Study of the antioxidant, anti-inflammatory and analgesic properties of extracts of *Gomphrena serrata* L. (Amaranthaceae), a plant used in traditional medicine for the treatment of gastrointestinal parasitosis in Burkina Faso.

ABSTRACT

Aims: to determine the antioxidant, anti-inflammatory and analgesic potential of aqueous decoctate (AD), aqueous macerate (AM) and hydroethanol macerate (HEM) of the plant.

Methodology: Thin-layer chromatography (TLC) was performed according to the method described by Hilderbert et al., 1996. In vitro evaluation of the antioxidant potential of the extracts was carried out using 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic) acid (ABTS), Ferric Reducing Antioxidant Power (FRAP) and lipid peroxidation (LPO) tests. In vivo anti-inflammatory activity was evaluated using the 1% carrageenan anti-edema test. The analgesic test was performed with 0.6% acetic acid.

Results: Phytochemical screening revealed the presence of tannins, saponosides, reducing compounds, coumarins and derivatives, anthocyanosides, steroids, triterpenes and flavonoids. The aqueous decoctate showed a lipid peroxidation inhibition rate of 48.30 ± 3.43%, respectively lower than hydroethanol macerate (58.08 ± 4.65%) and aqueous macerate (60.07 ± 4.52%). In the ABTS free radical scavenging test, hydroethanol macerate had an IC50 (35.92±5.04 µg/mL) lower than aqueous decoctate (44.75±1.04 µg/mL) and aqueous macerate (46.81±0.30 µg/mL) respectively. For the ferric ion reduction assay (FRAP), aqueous decoctate had the best reducing power of 1092.30± 18.50 Eqaa(µM/mL) respectively, compared with hydroethanolic macerate 957.99± 15.49 Eqaa(µM/mL) and aqueous macerate 716.13± 48.93 Eqaa(µM/mL). The carrageenan anti-inflammatory test, at a dose of 600 mg/Kg.b.w., gave an edema inhibition rate of 70.57% for the aqueous macerate, 73.07% for the aqueous decoctate and 75.56% for the hydroethanol macerate. Finally, the analgesic test at a dose of 600 mg/Kg.b.w. showed a contortion inhibition rate of 53.41% for the aqueous macerate, 60.80% for the aqueous decoctate and 69.32% for the hydroethanol macerate.

Conclusion: These results suggest that Gomphrena serrata is a plant with antioxidant, anti-inflammatory and analgesic properties that could alleviate the effects of inflammation during parasitic infections.

1. INTRODUCTION

Inflammation is a physiological response of defence or adaptation of the organism to aggression, which can be a microorganism or any particulate or soluble substance foreign to the organism (Pasquier, 1995). Recent pathophysiological studies indicate that there is a close relationship between pain and inflammation due to a bidirectional interaction between the neurosensory system and the immune system (Bertin and Vergne-Salle, 2019; Nko'o Moise et al., 2024). Also, parasitic infections such as helminthiasis manifest as chronic colitis (pain, obstruction, fever) with severe visceral lesions by eosinophilic granulomas (Rey et al., 1968). Furthermore, inflammation represents an immune reaction whereby immune system cells intentionally generate excess free radicals or reactive oxygen species (ROS), inducing oxidative stress and resulting in organ damage (Dieng et al., 2017; Mohammed et al., 2015). Free radicals are highly reactive unstable compounds with a single electron, namely the superoxide anion, hydroxyl radicals and hydrogen peroxide, singlet oxygen, and transition metals such as iron and copper (Cillard and Cillard, 2006; Inbathamizh et al., 2013). Free radicals attack and damage numerous cellular components such as proteins, lipids or DNA (Favier, 2003; Lobo et al., 2010). Indeed, the lipid peroxidation of lipoproteins, such as LDL, which are rich in cholesterol and phospholipids, is a primary factor in the development of chronic diseases, including atherosclerosis, neurodegenerative diseases, diabetes, cancer, inflammatory diseases, and ageing (Cillard and Cillard, 2006). To combat inflammation and the pain it induces, antioxidant substances would be best suited, as they have the advantage of capturing free radicals (Bene et al., 2017) , reducing and inactivating them (Siddhuraju and Becker, 2007). Plants also experience stress, both biotic and abiotic, and therefore produce free radicals (Garrett et al., 2006). In order to adapt to their environment, survive, develop and reproduce, plants synthesize antioxidants. Plants are therefore natural sources of antioxidants that protect them from stress (Sarr et al., 2015). In fact, all living organisms possess antioxidants and enzyme systems such as superoxide dismutase, catalase, glutathione peroxidase, and glutathione reductase to protect them from oxidative damage. However, these systems are not sufficient to entirely prevent and correct stress-related damage. Hence, the a need for antioxidant supplements or antioxidant-rich foods that can help scavenge free radicals and reduce oxidative damage (Shah and Modi, 2015). To treat diseases caused by oxidative stress, people turn to synthetic antioxidants and anti-inflammatories, such as non-steroidal anti-inflammatory drugs (NSAIDs) and steroidal anti-inflammatory drugs (SAIDs), which are the most widely sold drugs. However, the potential toxicological risks associated with the use of antioxidant and anti-inflammatory reference molecules (Lobo et al., 2010; Renfrey et al., 2003) and the high cost of health services and

