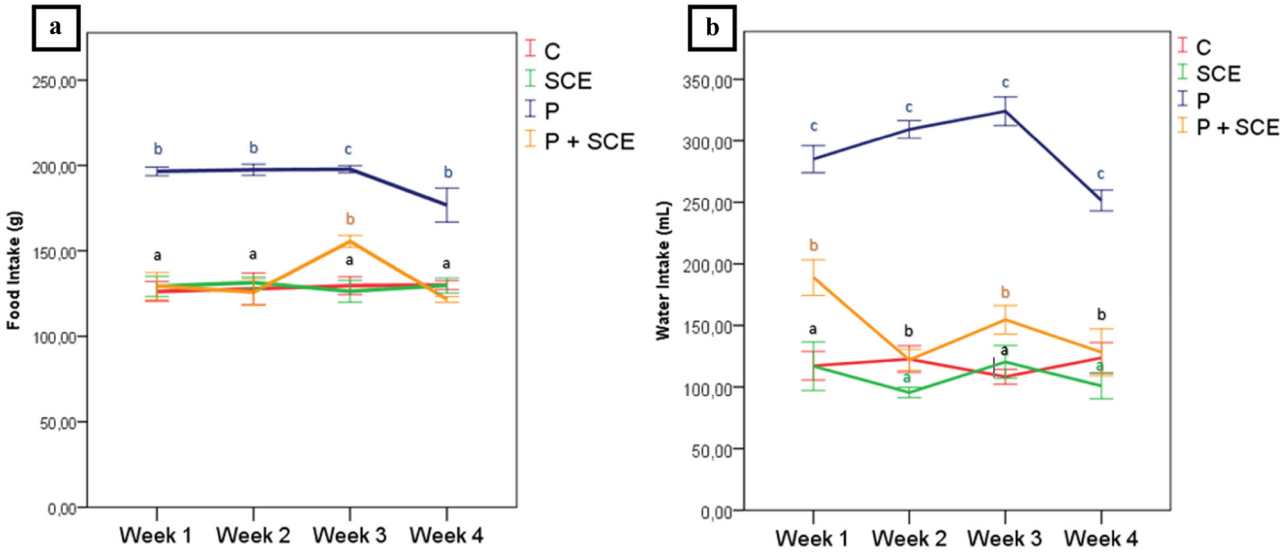

Figure 2: Food and water consumption during the 4 weeks of the experiment. (a) The fluctuation of food intake for all the experimental groups and during the 4 weeks of treatment. (b) The fluctuation of water intake for all the experimental groups and during the 4 weeks of treatment. Analysis of variance (ANOVA one-factor and Tukey post-hoc tests) revealed statistical difference ($P < 0.05$). Different superscripts (a, b, and c) for the values in the same time point are statistically different.

and to compare its variance between experimental groups. Besides, it enables us to estimate the loss or the gain of weight in each group and to notice the influence of pirimicarb and SCE. The results showed that all rats have gained weight, and those of group P represented the most elevated value, 79.36 ± 12.03 (g), as shown in Figure 3a. However, the variance of relative organ weight of liver, kidney, and spleen through the four experimental groups was not significant ($P > 0.05$), as shown in Figure 3b.

9 Hepatic and renal biochemical parameters

9.1 Hepatic

The assessment of hepatic biochemical parameters revealed a remarkable increase in terms of ALAT, ASAT, and ALP in rats of "P," accompanied by a moderate elevation in rats of

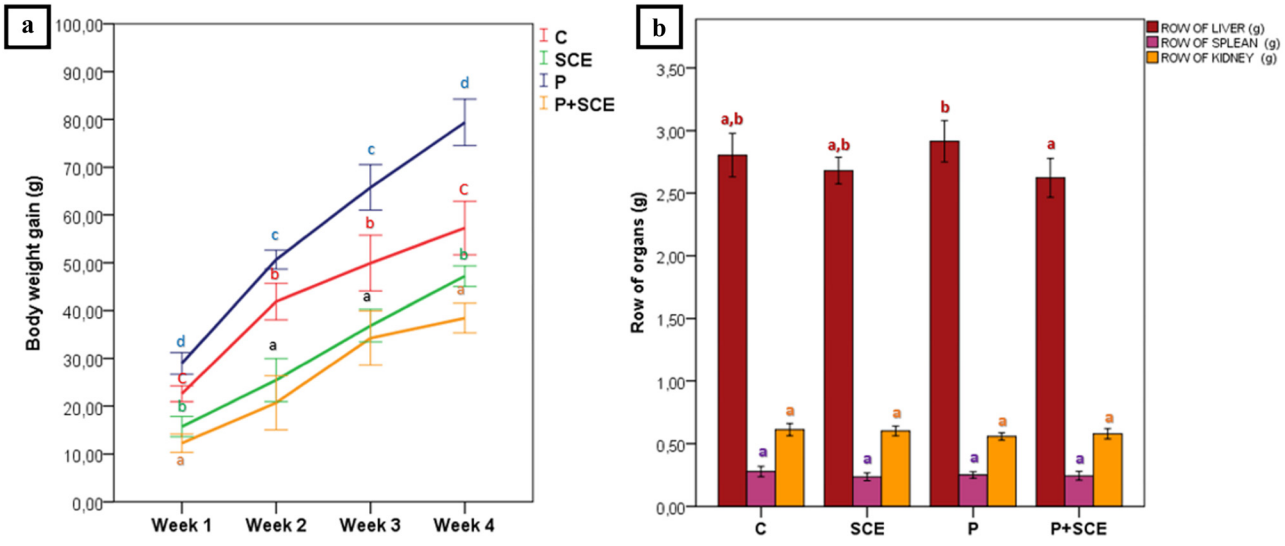

Figure 3: Body weight gain (a) and relative organ weight (b). Analysis of variance (ANOVA one-factor and Tukey post-hoc tests) revealed statistical difference ($P < 0.05$). Different superscripts (a, b, and c) for the values in the same time point are statistically different.

