In Vivo Evaluation of Genotoxicity and Antioxidant Capacity of *Ajuga Orientalis* L. (Lamiaceae) Leaf Extracts

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ABSTRACT

Ajuga orientalis L. (Lamiaceae) is a fragrant herb native to the Eastern Mediterranean region, widely used in traditional healing practices in Jordan and neighboring countries. Despite its extensive use, there is a lack of toxicological studies on its leaf extracts. This study aims to address this gap by evaluating the genotoxic potential of ethanolic and aqueous leaf extracts using a micronucleus (MN) assay on mice DNA, alongside assessing their antioxidant status. The median lethal dose 50% (LD_{50}) was tested in ten groups of sixty male *Balb/c* mice to determine the acute toxicity of *A. orientalis* leaf extracts. Four groups of male *Balb/c* mice (n=6) were used to evaluate micronucleus (MN) formation and total antioxidant capacity for each extract. Each group received daily intraperitoneal injections of one of the following concentrations: 4000, 2000, 1000, and 500 mg/kg over 28 days. Additionally, three control groups were included for comparison purposes. Peripheral blood samples were screened for MN formation, and liver samples were assessed for total antioxidant capacity. Results revealed an LD50 of 4000 mg/kg for both extracts, alongside a significant dose-dependent increase in MN formation and lower antioxidant capacity compared to controls. The findings indicate the genotoxicity of *A. orientalis* leaf extracts in *Balb/c* mice, urging caution in human consumption. Further research is warranted to comprehensively assess their safety and toxicity, especially considering their traditional medicinal use.

Keywords: medicinal plant; micronucleus; oriental bugle; total antioxidant capacity.

1. INTRODUCTION

The utilization of medicinal plant products and supplements has seen considerable growth in recent decades. Approximately 70-80% of the global population relies on traditional medicinal plants as a primary source of healthcare according to the World Health Organization (WHO). In spite of the accessibility of modern synthetic medications, individuals continue to opt for natural herbs for certain aspects of their primary healthcare [1]. Because they are natural, there is a prevalent perception that herbal products are inherently safer than allopathic medicines [2]. This perception in addition to low cost makes the use of herbal medicine more prominent. However, the use of herbal medicines without adherence to safety standards and without conducting toxicological studies is incorrect and harmful [3], potentially leading to adverse health effects due to their inherent toxic chemical composition [2].

Over the past few years, there has been increasing attention on investigating the toxicity, mutagenic properties, and potential carcinogenic effects of commonly used traditional medicinal plants [4]. The toxic effect of some medicinal plant extracts can lead to numerous chromosomal structure abnormalities, changes, and

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damage deoxyribonucleic acid (DNA) [5]. Such plants possessing DNA-damaging effects are classified as genotoxic plants [6]. Interestingly, it has been discovered that certain plants commonly used in traditional medicine, such as *Artemisia absinthium* L., *Crataegus oxyacantha* L. *Equisetum arvense* L., *Plantago lanceolata* L. and *Synadenium umbellatum* Pax, possess genotoxic potential [7–11]. DNA damage plays an etiological role in several human diseases including cancer [3]. The genotoxic potential was found not confined to a particular plant family, several studies have reported that proved genotoxic plants are common and frequent in the following families: *Fabaceae*, *Asteraceae*, *Euphorbiaceae*, *Rosaceae*, *Lamiaceae*, and *Apocynaceae* members [6]. Therefore, and for safety measures, a genotoxicity assessment of medicinal plants used in folkloric medicine appears to be required [8,12].

In Jordan, about 20% of the total flora are listed as medicinal plants [13]. Many of these plants have a long history of traditional use in medicine and are also utilized in the pharmaceutical industry [14]. While the biological effects and chemical composition of several herbal remedies used in traditional medicine have been investigated, many of them are still used without any validation of their safety and efficacy [1,15]. One of the most important and common herbal families worldwide including Jordan is the *Lamiaceae* family, members of this family genera such as *Salvia*, Teucrium, Thymus and Origanum showed wide medical and biological uses [16–18].