drugs are driving a large proportion of the population to use medicinal plants for treatment (Agban et al., 2013) . Plants rich in phenolic compounds (flavonoids and tannins) are best suited to fight free radicals (Ipona et al., 2023). *Gomphrena serrata*, an anthelmintic medicinal plant, contains secondary metabolites with antioxidant potential such as flavonoids, tannins and saponosides (Ouedraogo et al., 2024) could play an important role in combating oxidation, inflammation and pain; therefore, it is necessary to investigate the antioxidant, anti-inflammatory and analgesic properties of extracts of this plant, namely aqueous macerate (AM), decoctate (AD) and hydroethanolic macerate (HEM).

2. MATERIAL AND METHODS

2.1 Materials

The material used in this study consists of biological material (plant and animal), technical equipment and chemicals.

2.1.1 Plant material

The plant material consists of lyophilized aqueous and hydroethanolic extracts of the whole plant of *Gomphrena serrata.*

2.1.2 Animal material

The animal material consists of Wistar rats, whose livers are used for the lipoperoxidation inhibition test, and NMRI mice, which are used for anti-inflammatory and analgesic studies. The animals used were obtained from the animal facility of the Institut de Recherche en Science de la Santé/Centre National de Recherche Scientifique et Technologique (IRSS/ CNRST), where the average temperature is 25 ± 2 ◦C with a relative humidity of 50 to 70%. The photoperiod is 12/24 hours. The diet consists of tap water and cereal pellets containing 29% protein.

2.1.2 Technical equipment

The technical equipment consists of apparatus and instruments:
A plethysmometer, a spectrophotometer, a computer, a water bath, syringes, cages, test tubes, micropipettes, tips, beakers, an Erlenmeyer flask, Falcone tubes, Eppendorf tubes, a rack, HPTLC plate.

2.1.3 Chemicals

Carrageenan, acetic acid, ABTS, NaCl, vanillin, Trolox, ascorbic acid, analytical ethanol, FeCl3, FeCl2, H2O2, AlCl3, HCl, KIO3, paracetamol, acetylsalicylic acid, potassium hexacyanoferrate, trichloroacetic acid, tannic acid.

2.2 Methods

2.2.1 Thin-layer chromatography

Phytochemical screening of Gomphrena serrata extracts was performed on HPTLC plates (10cm x 10cm) silica gel 60F254 (Merck, Darmstadt, Germany)(Alphonsine et al., 2019). Approximately 15 µL of each extract was deposited in 8 mm strips along the baseline 8 mm from the bottom edge of the plate using a semi-automatic sample dispenser (CAMAG, Linomat V, Switzerland). The distance between the spots is 3.4 millimetres. The distance between the initial spot and the left edge of the plate, as well as the distance between the final spot and the right edge of the plate, is 20 mm. Following deposition, the plate was placed in a tank containing the eluent (20 x 10 cm, saturation time: 30 minutes). The presence of sterols, triterpenes, flavonoids, coumarins, tannins, saponosides, and alkaloids was determined in accordance with the methodologies outlined by H. Wagner and S. Bladt (Hildebert Wagner, 1996). HPTLC profiles were primarily utilized to identify these chemical families.