Table 3: Assessment of some hepatic biochemical parameters

	C	SCE	P	P + SCE
ALAT (UI/L)	45.83 ± 3.71[a]	44.35 ± 2.15[a]	57.70 ± 1.27[b]	55.87 ± 1.33[b]
ASAT (UI/L)	110.95 ± 7.09[a]	139.80 ± 0.69[b]	202.01 ± 4.75[c]	151.67 ± 12.69[b]
ALP (UI/L)	144.75 ± 3.37[a]	146.66 ± 2.73[a]	229.00 ± 1.18[c]	167.00 ± 7.77[b]

The outcomes were expressed as mean ± SD ($N = 6$). Analysis of variance (ANOVA one-factor and Tukey post hoc tests) revealed statistical difference ($P < 0.05$). Different superscripts (a, b, and c) for the values in the same lines are statistically different.

"P + SCE" comparatively to reduced values in rats of "C" and "SCE," as well numbered in Table 3.

9.2 Renal

The analysis of urea and creatinine from the serum of all animals gave values that are raised in "P," slightly raised in "P + SCE," and reasonable in "C" and "SCE," for both parameters, as indicated in Table 4.

10 OSS evaluation

10.1 Liver

The antioxidant activity of "P" rats was very enfeebled and characterized by a high level of LPO, where MDA was the most elevated, all with a very low amount of GSH and minimized activity of CAT. In contrast, the liver antioxidant activity of healthy groups was satisfactory, notably for "SCE." These defined parameters were slightly enhanced in "P + SCE" liver, presumably under the effect of SCE (Table 5).

10.2 Kidney

The kidney antioxidant activity of rats from "P" was importantly exhausted. This is well demonstrated by the intensely raised amount of MDA and the reduced expression of GSH and CAT comparatively to the normal kidney antioxidant activity of healthy groups. The effect of pirimicarb was apparently and relatively redressed in "P + SCE" due to the use of SCE combined treatment (Table 6).

10.3 Hematological parameters

Figure 4 shows the changes in the count of various blood elements among different rats of each experimental group. The findings indicated a reduction in lymphocytes and

Table 4: Evaluation of urea and creatinine titers

	C	SCE	P	P + SCE
Creatinine (mg/L)	8.24 ± 0.45[a]	8.13 ± 0.19[a]	9.13 ± 0.44[b]	8.36 ± 0.66[a]
Urea (g/L)	0.338 ± 0.086[a]	0.485 ± 0.193[b]	0.611 ± 0.009[d]	0.547 ± 0.025[c]

The outcomes were expressed as mean ± SD ($N = 6$). Analysis of variance (ANOVA one-factor and Tukey post hoc tests) revealed statistical difference ($P < 0.05$). Different superscripts (a, b, c, and d) for the values in the same lines are statistically different.

Table 5: Liver antioxidant activity

	C	SCE	P	P + SCE
MDA	1.06 ± 0.01[a]	1.45 ± 0.00[b]	6.38 ± 0.00[d]	4.23 ± 0.00[c]
GSH	0.645 ± 0.024[d]	0.564 ± 0.017[c]	0.341 ± 0.017[a]	0.523 ± 0.021[b]
CAT	82.42 ± 0.01[d]	91.56 ± 0.85[c]	27.46 ± 0.39[a]	40.65 ± 0.30[b]
Total protein	1.75 ± 0.00[b]	2.33 ± 0.00[d]	1.98 ± 0.00[c]	1.73 ± 0.00[a]

Measurement units of MDA, GSH, CAT, and total proteins were respectively: (nmol/mg protein), (μmol/mg protein), (μmol H_2O_2/min/mg protein), (mg/mL). The outcomes were expressed as Mean ± SD ($N = 6$). Analysis of variance (ANOVA One-Factor and Tukey post hoc tests) revealed statistical difference ($P < 0.05$). Different superscripts (a, b, c, and d) for the values in the same lines are statistically different.

Table 6: Renal antioxidant activity

	C	SCE	P	P + SCE
MDA	1.67 ± 0.01[a]	1.78 ± 0.00[b]	9.84 ± 0.00[d]	6.06 ± 0.00[c]
GSH	0.561 ± 0.010[c]	0.548 ± 0.020[c]	0.362 ± 0.008[a]	0.387 ± 0.008[b]
CAT	52.47 ± 0.30[d]	51.69 ± 0.41[c]	31.65 ± 0.13[a]	47.48 ± 0.79[b]
Total protein	2.26 ± 0.00[c]	2.24 ± 0.00[b]	1.95 ± 0.00[a]	2.55 ± 0.00[d]

Measurement units of MDA, GSH, CAT, and total proteins were respectively: (nmol/mg protein), (μmol/mg protein), (μmol H_2O_2/min/mg protein), (mg/mL). The outcomes were expressed as Mean ± SD (N = 6). Analysis of variance (ANOVA one-factor and Tukey post-hoc tests) revealed statistical difference (P < 0.05). Different superscripts (a, b, c, and d) for the values in the same lines are statistically different.

granulocytes in rats from "P" compared to the other groups. Conversely, there was a notable increase in monocytes and a significant rise in platelets of rats from "P" when compared to "C,"" SCE," and "P + SCE." Red blood cell counts, however, exhibited no significant variation across the different groups.

10.4 Histological slides

The histological slides corresponding to liver, kidney, and spleen tissues are illustrated in Figures 5, 6 and 7, respectively. Besides, the semiquantitative scoring of liver and kidney microscopic changes in different treatment groups are listed in Tables 7 and 8.

