

PHYTOCHEMICAL ANALYSIS, ANTI-INFLAMMATORY AND ANALGESIC PROPERTIES OF *ALLIUM TRIQUETRUM* L. IN MICE

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Abstract

Description of the subject: *Allium triquetrum* L. is a spontaneous plant from the Mediterranean basin used in traditional medicine, but rarely studied.

Objective : The objective of this study is to evaluate *in vivo* the anti-inflammatory and analgesic profiles of polar extracts from leaves and bulbs (AqEL, AqEB) of *Allium triquetrum* L. and its phytochemical analysis.

Methods : HPLC-DAD analysis was used to determine the chemical composition of the samples. A preliminary acute toxicity was also studied. The anti-inflammatory activity was assessed using the carrageenan-induced paw edema test and the analgesic effect was examined by the acetic acid-induced writhing model.

Results : phytochemical analysis showed the presence of phenolic compounds. Oral administration of extracts at doses up to 2,000 mg/kg did not induce death or toxic symptoms in mice. The treatment with AqEL and AqEB, produced significant dose-dependent inhibition of edema development. The AqEL at the dose of 250 mg/kg exhibited the best anti-inflammatory activity with an edema decrease rate of 59.45%, while the AqEB exhibited a percentage inhibition of 39.45%, at the same concentration. In addition, pretreatment of mice with AqEL, AqEB significantly reduced the number of writhes and the effect was found to be dose-dependent. AqEL and AqEB at the dose of 250 mg/kg reduced the number of writhes with a percentage of analgesia of 72.94% and 52.42%, respectively.

Conclusion : The present study suggests that aqueous extracts of *A. triquetrum* possess potential analgesic and anti-inflammatory effects.

Keywords : *Allium triquetrum* L.; Phenolic compounds; Acute toxicity; Anti-inflammatory effect; Analgesic effect.

ANALYSE PHYTOCHIMIQUE, PROPRIETES ANTI-INFLAMMATOIRE ET ANALGESIQUE DE *ALLIUM TRIQUETRUM* L. CHEZ LES SOURIS

Résumé

Description du sujet : *Allium triquetrum* L. est une plante spontanée du bassin méditerranéen utilisée en médecine traditionnelle, mais rarement étudiée.

Objectifs : L'objectif de cette étude est d'évaluer *in vivo* les profils anti-inflammatoire et analgésique des extraits polaires des feuilles et bulbes (AqEL, AqEB) de *Allium triquetrum* L. et sa composition phytochimique.

Méthodes : L'analyse HPLC-DAD a été utilisée pour déterminer la composition chimique des échantillons. La toxicité aiguë préliminaire a également été étudiée. L'activité anti-inflammatoire a été évaluée en utilisant le test d'œdème de pattes postérieures induit par la carragénine et l'effet analgésique a été examiné par le modèle d'induction des spasmes par l'acide acétique.

Résultats : L'analyse phytochimique a montré la présence de composés phénoliques. L'administration orale d'extraits à des doses allant jusqu'à 2 000 mg/kg n'a pas entraîné de mort ni de symptômes toxiques chez les souris. Le traitement par AqEL et AqEB a produit une inhibition dose-dépendante significative du développement de l'œdème. L'AqEL à la dose de 250 mg/kg a présenté la meilleure activité anti-inflammatoire avec un taux de diminution de l'œdème de 59,45%, tandis que l'AqEB a présenté un pourcentage d'inhibition de 39,45%, à la même concentration. De plus, le prétraitement des souris avec AqEL et AqEB a réduit de manière significative le nombre de spasmes et l'effet s'est avéré être dose-dépendant. AqEL et AqEB à la dose de 250 mg/kg ont réduit le nombre de contorsions avec un pourcentage d'analgésie de 72,94% et 52,42%, respectivement.

Conclusion : La présente étude suggère que les extraits aqueux d'*A. triquetrum* possèdent des effets analgésiques et anti-inflammatoires potentiels.