In Jordan and neighboring countries, there is an increasing demand for wild herbal plants, including *A. orientalis*, driven by the growing belief among local communities in their therapeutic properties, especially in cancer treatment. Moreover, the plant leaves are edible, either consumed fresh in salads or dried as a component of Jordanian manasaf spices. *Ajuga orientalis* L., commonly known as Eastern bugle, is a flowering species in the Lamiaceae family native to the eastern Mediterranean regions, including Jordan [19]. Due to its medicinal attributes, this species has found applications in traditional medicine across the globe for the treatment of conditions such as rheumatism, gout, malaria, and asthma, hypertension, hyperglycemia, joint pain, and gastrointestinal diseases [1,16,20–22]. Furthermore, members within the genus *Ajuga* have been documented for possessing medicinal properties such as: antibacterial, anti-inflammatory and antioxidant [23,24]. Notably, the ethanolic extract derived from the aerial parts of *A. orientalis* has exhibited noteworthy cytotoxic effects against breast and colon cancer [1]. In general, most cytotoxic plants are bears to have a genotoxic potential [2]. However, despite its widespread use and potential therapeutic benefits, there is a notable absence of comprehensive toxicological studies on *A. orientalis* leaf extracts. Our research aims to address this critical gap by evaluating the genotoxic potential and antioxidant status of these extracts. Assessing the safety profile and potential adverse effects of *A. orientalis* is crucial, especially considering the rising demand for this plant in traditional medicine.

2. MATERIALS AND METHODS

2.1. Plant material

The aerial parts of *A. orientalis*, at flowering period, were collected from roadside of Ishtafina forest, Ajloun city, north Jordan (32°35′47″N 35°76′85″E) in March 2021. The plant materials have been collected and authenticated by Dr. Mohammad Al-Gharaibeh (Department of Plant Production, J.U.S.T) and a voucher specimen (PHS-136) was deposited in the herbarium of the Faculty of Pharmacy, JUST.

2.2. Ethanolic and aqueous extracts preparation

Fresh leaves of *A. orientalis* were detached from stems, washed with tap water, and allowed to air-dry at room temperature in the shade for a period of three weeks. Afterward, they were finely ground into a powder. Ten grams of the powdered material were added to 100 ml of both solvents (distilled water and 90% ethanol) for extract preparation. For aqueous extract, the traditional way of

decoction preparation was followed by boiling the mixture over low heat for 5 minutes. Subsequently, the mixture was left to reflux for 30 minutes until it reached room temperature (approximately 25°C). Following this, the extract was centrifuged and filtered. For the ethanolic extract, the soaked powdered leaves with 90% ethanol were exhaustively extracted using the maceration technique for 24 h, with regular shaking. Subsequently, the extracts were centrifuged and filtered. To avoid the potential toxicity of ethanol, the extracts were diluted at a ratio of 1:5 (1 ml plant extract with a solution of 4 ml distilled water and 10% polyethylene glycol). The obtained crude extracts, both aqueous and ethanolic, were sealed in an airtight container and preserved at a temperature of -20°C for subsequent analysis. Different concentrations of the plant extracts were prepared by serial dilution.

2.3. Animal Study

Healthy 8-week-old male *Balb/c* mice with an average weight of 27.5 ± 3 g were employed to test the LD_{50}, MN formation, and total antioxidant capacity activity (TAC) of *A. orientalis* leaf extracts. The mice were housed in the animal facility at Yarmouk University, specifically in an animal room where standard laboratory conditions were maintained. These conditions included a temperature range of 20-22°C, relative humidity between 60-80%, and a lighting cycle of 12 hours of light/ 12 hours of darkness. The study protocol was approved by the Department Research Committee at Yarmouk University.

2.3.1. The median lethal dose (LD_{50})

To measure the median lethal dose (LD_{50}) of *A. orientalis* leaf extracts, 60 mice were utilized. The animals underwent an overnight fast lasting approximately 14 hours. They were then distributed among 10 groups, with each group consisting of 6 mice per group. Both extracts were administered intraperitoneally (IP) for 24 hours at varying doses, with each group receiving a daily single injection of one of the following doses: 5000, 4000, 2000, 1000, and 500 mg/kg. Mortality cases or indications of toxicity were monitored in the tested mice and the lowest dose concentration that killed 50% of mice in the tested groups indicated the value of LD_{50}.