11 Discussion

Inflammation and edema are interconnected physiological responses linked to tissue damage resulting from prolonged mechanical loading [24]. Researchers are constantly searching for new anti-inflammatory compounds without negative effects when treating inflammation. Large amounts of naturally occurring polyphenolic compounds are ingested in daily diets and can have a significant impact on disease treatment [25]. Recent research has shown that Ephedra ciliata's quercetin-rich methanol extract has anti-inflammatory properties that helped wounds heal in two distinct models. Downregulating TNF-α was hypothesized to be the indicator factor of these properties [26]. By blocking the expression of several inflammatory reaction targets, such as SELE, IL-2, and CXCL10, at the mRNA and protein levels, essential compounds of ephedra, such as quercetin, luteolin, kempferol, naringenin, and beta-sitosterol, have been found to be effective in treating asthma. These compounds are engaged in the biological mechanisms of immune reactions, cell signal transduction, and in responding to inflammation provoked by lipopolysaccharide [27]. Indeed, the SCE exhibited an interesting protective response from induced paw edema with the three tested concentrations. Although 200 and 400 mg/kg of SCE were more effective than diclofenac (25 mg/kg), giving a quite reduced edema percentage after 240 min: 17.10 ± 0.61 and 9.91 ± 0.79, respectively (Figure 1). The richness of plants in terms of phenols, flavonoids, saponins, tannins, phytosteroids, quinones, steroids, anthraquinones, triterpenes, glycosides, anthocyanins, cardiac glycosides, and reducing sugars provide them anti-inflammatory properties. *Ephedra alata Decne* appertains to plants with high amount of these metabolites [9,10,28–33]. Indeed, our previous study conducted on the same plant has shown the presence of a considerable amount of polyphenols and flavonoids, with a tentative identification of some of them using LS-MS/MS analysis [9]. Besides, in the current study, we have also revealed the countenance of the plant in terms of different other metabolites (Table 2).

In relation to pesticide-induced toxicity experiment, the findings showed a positive association between food and water consumption and WG assessment. Effectively, the increased consumption of food and water in P, in contrast to the reduced intake in C, SCE, and P + SCE, is linked to a higher WG in P compared to the diminished WG observed in the other groups (Figures 2 and 3). A systematic review has highlighted that exposure to various pesticides, including organophosphate, pyrethroid, organochlorine, carbamate, and neonicotinoid, as well as a combination of pesticides, has direct impacts on body WG and obesity in both humans and experimental animals at different life stages [34]. Similarly, a prior study on the impact of chlorpyrifos pesticide intake in mice revealed that chlorpyrifos adversely affected intestinal integrity, leading to increased entry of LPS bacteria into the body, and it included the induction of low-grade inflammation and alterations in the microbiota, ultimately culminating in insulin resistance and obesity, as indicated in the survey of Liang et al. [35]. A separate survey, encompassing 6,770 subjects aged 6–19 years, discovered a positive dose-dependent correlation between urinary levels of the dichlorophenol pesticide and obesity [36]. Our previous study indicated that pirimicarb induces a stressful state leading to anxiety and depression [10], which could be a key factor contributing to WG in P rats. Numerous studies support and reinforce our findings. The heightened perception of stress in modern society affects eating behavior, with sadness favoring the consumption of high-fat/sweet foods, hedonistically pleasant, while a delightful state is associated with an inclination