Mots clés : *Allium triquetrum* L., composés phénoliques, toxicité aiguë, effet anti-inflammatoire, effet analgésique,

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INTRODUCTION

Inflammation is the immune system's response to harmful stimuli, such as pathogens, damaged cells, toxic compounds, or irradiation [1]. Inflammation progression is usually associated with pain, which is the one of most cardinal signs including swelling, redness, lack of function, and fever [2]. Non-steroidal anti-inflammatory drugs (NSAIDs) are widely used in the treatment of acute and chronic inflammation, pain and fever. But the greatest disadvantage in presently available synthetic drugs is that they cause gastrointestinal irritation and reappearance of symptoms after discontinuation [3]. As a result, the use of medicinal plants to treat and manage diseases including inflammatory and body pain which has been in existence since time immemorial is becoming more popular globally [4]. The research on plants with apparent folkloric use, as agony relievers, anti-inflammatory agents, should therefore be regarded as a prolific and a rational research strategy in the search for new anti-inflammatory and analgesic drugs [5]. *Allium* species have a long history in common folklore and as sources of therapeutic principles [6]. In fact, the edible parts of *Allium* plants are used for the treatment and prevention of a number of diseases: coronary heart disease, cancer, obesity, diabetes, disturbances of the gastrointestinal tract, hyper-cholesterolemia and inflammatory diseases [7]. *Allium triquetrum* L., also known as three-cornered leek, triangle onion or triangular stalked garlic, belonging to the genus *Allium*, is a bulbous perennial flowering plant, native to the western and central Mediterranean region, being commonly found in wetland ecosystems. It is characterized by green striped, white, pendulous flowers looking like small lilies [8]. *A. triquetrum* L. is reported in Tell region, but quite rare in west of Algeria [9] [10]. In some regions in Algeria (Kabylia and Blida), the whole plant is eaten as an omelet and used to flavor bread or to season salads.

Thus, the young leaves are eaten raw or cooked and can accompany other vegetables in couscous. According to several ethnobotanical studies, the triangle onion is used by local population for its anti-inflammatory, antiseptic and anti-parasitic properties [11]. The bulb is hypotensive and deworming [12, 13]. Thus, the whole plant and flowers are used for the prevention of kidneys and intestines and also exhibits soporific effects [14].

Some studies have demonstrated the antimicrobial [15, 16] and antioxidant effects of *A. triquetrum* [17] and suggest that this properties due to the presence of secondary metabolites such as phenols, saponins, fatty acids and aromatic compounds [15-17]. However, no pharmacological study has been conducted to evaluate anti-inflammatory and analgesic activity of three-cornered leek, supporting traditional uses of this plant in folklore medicine. Hence, this topic has been chosen to evaluate in vivo, for the first time, the analgesic and anti-inflammatory activities of the aqueous extracts of *A. triquetrum* L. bulbs and leaves. Furthermore, HPLC analysis was performed to identify some bioactive compounds. The acute toxicity of extracts is also evaluated.

MATERIAL AND METHODS

1. Plant collection

A. triquetrum L. samples were collected during the flowering period (March-May, 2014), in the region of Mitidja (Chebli, Blida, Algeria). Bulbs and leaves of the triangle onion were separated, washed with distilled water and cut into small pieces. Thereafter, biomass was dried and protected from light at room temperature (25-28°C). The plant material is finally reduced to powder.

2. Sample preparation

20 g of powdered samples of leaves and bulbs were added to 500 mL of boiling distilled water (100°C) under magnetic stirring, for 30 minutes. The aqueous extracts (AqEL, AqEB) are then cooled, filtered by Whatman filter paper 4 and evaporated using a vacuum rotary evaporator (Laborota 4001-efficient Heidolph 2, Schwabach, Germany) to obtain dry concretes and kept at 4 °C until further use.

3. Animals

Healthy Male and female Swiss albino mice (18–21 g) were used to evaluate preliminary acute toxicity, anti-inflammatory and analgesic effects. Females were nulliparous and not pregnant. They were housed in cages and maintained on a 12 h light/dark cycle, at 25°C, with constant humidity. Animals were obtained from the laboratory of pharmaco-toxicology of the Center of Research and Development (CRD SAIDAL, Algiers, Algeria) and the laboratory of pharmaco-toxicology of Antibiotical Saidal Company (Medea, Algeria). Commercial pellet diet and water were provided *ad libitum*. The animals were fasted overnight prior to each experiment but had free access to water.