Condensed tannin content (TTC)

The condensed tannin content (TTC) was determined using the acidified vanillin (HCl) method. This method is based on the reaction of vanillin with the terminal flavonoid group of condensed tannins, resulting in the formation of red complexes (Schofield et al., 2001). This complexation provides an explanation for the property of tannins to be transformed into red anthocyanidins by reaction with vanillin (Sun et al., 1998).

The determination of condensed tannins is conducted on the various plant extracts under investigation following the methodology delineated by Broadhurst (Broadhurst and Jones, 1978) and Heimler (Heimler et al., 2006) . To 0.5 ml of each suitably diluted sample or standard, 3 ml of vanillin solution (4%, m/v, in methanol) and 1.5 ml of concentrated HCl are added. Following a 20-minute reaction period, the absorbance of the reaction mixture is measured at 500 nm. Tannin concentrations are calculated from the calibration curve generated with catechin (0-1 mg/ml) and expressed in milligrams of catechin equivalent per gram of dry extract (µg CE/mg dry extract).

Hydrolyzable tannin content (TTH)

The content of hydrolyzable tannins (TTH) was determined.

TTHs were determined by a slightly modified version of the method described by Çam and Hişil (Çam and Hişil, 2010). One of the appropriately diluted extract was combined with five millilitres of 2.5% KIO3 in a test tube. Following a four-minute incubation period, the absorbance of the red-coloured mixture is read at 550 nm against a blank (distilled water). Different concentrations of tannic acid solutions (0 to 1 mg/ml) were utilized to establish the calibration curve. The final results are expressed in µg tannic acid equivalent per g dry extract (µg EAT/g ES).

2.2.2. Evaluation of Antioxidant Activity

2.2.2.1 ABTS Free Radical Scavenging Method

This test, which is based on the redox mechanism of ABTS (ammonium salt of 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)), was conducted following the methodology described by Arts (Arts et al., 2004) and Re et al. (Re et al., 1999). In this test, the ABTS salt undergoes an electron transfer reaction, forming a dark-coloured cation radical (ABTS+) in solution. In the presence of the antioxidant agent, the radical is reduced to a cation (ABTS+) discoloration of the solution and discolored. 19.2 mg ABTS was dissolved in 5 ml distilled water, and 3.312 mg potassium persulfate was added. The mixture was then stored in the dark at room temperature for 12 to 16 hours. Subsequently, a 4.5-ml volume of the mixture was diluted in 220 ml of analytical ethanol. Twenty microliters of varying concentrations of ethanolic and aqueous extracts or the reference substance (Trolox) were combined with 200 microliters of ABTS solution in a 96-well microplate. The mixture was incubated for 30 minutes at room temperature, and the absorbance was read at 415 nm with a Bio-Rad model 680 spectrophotometer (Japan). The blank was the solvent diluent of the extract or standard. The inhibition curve of absorbance versus extract or Trolox concentration was plotted to determine the 50% inhibitory concentration (IC50).

2.2.2.2 Ferric Reducing Antioxidant Power (FRAP) test

The FRAP method is used to determine the chelating capacity of metals, exclusively iron. It is based on reducing ferric ions (Fe^{3+}) to ferrous ions (Fe^{2+}). The extract's ability to reduce Fe^{3+} to Fe^{2+} by donating electrons is referred to as its reducing potential. Ferric iron, initially yellow, reduces to blue or green in the presence of an electron. The change in color from yellow to blue or green is proportional to the antioxidant activity.