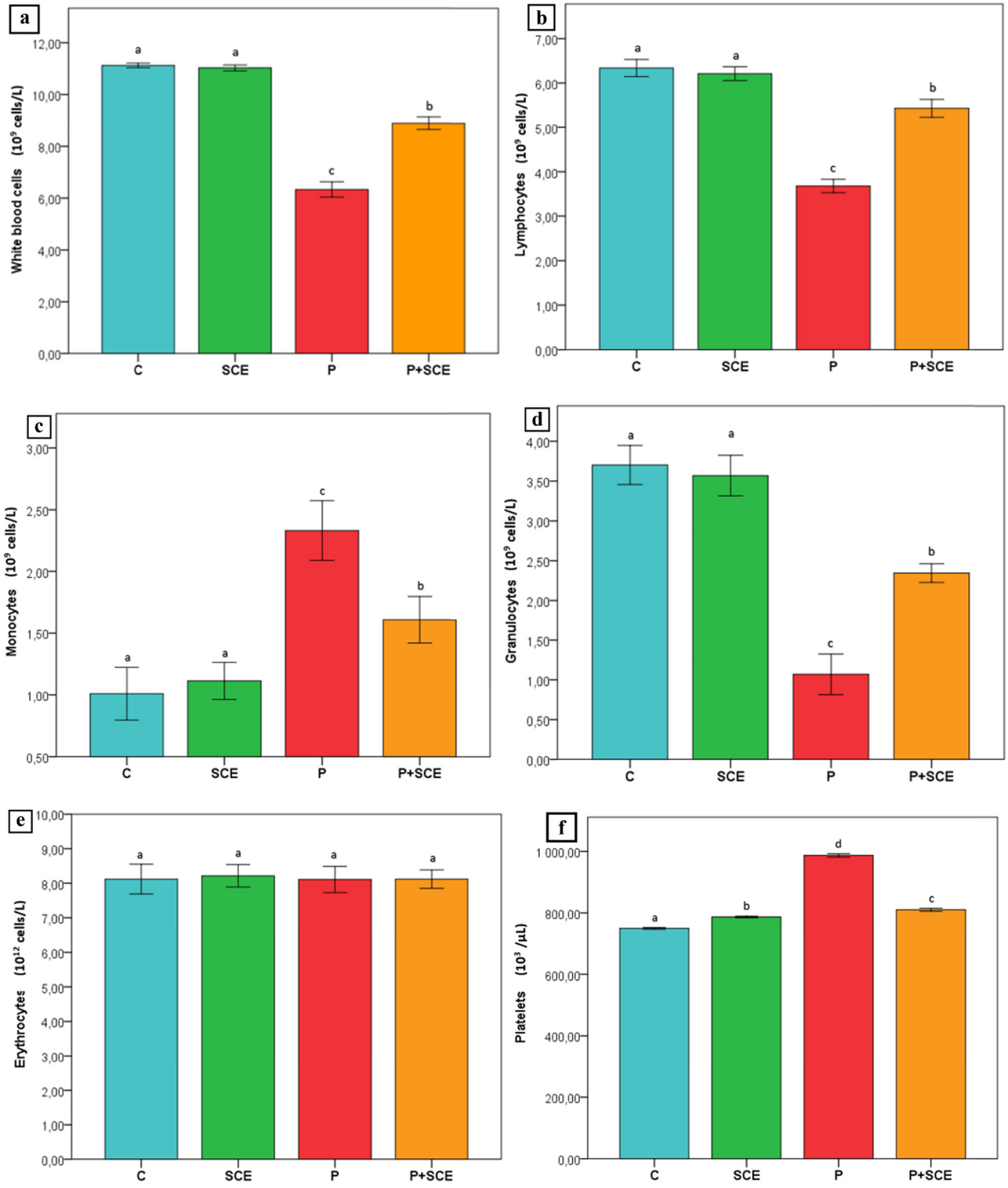

Figure 4: The count of blood elements. (a) White blood cell count, (b) Lymphocyte count, (c) monocyte count, (d) granulocyte count, (e) red blood cell (erythrocyte) count, and (f) platelet count. The outcomes were expressed as mean ± SD (N = 6). Analysis of variance (ANOVA one-factor and Tukey post-hoc tests) revealed statistical difference ($P < 0.05$). Different superscripts (a, b, c, and d) for the values in the same lines are statistically different.

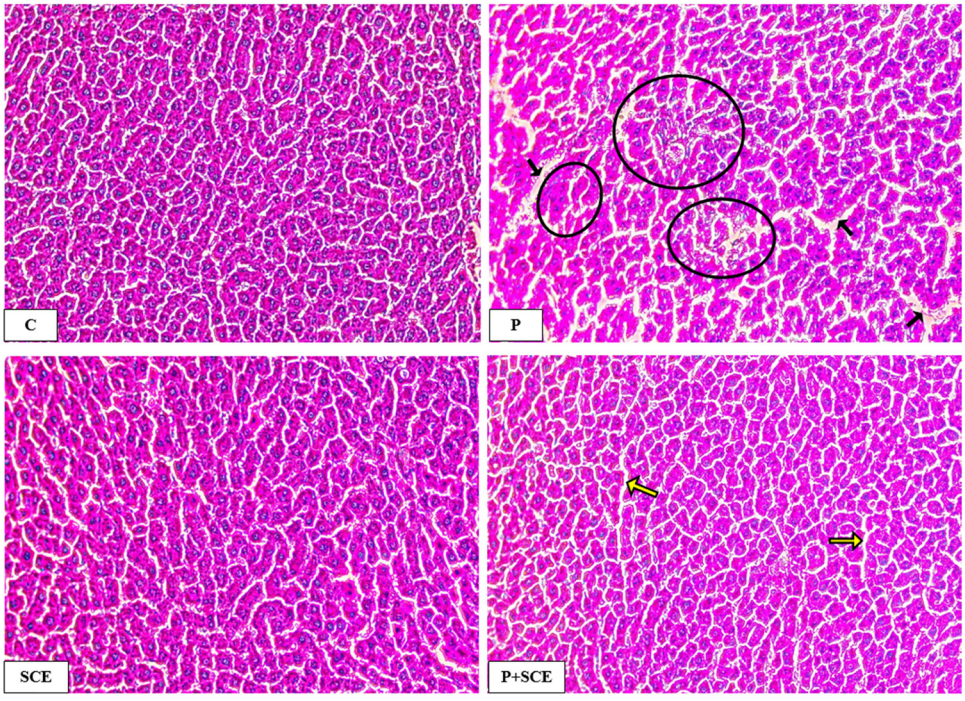

Figure 5: Photomicrographs of H&E-stained liver sections from experimental rats (400×). Control (C) rat showing normal histological structure. P (pirimicarb) rat's liver showing degenerative changes in hepatocytes (circle), loss of typical hepatic cord organization and sinusoidal dilatation (black arrow). P+SCE (shrub crude extract): treated rat liver showing a lesser degree of sinusoidal dilatation (yellow arrow) and normal cell morphology compared to the P group. SCE: treated rat liver showing normal appearance of hepatocytes.