4. HPLC-DAD ANALYSIS

Qualitative phytochemical investigation was carried out at the laboratory of analysis of the centre of biotechnology of Sfax (CBS-SFAX, Tunisia). Dried extracts were re-dissolved and diluted to 1 mg/mL in MeOH (water/methanol 30/70, v/v). Chromatography study was performed on an HPLC-DAD (Agilent, Series 1200, 454 Waldbronn, Germany). The separation was achieved on an Agilent column Zorbax Eclipse x DB - C18 (5 µm, 3×250 mm) with two mobile phases: solvent A (0.1% acetic acid in water) and solvent B (100 % acetonitrile). Flow rate was set at 0.4 mL/min. The analysis was maintained at room temperature and the injection volume was 20 µL. The detection was performed in DAD (UV-VIS) at a wave-length of 280 nm. The identification is done by extrapolation of the chromatograms of the extracts to that of the standard molecules and comparison of their retention times. All standards (gallic acid, naringenin, chlorogenic acid, syringic acid, coumarin, protocatechic acid, methylgallate, ferulic acid, rutin, hydroxycinnamic acid and catechin) at purity of 99.9% HPLC grade were purchased from Sigma Chemical USA). These molecules are prepared (water/methanol 30/70, v/v) and analyzed under the same operating conditions as for the samples.

5. Preliminary acute toxicity

The acute toxicity of the polar extracts was carried out on the laboratory of pharmacotoxicology of the Center of Research and Development (CRD SAIDAL, Algiers, Algeria), The assay was performed, in vivo, according to the protocol of N'GUESSAN *et al.* [18]. Animals were divided into seven experimental groups of 10 mice each (5 male, 5 female). Group 1 received 0.5 mL of 0.9% physiological water and served as control. Groups 2 to 4 treated with 0.5 mL of the aqueous extract of *A. triquetrum* leaves (AqEL) at the doses of 1.200, 1.600 and 2.000 mg/kg

$$\% \text{ inhibition of inflammation} = \frac{\% \text{ AUG control group} - \% \text{ AUG treated group}}{\% \text{ AUG control group}} \times 100$$

% AUG is the percentage increase in the volume of the mouse paw; it is calculated by the following equation:

$$\text{AUG}(\%) = \frac{\text{Average left hind foot weights} - \text{Average right hind foot weights}}{\text{Average right hind foot weights}} \times 100$$

7. Analgesic effect

The analgesic effect was examined by the acetic acid-induced writhing test [21]. Animals were divided into 6 groups of 6 mice each (3 males, 3 females). Group 1 received the normal saline (0.9% NaCl, 0.5 mL) and served as a control.

and groups 5 to 7 were treated with 0.5 mL of the aqueous extract of *A. triquetrum* bulbs (AqEB) at the doses of 1,200, 1,600 and 2,000 mg/kg respectively. All treatments were administered once by oral gavage. Mice were kept under regular observation for 4 h following administration for any adverse effects, including mortality. Other behavioral changes and parameters, food intake, water intake, diarrhea, and locomotor activity were also monitored. Observations were further extended up to 14 days for any signs of mortality. If possible, the LD50 was estimated by the log dose-probit analysis method [19].

6. Anti-inflammatory effect

Anti-inflammatory assay was realized at the laboratory of pharmaco-toxicology of Antibiotical Saidal Company (Medea, Algeria). The carrageenan-induced hind paw edema test was performed according to the protocol described by Winter *et al.* [20], with slowly modification. Animals divided into 6 groups with each group containing 6 mice (3 male, 3 female). The control group and the reference group received normal saline (0.9% NaCl, 0.5 mL) and diclofenac (0.5 mL, 12.5 mg/kg), respectively. Groups 3 and 4 treated with 0.5 mL of AqEL at the concentrations of 100 and 250 mg/kg and groups 5 and 6 were treated with 0.5 mL of AqEB at the concentrations 100 and 250 mg/kg respectively. Saline, extracts and diclofenac were all administered orally. After 30 min, inflammation was induced by injection of 0.1 mL of a freshly prepared 1% λ carrageenan suspended in sterile physiological saline into the right hind foot of each mouse under the sub-plantar aponeurosis. After 5 hours, the mice are sacrificed and the hind feet are cut at joint height and then weighed using an analytical balance. The percentage inhibition of inflammation is calculated according to the following formula:

Groups 2 received the standard drug indomethacin (25 mg, 80 mg/kg) and served as a reference group. Groups 3 and 4 received AqEL at the concentrations of 100 mg/kg and 250 mg/kg, respectively. Groups 5 and 6 treated with 0.5 mL of the AqEB at the concentrations

of 100 mg/kg and 250 mg/kg, respectively. All oral administrations were performed 30 min before injection of acetic acid. Writhing was induced in mice by intraperitoneal injection of 0.2 mL of 1% acetic acid solution. The mice were then placed in transparent observation cages for observation and the stretching reaction

resulting from abdominal constriction was evaluated. The number of writhes (muscular contractions) was counted for 20 min immediately after the acetic acid injection for each group. The percentage inhibition of writhes (%) was calculated by the following formula:

$$\% \text{ Inhibition of writhes} = \frac{\text{Average number of writhes (control)} - \text{Average number of writhes (treated)}}{\text{Average number of writhes (control)}} \times 100$$

8. Statistical analysis

Data are expressed as mean ± SD. The data obtained were evaluated through the one-way analysis of variance (ANOVA) followed by Tukey’s test. In all cases, differences were considered significant if *p* >0.05. All statistical analyses were performed using the software XLStats 2013 (statistic software Pros, Addinsoft, Paris, France).

RESULTS

1. HPLC Analysis

The chromatograms profiles of aqueous extracts of *A. triquetrum* are shown in the figure 1 and figure 2. Results of tentative identification of some phenolic compounds are presented in the table 1 and table 2. HPLC analysis of aqueous

extracts of triangle onion revealed the presence of some phenolic compounds which are mainly coumarin, phenolic acids (gallic acid, ferulic acid and hydroxy-cinnamic acid), flavonoids such as flavonols (rutin) and flavanones (naringenin). However, catechin (flavanols) was only identified in the AqEL. In addition, except gallic acid, which is the major component of all the substances identified in aqueous extracts (AqEB : 1.50%, AqEL : 7.28%), the other compounds were presented in small amounts. Furthermore, it was not possible to identify the major peaks of the obtained chromatographic profiles through the comparison of retention time at the analytical standards. These compounds are under investigation.

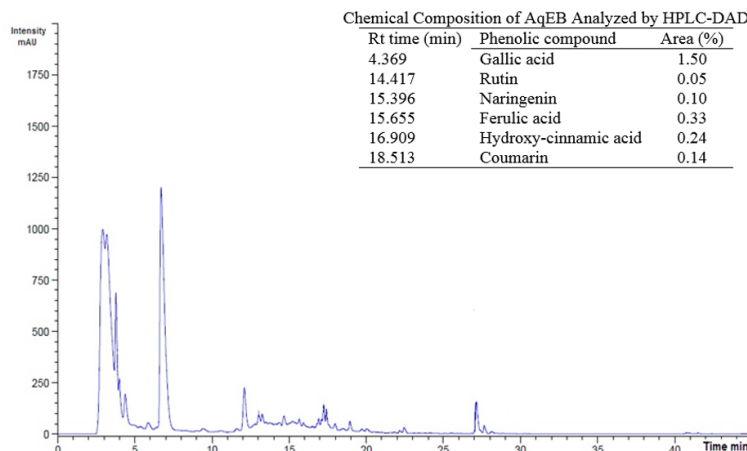


Figure 1: Chromatogram profile of aqueous extract of *A. triquetrum* bulbs AqEB (280 nm)

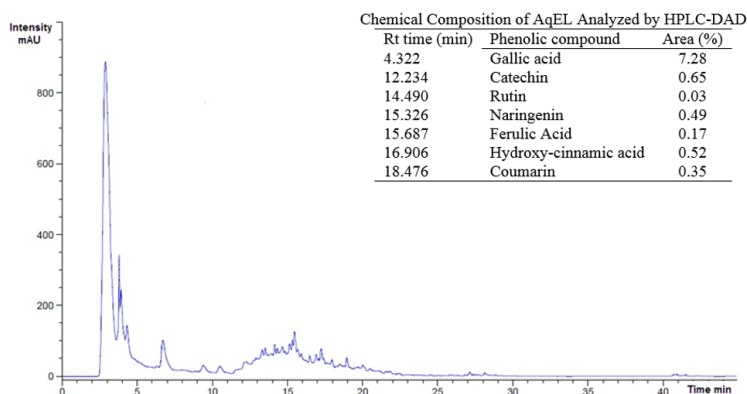


Figure 2: Chromatogram profile of aqueous extract of *A. triquetrum* leaves AqEL (280 nm)

2. Acute toxicity

All of the mice treated with aqueous extracts of *A. triquetrum* (AqEB, AqEL) at the different doses (1.200, 1.600 and 2.000 mg/kg) were alive for all 14 days of observation. No abnormal gross findings were observed in any of the animals. The acute toxicity of *A. triquetrum* polar extracts was therefore

considered to be unclassified; doses up to 2.000 mg/kg did not induce death or toxic symptoms.