2.3.1. Micronucleus (MN) formation, and total antioxidant capacity

In vivo animal study was conducted using 66 *Balb/c* male mice to assess the genotoxic potential of *A. orientalis* leaf extracts. After one week of environmental acclimation, mice were divided into a total of 11 groups (n=6 mice/group). Four groups were assigned to the aqueous extraction method, and four groups were assigned to the ethanolic extraction method. Each group received a daily dose of one of the following concentrations: 4000, 2000, 1000, and 500 mg/kg of plant extract given via the IP route of injection. Additionally, two groups served as solvent negative controls (distilled water and 20% ethanol), and one group for the positive control (mitomycin C, 14 mg/kg, Shabbar *et al*., 2012). Both treatment and control experiments were run simultaneously and terminated after 28 days. At study termination, and after 30 hours of injection, the mice were euthanized by cervical dislocation. The ethylenediaminetetraacetic acid (EDTA) as an anticoagulant was used to collect peripheral blood samples. In addition, the liver of mice was collected in pre-sterilized tubes.

2.4. Micronucleus assay

2.4.1. Blood smears preparation

Clean pre-washed glass slides were used to prepare the blood smears. From each mouse, three slides were created, and these blood films were allowed to air-dry. Subsequently, they were fixed in absolute methanol for a duration of 3 minutes, following the methods described by Heddle in [26] and Schmid in [27]

2.4.2. Staining and MN evaluation

All the prepared slides were double-stained with Mayer's hematoxylin (BioGnost, Zagreb) for 10 minutes and then 10% Giemsa (GCC, UK) for 20 minutes [28]. The slides were thoroughly rinsed in tap water, followed by a 10-minute differentiation step in Sorensen's buffer (pH 6.8). Afterward, they were left to air-dry. Observations were conducted using

a BioBlue light microscope (Euromex, Netherlands) equipped with an oil immersion lens (100X). For each treatment, 2000 normochromic erythrocytes from each animal were screened for MN formation.

2.5. TAC assay

To measure the total antioxidant capacity of *A. orientalis* leaf extracts, MyBioSource CheKine ™ Total TAC assay kit (Cat. number MBS9718973, lot. number ATTJL2301, USA) was used. To conduct this test, liver samples were collected in pre-sterilized tubes. Subsequently, 0.1 gram of each liver sample was homogenized with 1 ml of assay buffer, and the resulting mixture was centrifuged at 12,000 x, at 4°C for 10 minutes. The supernatant was then transferred to a new tube and kept on ice for further detection. For sample detection, 10 µl of each sample was combined with 150 µl substrate diluent, 15 µl substrate, and 15 µl reaction buffer. The sample mixtures were then incubated in a 96-well plate at room temperature for 5 minutes, and the optical density (O. D) was measured at 593 nm. The Total Antioxidant Capacity (TAC) for each sample (expressed in U/g) was determined using the following formula:

$$TAC(U/g) = \frac{0.6 \times (O.D_{\ sample} - O.D_{\ blank})}{(O.D_{standard} - O.D_{blank})} \times \frac{1}{W}$$

Where: *O.D* is the optical density, *W* is the weight of the sample in grams.

2.6. Statistical analysis

The data underwent analysis through a one-way analysis of variance (*ANOVA*) using SPSS, version 26, USA. The data were presented as mean values ± standard error of the mean. Following the ANOVA, Tukey's multiple comparisons tests were conducted as a post-hoc analysis. Data were deemed statistically significant when the *p* value was less than 0.05.

3. RESULTS

3.1. The median lethal dose LD_{50}

After a 24-hour period following a single injection, there were no indications of toxicity or mortality within the dosage range of 500-2000 mg/kg for both leaf extract methods. However, at higher doses of 4000 and 5000 mg/kg, the mortality rate resulting from intraperitoneal administration increased progressively with the rising dose, as detailed in Table (1). Nonetheless, the mortality percentage at the dose of 4000 mg/kg was equal for both aqueous and ethanolic extract methods (Fig.1). Conversely, at the highest tested dose (5000 mg/kg), the number of mouse fatalities was greater in the aqueous extraction group compared to the ethanolic group. The median lethal dose LD_{50} of *A. orientalis* extract was detected in the experimental group of mice that were administered a dose of 4000 mg/kg for both types of extracts, as indicated in Figure (1).