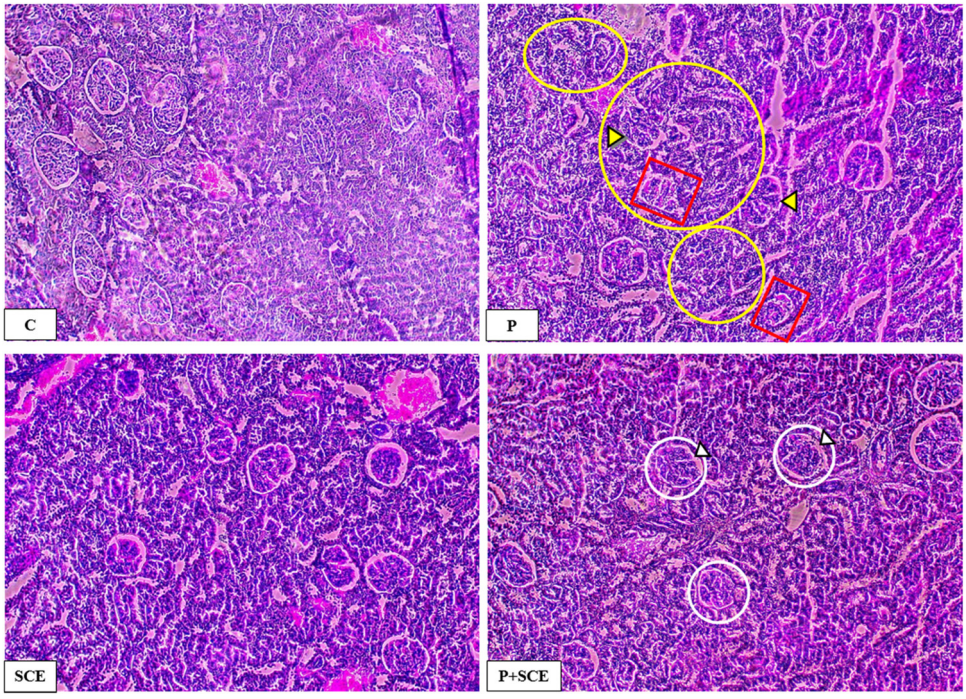

Figure 6: Photomicrographs of H&E-stained kidney sections from experimental rat kidney (100×). Control (C) rat showing normal renal histology. P (pirimicarb) rat's kidney showing mild glomerular atrophy (red square), narrowed Bowman's spaces (yellow arrowheads), and congestion (yellow circle). P + SCE (shrub crude extract)-treated rat kidney displaying normal renal architecture, slightly narrowed Bowman's spaces (white arrowheads) with focal congestion (white circle) compared to the P group. SCE-treated rat kidney showing mild congestion.

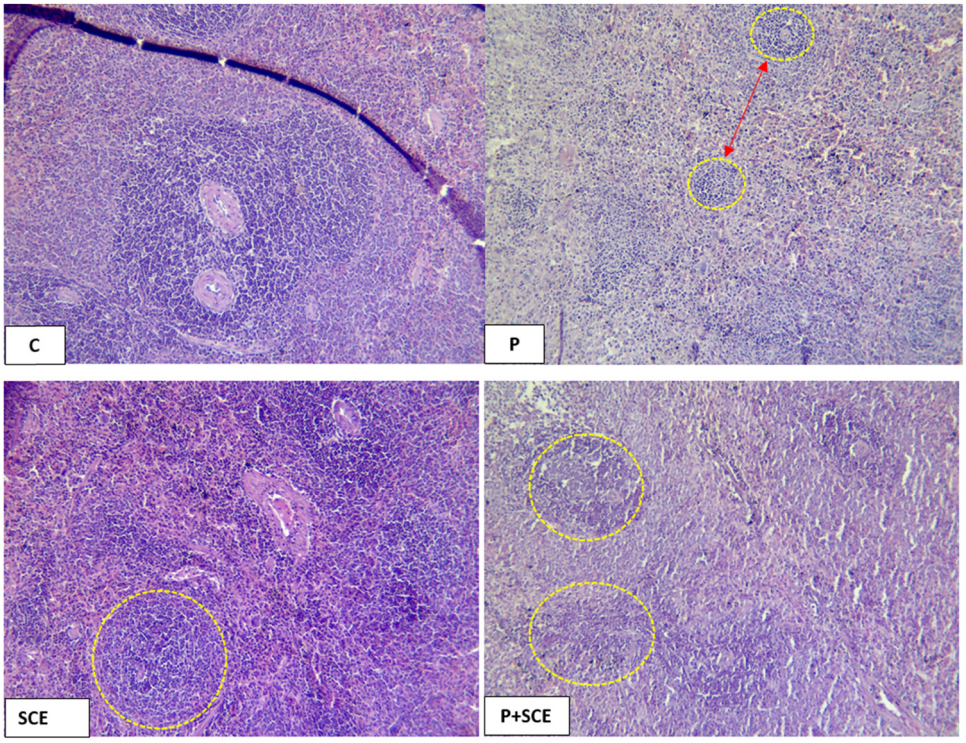

Figure 7: Photomicrographs of H&E-stained spleen sections from experimental rats. Control (C) rat showing normal splenic histology. P (pirimicarb) rat's spleen showing severe lymphocytic hypoplasia. P + SCE (shrub crude extract)-treated rat spleen demonstrating restored lymphoid follicular architecture with near normal cellularity compared to the P group. SCE C (control), treated rat spleen showing normal splenic histology. The double-headed red arrow demonstrates the increase in distance between the atrophied lymphoid follicular nodules.

Table 7: Semi-quantitative scoring of liver microscopic changes in different treatment groups of rats

Lesion	HC	SCE C	P	P + SCH
Degenerative changes	–	–	++	–
Loss of typical hepatic cords	–	–	++	–
Sinusoidal dilatation	–	–	++	+

(–) Absent, (+) mild and (++) moderate.

Table 8: Semi-quantitative scoring of renal microscopic changes in different treatment groups of rats

Lesion	C	SCE	P	P + SCH
Narrowing of the Bowman's space	–	–	++	–
Glomerular atrophy	–	–	+++	–
Congestion	–	–	+++	+

(–) Absent, (+) mild, (++) moderate and (+++) severe.

toward dried fruit. Stress triggers the secretion of glucocorticoids, increasing motivation for food, and insulin, promoting food intake and obesity [37]. Stress can also disrupt activity patterns, either by reducing physical activity or by increasing sedentary behavior. Additionally, it is known to disrupt sleep, leading to a reduction in sleep duration, which is tied to a higher susceptibility to obesity [38]. Several studies have endorsed the anti-diabetic and anti-obesity properties of *Ephedra alata Decne* [39,40]. Foods high in flavonoids and polyphenols have been shown to have a positive correlation with the prevention of type 2 diabetes and obesity. Numerous substances found in *Ephedra alata Decne* include apigenin, rutin, luteolin 7-O-glucoside, *p*-coumaric acid, gallic acid, vanilic acid, quercetin, cinnamic acid, apigenin-8-C-glucoside, epi-catechin, and quercetin-3-O-glucuronide. These substances have been shown to have inhibitory effects on the activity of lipase in the intestine and pancreatic tissues. Several of these substances, such as gallic acid, apigenin, and cafeic acid, show protective effects against hyperglycemia and atherosclerosis by inhibiting α-amylase, increasing insulin secretion, preventing insulin resistance, and decreasing body WG and adipose tissue weight [41]. Most of these potent anti-diabetic and anti-obesity metabolites were identified in our extract (SCE). Effectively, our extract significantly reduced WG in P + SCE rats treated with both pirimicarb and SCE (from 71.36 ± 12.03 g to 41.55 ± 11.71, Figure 3).