3. Anti-inflammatory activity

The anti-inflammatory effect of polar extracts of *Allium triquetrum* (L.) leaves and bulbs evaluated by the carrageenan-induced paw edema is shown in figure 3.

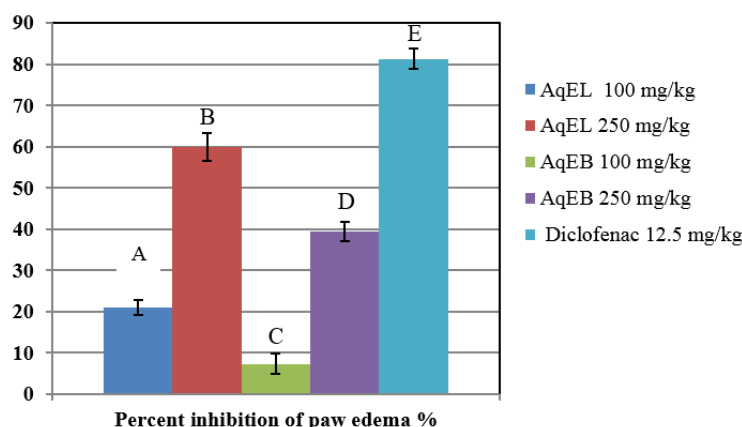


Figure 3: Inhibition rate of paw edema (%) in mice receiving polar extracts (AqEL, AqEB) of *A. triquetrum* and diclofenac. Values are reported as mean ± SD with n= 6 per group. The means with different letters are significantly different ($p < 0.05$) according to a one-way ANOVA followed by Tukey's multiple test comparisons.

Histogram shows that the anti-inflammatory activity of the polar extracts of *A. triquetrum*, at different doses, is considerably lower than that elicited by 12.5 mg/kg of the diclofenac (estimated percentage of inhibition is $81.28 \pm 2.45\%$). Nevertheless, the extracts tested produced significant ($p < 0.05$) dose-dependent inhibition of edema development. The aqueous extract of leaves AqEL at the dose of 250 mg/kg exhibited the best anti-inflammatory activity with an edema decrease rate of $59.45 \pm 3.45\%$, while the polar extract of bulbs AqEB exhibited a percentage inhibition of $39.45 \pm 2.39\%$, at the same concentration. The anti-inflammatory power of AqEL and AqEB at the dose of 100 mg/kg is less interesting. In fact, the percentages of inhibition are equal to $20.99 \pm 1.74\%$ and $7.39 \pm 2.44\%$, respectively.

4. Analgesic effect

The analgesic effect of polar extracts of *Allium triquetrum* (L.) leaves and bulbs evaluated by the acetic acid-induced abdominal writhing

response is shown in table 3 and figure 4. Pretreatment of mice with AqEL, AqEB significantly reduced the number of writhes ($p < 0.001$) and the effect was found to be dose-dependent. Thus, the analgesic power of AqEL at dose of 200 mg/kg is close to that of the molecule used in therapy, indomethacin (80 mg/kg), which showed an inhibition rate of writhes equal to $71.45 \pm 1.17\%$. ANOVA analysis followed by Tukey's multiple test comparison showed no significant difference at ($p > 0.05$) between the mean inhibition rates of the two groups. Oral administration of AqEL reduced the number of writhes caused by the injection of acetic acid, with a percentage of analgesia of $30.78 \pm 3.45\%$ and $72.94 \pm 1.10\%$ at doses of 100, 250 mg/kg, respectively. On the other hand, the AqEB showed a significant decrease of writhes compared to the control group with percentage of analgesia of $19.4 \pm 2.35\%$ and $52.42 \pm 1.17\%$ at doses of 100 and 250 mg/kg, respectively.