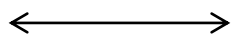


Diagram of Fe3+ reduction by an antioxidant

Reducing power was assessed using the spectrophotometric method described by Apati et al. (Apáti et al., 2003). To a test tube containing 0.5 mL sample solution (1mg/mL) is added 1.25 mL phosphate buffer (0.2 M, pH 6.6) followed by 1.25 mL potassium hexacyanoferrate [K 3 Fe(CN)6] 1% in water. The mixture is heated to 50°C in a water bath for 30 minutes. 1.25 mL trichloroacetic acid (10%) was ,added and centrifuged at 3000 rpm for 10 minutes. Three 0.625 mL aliquots were made in 3 tubes, to which 0.625 mL distilled water was added, followed by 0.125 mL freshly prepared 1% FeCl3 in water. A blank without a sample was prepared under the same conditions. Readings were taken at 700 nm against a Trolox standard curve (0.2-0.003 mg/ml). The concentration of reducing compounds (antioxidants) in the extract is expressed in mmol Trolox Equivalent (TE)/g dry extract according to the following formula:

$$C = \frac{c \times D}{M \times Ci} \times 1000$$

C = concentration of reducing compounds in mmol TE/g dry extract
c = sample concentration read
D = dilution factor of extract stock solution
Ci = concentration of extract stock solution
M = molar mass of Trolox (250 g/mol)

2.2.2.3 In vitro inhibition of lipid peroxidation

In vitro lipid peroxidation of rat liver homogenate is induced with ferric bichloride ($FeCl_2$) and hydrogen peroxide (H_2O_2). Peroxidation is inhibited in the presence of a substance with inhibitory activity.

The inhibitory activity of rat liver lipid peroxidation was determined using 2-thiobarbituric acid. $FeCl_2$-H_2O_2 was used to induce peroxidation of liver homogenate (207). 0.2 ml of each extract at the concentration of 1.5 mg/ml was mixed with 1 ml of 1% liver homogenate, then 50 µl of $FeCl_2$ (0.5 mM) and 50 µl of H_2O_2 (0.5 mM) were added. The mixture was incubated at 37°C for 60 minutes, then 1 ml trichloroacetic acid (15%) and 1 ml 2-thiobarbituric acid (0.67%) added, and the mixture was heated in boiling water for 15 minutes. Absorbance was read at 532 nm. Ascorbic acid was used as a reference product. Aqueous decoctate, aqueous macerate and hydroethanol macerate of *Gomphrena serrata* were used, and their ability to inhibit lipid peroxidation was expressed as percentage inhibition according to the following formula: Percentage inhibition (%) = [1 - (A1-A2)/A0x100].

A0 = absorbance of the control, A1 = absorbance with sample and A2 = absorbance without liver homogenate.

2.2.3 Study of anti-inflammatory activities

Carrageenan anti-edema test

Carrageenan is injected under the plantar fascia of the mouse's hind leg to provoke an inflammatory reaction, which can be reduced by any substance with anti-inflammatory properties (Winter et al., 1962).

Mice were fasted for 17 hours prior to testing. An injection of 0.05 mL of carrageenan (1% suspended in 0.9% NaCl) was made under the plantar fascia of the hind paw to induce oedema in the metatarsal region.

Batches of six mice were formed. The different batches were treated with either the plant drug or the reference substances one hour before carrageenan injection. The reference substance used was acetylsalicylic acid as an NSAID. The plant drugs used were *Gomphrena serrata* aqueous decoctate, aqueous macerate and hydroethanol macerate. The doses used for each extract were 200, 400 and 600 mg/kg (per os).

The volume of the treated paw was measured before and 1, 3 and 5 hours after the carrageenan injection. Variation in treated paw volume was used to assess the anti-inflammatory potency of each Gomphrena serrata extract. The mean oedema volume in the treated paw was calculated from 3 measurements of deviation not exceeding 4%. Anti-inflammatory activity was assessed as the percentage reduction in edema in treated rats versus blank controls, using the following formula:

$$\%\text{Inhibition} = \frac{A - B}{A} \times 100$$

A = mean difference in paw enlargement volume of white control mice;
B = mean difference in paw enlargement volume of treated mice.