The gastrointestinal tract is the site of xenobiotic absorption, immediately after absorption, the liver becomes the

primary site for their metabolism. Among all organs in the body, the liver possesses the highest concentration of biotransformation enzymes. This makes the liver crucial for the detoxification of xenobiotics and safeguarding against chemical toxicity. Due to its involvement in the transformation of xenobiotics, the liver is susceptible to potential damage [42]. In the field of liver disease diagnosis, ALT, AST, and ALP are commonly utilized laboratory indicators. These enzymes are elevated in most prevalent liver problems [43]. ALT and AST are traditionally recognized as markers of hepatocellular injury, while ALP serves as an indicator of cholestasis [44].

Our results showed a significant increase in ALT and AST levels in rats treated with pirimicarb when compared to the control group. These results concur with numerous earlier research studies [45,46]. The outcomes of this investigation indicate that the elevated levels of ALT and AST may be indicative of liver tissue damage caused by pirimicarb. Normally, AST and ALT are found in low concentrations. Nevertheless, when there are cellular lesions or variations in the permeability of cell membranes, these enzymes can leak into the bloodstream. Given that AST levels can also rise in cases of cardiac arrest or muscle injury, ALT is thought to be a more sensitive and specific test for hepatocyte injury out of the two [47]. In the livers of rats treated with abamectin, histopathological lesions may be observed, suggesting damage. Additionally, the significant increase in LPO, which leads to changes in cell membrane permeability, reported in this study, may result in the leakage of enzymes into the blood due to free radical attack on the cell membrane. ALP, an enzyme present in the cell membrane, plays a crucial role in the dephosphorylation of various substances, such as nucleotides, proteins, and alkaloids, particularly in an alkaline pH environment. The administration of pirimicarb to rats can provoke a remarkable rise in ALP levels, which can be attributed to the degeneration and necrosis of hepatocytes, as well as damage to the membranes of cells [48]. When the liver cell membrane is impaired, several enzymes, including ALP, ALT, and AST, are released into the bloodstream from the hepatocyte cytosol [49]. Consequently, these serum enzymes serve as indicators of liver damage [50]. Previous studies have demonstrated that carbamate pesticides can enhance the enzymatic activities of ALP, ALT, and AST [49–52]. In our investigation, we observed significantly higher levels of PAL, ALT, and AST (Table 3) in animals treated with pirimicarb compared to the control group. This finding aligns with the damage observed in the hepatic tissues of pirimicarb-treated rats. Furthermore, the increase in PAL levels suggests an augmentation in lysosomal mobilization and cell necrosis due to pesticide toxicity [49].

Urea, creatinine, and uric acid are examples of waste materials and toxins that the kidney is essential in the body's removal. It is also important in controlling serum osmolality, electrolyte concentrations, and extracellular fluid volume. Furthermore, the kidneys produce renin, 1,25-dihydroxyvitamin D, and erythropoietin, among other hormones [53]. These hormones have important functions in the body. Due to its unique biochemical, anatomical, and physiological characteristics, the kidney is particularly susceptible to various toxic substances, including potentially harmful chemical elements found in the environment [54]. When assessing renal function and glomerular filtration, blood urea and creatinine levels are among the essential parameters measured in experimental animals. These parameters provide valuable insights into the overall health and functioning of the kidney [55].

Creatinine, a byproduct of regular muscle metabolism, undergoes a nonenzymatic conversion from creatine and phosphocreatine at a relatively constant pace, accounting for approximately 2% of the total creatine content each day. Due to its lack of binding to plasma proteins, it is easily filtered by the kidneys. Consequently, creatinine has gained significant recognition as a renal marker among medical professionals and researchers [56].

Our study revealed an important increase in creatinine levels of P rats compared to the control group (Table 4). Elevated serum creatinine levels are indicative of a notable decrease in the rate of glomerular filtration or obstruction in urine elimination. These findings align with previous research conducted on the toxicity of other pesticides within the same category [50,57,58]. The observed rise in plasma creatinine levels in P rats can be attributed to a preliminary reduction in the rate of glomerular filtration. Glomerular filtration is responsible for the excretion of various potential toxins, including pirimicarb, and compounds generated through cellular metabolism [59,60]. Consequently, progressive damage to the kidney may result in the retention of multiple substances, leading to an elevation in blood urea nitrogen and plasma creatinine levels [61]. It is worth noting that a significant loss of approximately 50% of kidney function is required prior to a creatinine level elevation becoming detectable [62]. Therefore, serum creatinine serves as a delayed indicator of acute kidney injury. Supporting our findings, other researchers have reported an increase in serum creatinine levels among agricultural workers who have been chronically exposed to high levels of pesticides [63,64].

Urea, a compound formed through the deamination of amino acids in the liver, is subsequently transported through the bloodstream to the kidneys for excretion in urine [65]. The findings of the current study revealed a significant rise in urea levels in rats treated with pirimicarb, in comparison to the control group. Previous research has also reported a notable increase in serum urea concentration as

a result of exposure to pesticides [45,66–68]. The elevated urea concentration observed in this study could potentially be attributed to the impact of pirimicarb on liver function (Table 4), as urea is the final product of protein breakdown. Additionally, it may indicate kidney dysfunction and disruption in protein metabolism, as indicated by the present results. Notably, excessive exposure to pesticides has been documented to induce cytotoxic changes in hepatic and renal biochemical markers, which exhibit a positive correlation with pesticide residue [69].