Table 1. Results of the tested doses of A. orientalis extract used to ascertain the LD_{50} following intraperitoneal injection in Balb/c male mice.

Extract	Group	N. of Mice	Doses (mg/kg)	Death number	Mortality %
Aqueous	A	6	500	0	0
	B	6	1000	0	0
	C	6	2000	0	0
	D	6	4000	3	50
	E	6	5000	5	83.3
Ethanolic	A	6	500	0	0
	B	6	1000	0	0
	C	6	2000	0	0
	D	6	4000	3	50
	E	6	5000	4	66.7

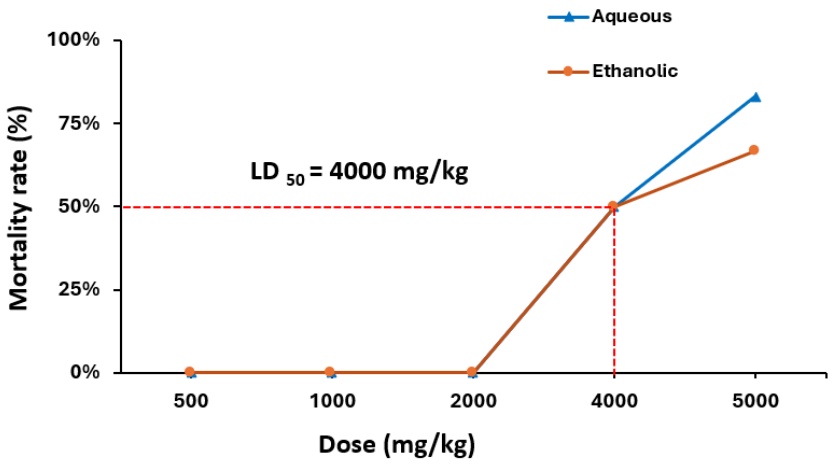

Figure 1. Dose-response (mortality rate %) curve. The red dashed line represents the LD $_{50}$.

3.2 MN assay

The percentage of micronuclei was determined after scoring approximately 2000 cells. As demonstrated in Table (2), a noticeable increase in the percentage of micronucleus (MN) formation was observed as the extract doses increased for both types of extracts. However, it's worth mentioning that this percentage was slightly higher in the aqueous extract (decoction). The highest observed percentages of MN were 0.0679% and 0.052% at the dose of 4000 mg/kg for the aqueous and ethanolic extracts, respectively. The percentage of micronucleus (MN) formation at a dose of 4000 mg/kg for the aqueous extract represents a substantial increase when compared to the control groups (distilled water and 20% ethanol), with percentages of 0.001% and 0.007%, respectively. For instance, a noticeable disparity in the incidence of MN formation was evident between the mice group that received 2000 mg/kg and the negative control group treated with the aqueous extract (Figure 1). This discrepancy in the means of MN formation subsequent to the administration of *A. orientalis* extracts was found to be statistically significant ($p < 0.05$) when compared to both the negative and positive control groups (Table 2).

Table 2. the micronucleus (MN) formation observed in *Balb/c* mice at various concentrations of *A. orientalis* extract.

	Aqueous extract		**Ethanolic extract**	
Treatment	Mean ± S.D $^{\Sigma}$	Freq. (%) ± S.D	Mean ± S.D $^{\Sigma}$	Freq. (%) ± S.D
N. control	15.75 ± 2.5	0.001 ± 0.001	14.8 ± 3.962	0.007 ± 0.002
P. control (MMC)	79.5 ± 5.259	0.04 ± 0.003	79.5 ± 5.259	0.04 ± 0.003
500 mg/kg	53.8 ± 5.263$^{*,\#}$	0.027 ± 0.003	44.2 ± 3.193$^{*,\#}$	0.022 ± 0.002
1000 mg/kg	83 ± 4.743$^{*,\#}$	0.042 ± 0.002	71.6 ± 2.873$^{*,\#}$	0.036
2000 mg/kg	93.66 ± 7.359$^{*,\#}$	0.046 ± 0.004	86 ± 4.516$^{*,\#}$	0.043 ± 0.002
4000 mg/ kg	135.83 ± 3.656$^{*,\#}$	0.0679 ± 0.002	103 ± 3.741$^{*,\#}$	0.052 ± 0.002