Table 3: Analgesic effect of *A. triquetrum* aqueous extracts

Extract	Number of writhes	Percent reduction (%)
Control (NaCl 0.9%)	89.33 ± 0.82	/
Indomethacin (80 mg/kg)	25.5 ± 1.05***	71.45 ± 1.17%
AqEL (100 mg/kg)	61.83 ± 3.12**	30.78 ± 3.45%
AqEL (250 mg/kg)	22 ± 0.73***	72.94 ± 1.10%
AqEB (100 mg/kg)	72 ± 2.09*	19.4 ± 2.35%
AqEB (250 mg/kg)	42.5 ± 1.05***	52.42 ± 1.17%

Values are reported as mean ± SD; n = 6 per group; the results are statistically significant at: * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$ according to a one-way ANOVA followed by Dunnett's test comparing the experimental group to the control group. AqEL: aqueous extract of leaves; AqEB: aqueous extract of bulbs.

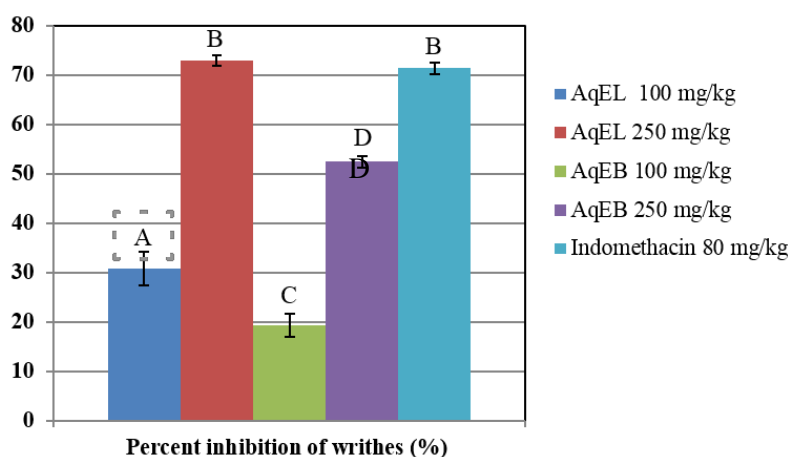


Figure 4. Percentage inhibition of writhes in mice treated by polar extracts (AqEL, AqEB) of *A. triquetrum* and indometacin. Values are expressed as mean \pm SD with $n=6$ per group. The means with different letters are significantly different ($p < 0.05$) according to a one-way ANOVA followed by Tukey's multiple test comparisons.

DISCUSSION

A. triquetrum is a medicinal plant used in the folk medicine of northern Algeria. However, little is known about its possible biological effects. Hence, this is the first report about the anti-inflammatory and analgesic activities of the aqueous extracts from bulbs and leaves of *A. triquetrum* using oral administration in experimental protocols in mice model. Data showed the presence of some phenolic compounds with noticeable anti-inflammatory and analgesic activities of samples at both doses of 100 and 250 mg/kg, especially the 250 mg/kg dose. Some studies have focused on determining the composition of the different organs of *Allium triquetrum* L. The work of Corea et al. (2003) showed that the bulbs and flowers of *A. triquetrum* contain high proportions of flavonoids, in particular kaempferol [8]. In recent study, it has been shown the presence of some phenolic acids (cinnamic acid, ferulic acid, hydroxybenzoic acid) in the lipophilic fraction from the leaves, bulbs and flowers of *A. triquetrum* [16]. Several studies have shown the anti-inflammatory and analgesic powers of *Allium* species. Indeed, the study of PAN et al. (2015) showed that the aqueous extract of *A. sativum* has appreciable anti-inflammatory activity with an edema inhibition of 68.1% at a dose of 100 mg/kg and 74.09% at a dose of 200 mg/kg [22]. Another study showed that the powder of *A. sativum* at doses of 75, 150 and 300 mg/kg produced abdominal cramp reduction rates of 33.65%, 57.44% and 72.10%, respectively [23]. Thus, the study of Ranjan et al. (2010) based on the comparison of the anti-inflammatory and analgesic effects of different extracts (aqueous,