The primary way in which pesticides exert their toxic effects is by generating a high level of free radicals, which in turn leads to damage to tissues and organs [70]. This process is characterized by LPO, which is the oxidative degradation of polyunsaturated fatty acids [71]. LPO has been found to play a significant role in the toxicity and carcinogenicity of various xenobiotics. When LPO occurs in biological membranes, it causes changes in their structure and function, resulting in decreased membrane fluidity and the inactivation of certain membrane enzymes [72]. In the case of rats administered with pirimicarb, there was a significant increase in LPO in the liver and kidney. The main mechanism of toxicity associated with pirimicarb was the notable increase in the biomarker of MDA. These findings align with previous studies that have reported the ability of insecticides and their degradation products to act on membranes, oxidize lipid components, and enhance the production of free radicals during exposure [50,51,68,73]. The increase in LPO observed following pirimicarb exposure (Table 5) can be attributed to the induction of reactive oxygen species (ROS), which further promotes the oxidation of polyunsaturated fatty acids.

GSH, known as reduced glutathione, functions as a non-enzymatic antioxidant that eliminates free radicals generated from oxidative metabolism and evades breakdown by antioxidant enzymes. During GSH's metabolic process, its sulfhydryl group undergoes oxidation, forming a disulfide compound [74]. The reduction of GSH levels serves as a crucial biomarker for oxidative stress due to its involvement in conjugation and its role as an antioxidant in counteracting free radicals produced by insecticides, thereby maintaining the intracellular redox equilibrium in mammalian cells [75]. These findings align with previous studies conducted by various researchers [51,50,57,68]. Additionally, GSH plays a role in the detoxification of xenobiotics by serving as a substrate for the enzyme glutathione S-transferase (GST) [76]. Fetoui et al. emphasized that the depletion of intracellular sulfhydryl groups by insecticides is a prerequisite for the generation of ROS [77]. Consequently, the significant reduction in GSH content observed in the current investigation may enhance susceptibility to damage caused by free radicals.

CAT, an important enzyme, utilizes hydrogen peroxide as its substrate to carry out its functions. This enzyme plays a crucial role in neutralizing hydrogen peroxide by breaking it down, thereby maintaining an optimal level of this molecule within the cell. This optimal level is essential for various cellular signaling processes [78]. In the pirimicarb group, the activity of CAT was found to be significantly reduced compared to the control group (Table 5), which is consistent with previous studies [68,79–81]. This reduction in CAT activity can be attributed to the presence of superoxide radicals, which are known to decrease the activity of CAT. Insecticides, such as pirimicarb, generate excessive ROS, disrupting the balance of antioxidant enzymes. Consequently, the inhibition of CAT, which is responsible for removing free radicals, leads to the accumulation of superoxide. This accumulation, in turn, promotes LPO, cell death, and tissue alterations [64].

Protein, the primary biochemical component found in abundance in the body, is essential for various metabolic reactions. The liver, known for its high protein content, serves as a hub for these reactions. In the study, it was observed that the pirimicarb group exhibited a significant decrease in total protein levels in the kidney compared to the control group [82–84]. This decline can be attributed to an increased rate of proteolytic activity or the repeated breakdown of protein to generate energy, which is a response to the stress caused by pesticide exposure [83]. This decrease in total protein concentrations in the kidney is considered an indicator of the toxicological adverse effects of pesticides [82]. However, contrasting results were observed in the liver, where the pirimicarb-treated group showed a significant increase in total protein levels compared to the control group. The liver, being a vital organ involved in metabolism, performs various functions such as glycogen storage, decomposition of red blood cells, synthesis of plasma proteins, hormone production, and detoxification of xenobiotics like pesticides [84]. Recent studies have suggested that exposure to insecticides can impact protein metabolism [85,86], which may explain the elevated levels of total protein in the liver of the pirimicarb-treated group in our study.

The hepatic dysfunction caused by pirimicarb can be inferred from the decreased activities of CAT, total proteins, and increased MDA levels in the liver and kidney, as well as increased serum AST and ALT activities. This suggests that pirimicarb may lead to the formation of free radicals through its metabolism in the liver, specifically through hydrolytic ester cleavage and oxidative pathways mediated by the cytochrome P450 microsomal enzyme system. It is likely that this metabolism process results in a decrease in P450 contents in the liver, which in turn induces oxidative stress. This oxidative stress leads to a depletion of CAT activity and an

increase in MDA levels, ultimately causing hepatic degeneration and necrosis [87].

The impact of pirimicarb on leucocytes is evident in the form of significant lymphopenia and agranulocytosis, with lymphocyte levels decreasing from 6.34×10^9 cells/L to 3.67×10^9 cells/L and granulocyte levels decreasing from 3.70×10^9 cells/L to 1.07×10^9 cells/L. Variations in white blood cell counts are recognized as reliable indicators of stress induced by environmental factors. Lymphopenia, a reported consequence of pesticide exposure, often accompanies an increase in monocytes as a response to stress triggered by pesticide exposure [88]. This may be explained by the potential involvement of free radicals and oxidative stress induced by the pesticide, leading to immune-cytotoxicity [89]. Indeed, the lymphocytic hypoplasia remarked in spleen slides of rats exposed to pirimicarb along with the atrophied lymphoid follicular nodules is associated with the toxic effect of the pesticide that led to cell death through several mechanisms. Similar studies have been undertaken on the impact of pesticides on immune responses; they have shown suppression of cell mediation immune response (decrease in the expression of CD4 helper T cells and CD8 cytotoxic T cells), spleen toxicity evidenced by a decrease in B lymphocyte number, reduction in total and differential leukocyte counts, phagocytic activity, phagocytic index, immunoglobulins, and lysosomal activity and a subsequent (these latest changes were observed in thymus and spleen tissues) [90–92].