"Σ" denotes the total count of cells with micronuclei (MN) in normochromatic erythrocytes, "#" is used to denote statistical significance ($p < 0.05$) when compared with the positive group, while "*" is used to denote statistical significance ($p < 0.05$) when compared with the negative group. MMC is the abbreviation of mitomycine C.

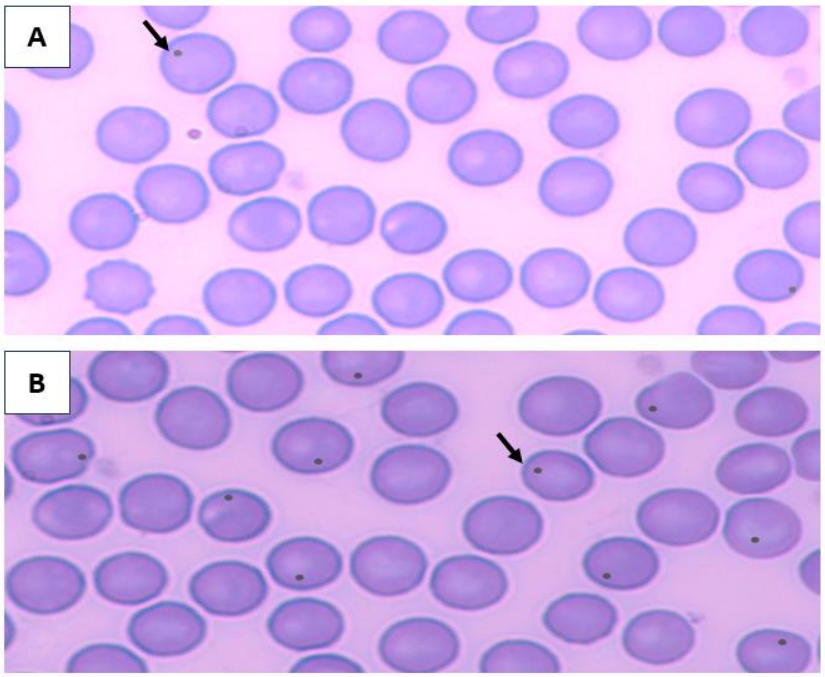

Figure 1. Representative photomicrographs of mouse blood smear stained with 10% Giemsa, (A) negative control group (B) 2000 mg/kg group of aqueous extract. The arrow indicates MN formation.

3.3 TAC assay

Table (3) presents the outcomes of Total Antioxidant Capacity (TAC) in the liver cells of mice following the administration of varying doses of aqueous and ethanolic *A. orientalis* extracts. For both types of extracts, the mean ± standard deviation (SD) values of the experimental groups exhibited an increase with rising doses. It is worth noting that the TAC, an indicator of the total antioxidant capacity of the extracts, was higher in the ethanolic extract as compared to the aqueous extract. Furthermore, both extracts exhibited lower antioxidant capacity across all concentrations compared to the positive and negative control groups. In the case of the aqueous extract, the results indicated statistically significant differences ($p < 0.05$) in the means of TAC for all doses when compared to both the negative and positive control groups. On the other hand, for the ethanolic plant extract, the group that received the lowest dose (4000 mg/kg) exhibited a TAC value that was in proximity to the positive control value ($p > 0.05$).

Table 3. The levels of TAC in *Balb/c* mice at different concentrations of *A. orientalis* extract.