methanolic and petroleum ether) of *A. Stracheyi* leaves showed a possible inhibitory capacity of edema and possible inhibition of writhes of all the extracts tested. The best activity given is that of the methanolic extract with 61% rate inhibition of edema and 64.62% rate inhibition of writhes at a dose of 100 mg/kg [24]. The inflammation model of a carrageenan induced edema is usually used to assess the activity of natural products in resisting the pathological changes associated with acute inflammation. Inflammation induced by carrageenan is acute, non immune, well-researched, and highly reproducible [25]. The edema formation is a biphasic event, the release of histamine, serotonin, and bradykinin occurs in the first phase, and the second phase is associated with the production of prostaglandin, protease, and lysosomal enzymes [26]. For our study, results tend to suggest that the inhibitory of the *A. triquetrum* extracts on edema formation is probably due to the inhibition of the synthesis and/or release of the inflammatory mediators, especially the cyclooxygenase (COX) products. The carrageenan induced paw edema test is effectively controlled with the arachidonate cyclooxygenase (COX) inhibition due to its COX-dependent mechanism [27], thus, it is suggested that the AqEL and AqEB may possess arachidonate COX inhibitory property. According to the literature, the anti-inflammatory activity of *Allium* species is attributable to organosulfur compounds and polyphenols [24]. These molecules exert an inhibitory action on inflammation, which is thought to involve the inhibition of the formation of the main pro-inflammatory mediators of arachidonic acid metabolism via the inhibition of COX and LPO [28].

In the present study, phytochemical analysis of the aqueous fractions (AqEL, AqEB) revealed the presence of phenolic compounds in particularly gallic acid which may be responsible of the anti-inflammatory power of the plant. However, this activity can also be linked to several other secondary metabolites belonging to other chemical classes such as saponosides, triterpenes and steroids [29]. Thus, this activity has been reported to be related to the synergistic effect of secondary metabolites [30]. Thus the phytochemical study of these extracts has shown that the greater content of phenolic compounds present in the leaves and explains its greater anti-inflammatory activity compared to the bulbs. On the other hand, the anti-edematous effect is probably linked to the antioxidant effects of the extracts already proven [17], since reactive oxygen species (ROS) are involved in the pathophysiology of diseases with an inflammatory component (cancer, diabetes, atherosclerosis, arthritis, infectious diseases). They induce the release of cytokines (TNF α , IL 1 β , IL 6) and the activation of pro-inflammatory enzymes (COX, LPO, inducible nitric oxide synthase) involved in the inflammatory process [31]. Acetic acid writhing test was adopted for evaluation of the analgesic power of *A. triquetrum* (L.) polar extracts. The abdominal constriction response induced by acetic acid is a sensitive procedure to evaluate peripherally acting analgesics. In general, acetic acid causes pain by liberating endogenous substances such as serotonin histamine, prostaglandins (PGs), bradykinins and substance P, endings. Local peritoneal receptors are postulated to be involved in the abdominal constrictions response. The method has also been associated with prostanoids in general that is, increased levels of PGE2 and PGF2 α in peritoneal fluids as well as lipoxigenase products [32]. The decrease in the number of writhes induced by aqueous extracts of leaves and bulbs suggests that the antinociceptive action may be peripherally mediated by the inhibition of synthesis and/or liberation (release) of prostaglandins. The analgesic power of *Allium* species is largely linked to the effect of organosulfur and phenolic compounds [24]. The results of the phytochemical study obtained in our experiment showed the presence of some phenolic compounds such as gallic acid, in the leaves and bulbs of *A. triquetrum*, and the best concentrations of phenolic compounds are found in the leaves which explains the results obtained.

In addition, it has been suggested that analgesic potency results from the synergistic effect between secondary metabolites presents in the extracts [30].

CONCLUSION

Data obtained in the present study showed that polar extracts of *A. triquetrum* bulbs and leaves possess significant anti-inflammatory and analgesic activities which are in accordance with folkmedicine of many *Allium* plants. However, the analgesic effect is more interest to the ant-inflammatory power. HPLC-DAD analysis of AqEL and AqEB revealed the presence of some phenolic compounds such as gallic acid, catechin, rutin, naringenin, ferulic acid, hydroxy-cinnamic acid and coumarin. The presence of phenolic compounds in the aqueous fractions might have some role in the observed pharmacological activities. In addition, the acute toxicity does not show any symptoms, changes in behavior or mortality at 2000 mg/kg doses that indicate a therapeutic safety for the doses pharmacologically active. Further investigations are required to identify all the active compounds present in *Allium triquetrum* L. and their precise mechanisms of action. Moreover, it could be a potential source for discovery of anti-inflammatory and analgesic drug development.

Conflict of interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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