Furthermore, pirimicarb has increased the level of monocytes from 1.00×10^9 cells/L to 2.33×10^9 cells/L. Studies in rodents have indicated that stress enhances the production of inflammatory cytokines, such as IL-1β or IL-6, by spleen cells and peritoneal macrophages. Stress can also reduce the sensitivity of macrophages to glucocorticoids. In humans, acute stress involving catecholamines like noradrenaline can activate the NF-κB pathway in monocytes, leading to the expression of inflammatory cytokine genes. Stress may also intensify the release of TNF-α, IL-1β, and IL-6 by macrophages through the induction of local release of CRH via free nerve endings. The observed elevation in IL-1β levels [10] may be parallel to the increased number of monocytes induced by pirimicarb-induced stress [93]. Platelet levels have also shown a notable increase (987×10^3/µL vs 750×10^3/µL) due to pirimicarb's impact. This finding aligns with a study by veterinarian researchers who observed a significant rise in platelet count in chickens after exposure to a pesticide (thiram), attributed to increased expression of thrombopoietin mRNA in the dysfunctional liver [94]. Similarly, our findings resonate with an Algerian survey on farmers using chemical pesticides, where changes in hemostasis were noted, including a procoagulant state with increased clotting factors, oxidative stress, and inflammation associated with elevated plasma CRP levels [10,95].

The SCE has played a general enhancing role against edema, OS, metabolism dysregulation, immune depression, and tissue damage. In preventing the disruptive mechanisms induced by pirimicarb in liver, kidney, and spleen function, the antagonistic reaction was established through the synergic effect resulting from antioxidant, anti-inflammatory, and euphoric metabolites, namely, phenolics, flavonoids, saponins, steroids, tannins, terpenoids, carbohydrates, and alkaloids [9,10,39,96–100]. Effectively, the SCE has exhibited impressive protection defined by its ability to inhibit the pirimicarb effect in regards to metabolic function (the values of the biochemical dosed parameters in P + SCE are nearer to normal experimental groups than those of P group) and anatomical integrity of liver, spleen, and kidney (the lesions are less important in the slides of rats exposed to pirimicarb in conjunction with the SCE administration). Moreover, enhancing the antioxidant activity by upregulating the expression of antioxidant elements (GSH and CAT) and reducing LPO. It is noteworthy to state that the SCE provided a satiety sensation and suppressed the negative impact of probable causative factors of excessive consumption of food and WG (dealing with emotional stress, gut microbiota imbalance along with its ability to inhibit lipase and alpha-amylase enzymes [9]). We come across the following assertions to give some insight into the detailed effect of each type of metabolite present in the SCE. In the past two decades, polyphenols have been extensively researched for their potential roles in various areas, such as cancer, cardiovascular diseases, inflammation, and microbial infections. Initially, their protective effects were linked to their antioxidant properties, as well as their ability to scavenge free radicals and chelate metals. However, further investigations revealed that polyphenols could also inhibit or reduce the activity of different enzymes. Emerging evidence indicates that polyphenols may interact with signal transduction pathways and cell receptors, enhancing their biological activity in diverse ways [101]. Tannins represent a diverse group of polyphenolic compounds characterized by their high molecular weight and solubility in water. These compounds offer various pharmacological benefits, including antioxidant properties and the ability to scavenge free radicals. Additionally, tannins exhibit antimicrobial, anti-cancer, anti-nutritional, and cardio-protective effects. They also demonstrate positive impacts on metabolic disorders and are believed to help prevent the development of diseases associated with oxidative stress [102]. Terpenoids, which represent the most prevalent compounds found in natural products, constitute a significant group of secondary metabolites in plants, exhibiting diverse structures. These compounds possess a wide array of beneficial properties, including antitumor, anti-inflammatory, antibacterial, antiviral, and antimalarial effects. They are also known

to enhance transdermal absorption, aid in the prevention and treatment of cardiovascular diseases, and exhibit hypoglycemic activities. Moreover, previous research has identified numerous potential applications for terpenoids, including immunoregulation, antioxidation, antiaging, and neuroprotection [103]. Phytosteroids have garnered attention within the realm of natural products due to their promising array of pharmacological activities. A comprehensive review of literature across various scientific search engines has compiled valuable information regarding the types of phytosteroids and their effectiveness against inflammation and allergic complications. It is suggested that phytosteroids exert anti-inflammatory effects through diverse mechanisms, including transrepression or selective inhibition of COX-2 enzymes [104]. In fact, there is a growing fascination with bioactive carbohydrates because of their wide-ranging biological uses. Researchers have conducted extensive studies to understand the structure and mechanisms of action of natural polysaccharides and their derivatives. Many of these compounds have garnered attention for their proven biological effects, including antitumor, antioxidant, anti-diabetic, antiviral, hypolipidemic, and immunomodulatory properties [105]. All ephedra plants inherently possess alkaloids derived from phenylalanine, such as ephedrine, pseudoephedrine, methylephedrine, and trace quantities of phenylpropanolamine. The long-term safety and effectiveness of ephedra alkaloids were investigated, revealing that they can effectively decrease body weight and fat content while also enhancing blood lipid profiles, all without causing significant adverse effects [106].

12 Conclusions

Pirimicarb belongs to carbamate pesticides that have noxious effects if consumed frequently. In effect, it interferes with metabolic homeostasis, provoking obesity, affecting histology, and disturbing the function of the liver and kidney. Besides, it reduces and depresses the immune potentialities by provoking severe leucopenia and aberration in lymphoid tissues like the spleen. The Algerian Saharan shrub named *Ephedra alata* Decne has the property to protect the function and the histologic integrity of liver, kidney, spleen, and blood. Notably, it has the ability to promote the antioxidant activity, which leads to preventing cell death. Certainly, the virtues exhibited by the plant in the context of chemically induced edema and toxicity are assumed to be attributed to its possession of a wide array of secondary metabolites, including polyphenols, flavonoids, saponins, steroids, terpenoids, tannins, alkaloids, and carbohydrates. In accordance with our previous study and based on the current outcomes, we could consider this edible plant as a general health care agent. To attain this goal, further surveys are required to adapt the pharmacological parameters for adequate use.

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Conflict of interest: The authors assert that they do not have competing interests.

Ethical approval: The study was authorized by the institutional ethic committee of the CRBt and granted an ethical approval under the reference of N07KH-2021/2023/CCE.

Data availability statement: The corresponding author can provide the data required to support the findings of this study upon request.

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