	Aqueous extract	**Ethanolic extract**
Treatment	Mean ± S.D $^{\Sigma}$	Mean ± S.D $^{\Sigma}$
N. control	158.350 ± 0.572	169.9 ± 3.140
P. control (MMC)	156.38 ± 0.571	156.325 ± 0.665
500 mg/kg	84.150 ± 0.641*, #	130.967 ± 3.149*, #
1000 mg/kg	89.472 ± 12.366*, #	135 ± 0.969*, #
2000 mg/kg	92.917 ± 8330*, #	146.40 ± 0.550*, #
4000 mg/ kg	116.90 ± 8.894*, #	155.34 ± 0.5550

"Σ" denotes the total amount of TAC (U/ g), "#" is used to denote statistical significance ($p < 0.05$) when compared with the positive group, while "*" is used to denote statistical significance ($p < 0.05$) when compared with the negative group. MMC is the abbreviation of mitomycine C.

4. DISCUSSION

The utilization of medicinal plants as the primary means of medical treatment has been a historical and enduring practice in developing nations [29]. Plant-based natural products remain an abundant source of novel phytochemicals and nutraceuticals [30]. It's important to note that some of these phytochemicals found in medicinal plant extracts may be associated with toxicity [31]. Hence, it is crucial and advisable to evaluate the toxicity of medicinal plant extracts during the preclinical assessment phase [11]. The current study demonstrates that the aqueous and ethanolic extracts obtained from *Ajuga orientalis* leaves exhibit toxicity when administered through the intraperitoneal (IP) route in mice, with toxic effects observed at doses of 2000 and 4000 mg/kg. Notably, mortality was only observed in mice when relatively high doses of both extracts were intraperitoneally injected, resulting in an LD_{50} value of 4000 mg/kg for these extracts. In a closely related and commonly used medicinal plant, *Ajuga iva*, a previous study by El Hilaly [32] found that the LD50 for acute intraperitoneal administration in mice and rats was 3600 mg/kg. Given that closely related plants tend to share similar or identical chemical constituents, this suggests that phytotoxicity is often consistent at the genus level [33].

One of the primary and recommended tests for evaluating product safety is the in vivo assessment of micronucleus formation (MN) frequencies [34]. Consequently, the utilization of MN as an indicator of chromosomal damage has become a widespread method for assessing genotoxicity and conducting human biomonitoring research [35]. In our research, we observed a dose-dependent increase in the percentage of MN formation in bone marrow cells of mice with escalating concentrations of the extracts. This rise in MN-containing erythrocytes among treated mice was statistically significant for both the aqueous and ethanolic extracts when compared to the control groups (both negative and positive). Such a dose-related increase in MN formation is considered an indicative sign of genotoxicity activity [34]. Where Phytochemicals present in plants are among the genotoxic agents known to induce MN formation [36], which can subsequently lead to DNA or chromosomal damage [37]. Several previous studies have explored the genotoxic effects of various plant species. These investigations have found that extracts from plants such as *Crataegus oxyacantha, Aristolochia debilis, Asristolochia heterophylla, Astagalus membranaceus, Bupleurum talcatum, Canrthamus tinctorius, Cinnamomum mairie, and Cuscuta chinensis* can increase the percentage of MN formation in peripheral cells in a dose-dependent manner [9,38]. Consequently, our results suggest that both extracts derived from *A. orientalis* leaves have a significant impact on the induction of MN formation, indicating high genotoxicity, particularly at higher doses. Nevertheless, it's important to note that in human use, exposure to crude extracts of medicinal plants typically occurs orally, often in the form of decoctions, rather than through injection. Therefore, the dose, extraction method, and route of administration for herbal medicines are crucial factors that should be thoroughly investigated for their potential toxicity and safety implications [2].

Despite the widespread use of *A. orientalis* in traditional medicine, the extracts derived from its leaves exhibited low antioxidant capacity. In general, both extracts showed significantly lower antioxidant activity compared to the control groups. This outcome aligns with similar findings reported in studies on various folk medicinal *Ajuga* species that used different assays, such as *A. orientalis* [1], *A. bracteosa* [39], *A. parviflora* [40], and *A. reptans* [41]. On the other hand, the choice of extraction method, specifically the solvent used, influenced the total antioxidant capacity (TAC). In the case of *A. orientalis* leaves, the ethanolic extract exhibited higher TAC compared to the aqueous extract. This trend has also been observed in the aforementioned studies on *Ajuga* species. Generally, the addition of 20%–60% ethanol or methanol to distilled water can significantly enhance the antioxidant

capacity during plant extraction [42]. This enhancement is attributed to their ability to extract higher levels of phenolic and flavonoid compounds while reducing the consumption rate of endogenous antioxidants, thus acting as antioxidants [43]. It's important to emphasize the significance of natural antioxidants, as they are regarded as a preventive approach against chronic diseases due to their capacity to reduce oxidative stress and enhance immune function [44]. Furthermore, natural antioxidants can contribute to improving healthcare systems by reducing the dependency on costly and frequently inefficacious treatments. In the specific case of our study species, *A. orientalis*, the low antioxidant capacity of its leaf extracts suggests that it may not be a promising natural source of antioxidants and may have limited potential as a medicinal plant in this regard.

5. CONCLUSIONS

In summary, the findings derived from this study indicate that *A. orientalis* leaf extracts display genotoxic potential while exhibiting a relatively modest antioxidant capacity. Both the aqueous and ethanolic extracts of *A. orientalis* leaves induced a statistically significant increase in micronuclei (MN) formation in normochromic erythrocytes among treated mice, following a dose-dependent pattern. Therefore, it underscores the critical importance of considering dosage and extraction methods in the evaluation of potential toxicity and safety associated with herbal medicines. This research underscores the imperative need for further investigations to thoroughly evaluate the safety and toxicity of *A. orientalis*, particularly focusing on its active chemical constituents that may induce potential genotoxic effects. This is especially pertinent given its continued utilization in traditional medicinal practices.

Declaration of competing interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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تقييم السمية الجينية والقدرة المضاداة للأكسدة في مستخلصات أوراق عشبة الدم الشرقية في الفئران *Ajuga orientalis* L. (Lamiaceae)

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ملخص

يعد نبات عشبة الدم الشرقية من النباتات العطرية المستوطنة لمنطقة شرق البحر الابيض المتوسط والتي تستخدم بشكل شائع في الطب الشعبي في الاردن والدول المجاورة. على الرغم من استعمالها الواسع، إلا أنه لا توجد دراسات سمية على مستخلصات اوراقها. تهدف هذه الدراسة لتقييم سمية هذا النبات في مسخلصات الاوراق عن طريق الايثانول والماء لتقييم باستخدام اختبار تكوين الأنوية الصغيرة في كريات الدم الحمراء الصبغية السوية في الحمض النووي الريبوزي منقوص الأكسجين للفئران (DNA) وايضا لتقييم إجمالي نشاط القدرة المضادة للأكسدة. لتحديد الجرعة القاتلة الوسيطة (LD_{50}) لمستخلصات أوراق نبات الدم الشرقي، تم اعطاء تراكيز متفاوته ل 66 من ذكور فأر *Balb/c*. تم حقن الفئران يوميا بداخل البطن بالتراكيز التالية 4000 و2000 و1000 و500 ملغ/كغ وتم استخدام 3 مجموعات كضوابط ولمدة 28 يوماً. تم فحص تكون الأنوية الصغيرة في كريات الدم الحمراء العادية، وتم تقييم عينات الكبد لقياس القدرة الكلية المضادة للأكسدة.كان تركيزالجرعة القاتلة الوسيطة لكلا المستخلصين 4000 ملغ/كغ، مع زيادة معنوية في تكوين الأنوية الصغيرة في كريات الدم الحمراء العادية كلما زاد تركيز الجرعة. اظهرت النتائج انخفاض في قيم إجمالي نشاط القدرة المضادة للأكسدة في كلا المستخلصين مقارنة بالضوابط. تشير النتائج إلى وجود سمية جينية لمستخلصات أوراق نبات الدم الشرقي في الفئران ، مما يحث على الحذر في استهلاكها من قبل البشر. يجب إجراء المزيد من الأبحاث لتقييم سلامتها وسميتها بشكل شامل، خاصةً مع استخدامها التقليدي الواسع في الطب الشعبي.

الكلمات الدالة: الأنوية الصغيرة، نبات الدم الشرقي، نبات طبي، إجمالي نشاط القدرة المضادة للأكسدة.

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