2.2.3 Analgesic activity

Evaluation of analgesic activity using the acetic acid test

Intraperitoneal administration of an acetic acid solution (0.6%) to mice causes abdominal contortions. The number of contortions observed after administration of a pharmacological substance is used to assess its peripheral analgesic effect. The analgesic effect was assessed using the method described by Sawadogo et al.,2006 and Bhuiya et al., 2017. Batches of six (06) mice were formed: The white control batch received distilled water, the other batches received the extracts to be studied, and the positive control batch received a

reference substance, acetaminophen or paracetamol (200 mg/kg.b.w.). The different doses were administered orally to the mice according to their body weight. The doses used for each extract were 200, 400 and 600 mg/kg (per os).

One hour after extract administration, the animals received acetic acid intraperitoneally at a dose of 10 mL/kg. Five minutes after acetic acid injection, the number of contortions was counted in each mouse for 15 minutes. The analgesic effect was evaluated according to the following formula:

$$\%\text{inhibition} = \frac{1 - W_t}{W_b} \times 100$$

Wb = average number of contortions for mice in the white control group.
Wt = an average number of contortions of mice in the treated batch.

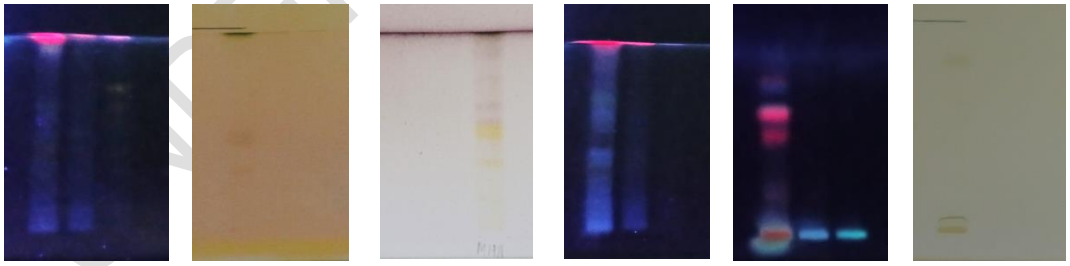
2.2.5 Statistical analysis

Means and standard deviations are obtained using Excel. Illustrative graphs, treatment comparisons and analysis of the significance of a dose effect and an effect are performed using GraphPad Prism 8 software. The effects of different doses on inflammation and pain were compared using Student's t-test. Differences are considered significant if the “p” value is less than 0.05 compared with the negative control group. n = 6. *P < 0.05; **P < 0.01; *** P < 0.001 and **** P < 0.0001.

3. RESULTS AND DISCUSSION

3.1 Results

3.1.1 Thin layer chromatography



Flavonoids:
Yellow
blue
brown, black
Yellow-green
Green
orange
purple

Tannin:
grey
brown

Saponins steroids:
Yellow
Saponins triterpenes:
pink-violet

Coumarins:
blue
green
purple

Triterpenes:
pink
orange
blue
Sterols :
brown
green

Alkaloi
Yellow
orange

Figure 1: TLC profile of some phytochemical compounds in hydroethanolic macerate (HEM), aqueous macerate (AM) and aqueous decoctate (AD).

Table1. Phenolic compounds assay

	EXTRACT		
CONTENT	HEM	AM	AD
TPC (mg EAG/g)	408,86±10,88	317,46±10,81	301,28±27,62
TF (mg/g ES)	27,36±0,49	11,49±0,95	10,11±0,60
CT (mg/g ES)	1,48± 0,04	16,86± 0,12	1,36±0,01
HT (mg/g ES)	88,01± 0,51	14,55± 0,27	2,34± 0,05

TPC= Total Phenolic Compounds, TF= Total Flavonoids, CT= Condensed Tannins, HT= Hydrolysable Tannins. AM=aqueous macerate, HEM=hydroethanolic macerate, AD=aqueous decoctate, mg GAE/g= milligrams Gallic Acid Equivalent, mg/gES= milligrams per gram of dry extract.

3.1.2 Assessment of antioxidant activity

Three methods were used to assess the antioxidant potential of the various extracts (Table 2). These included ABTS free radical scavenging activity, FRAP reduction of ferric ions to ferrous ions and rat liver lipid peroxidation inhibition activity (LPO test).

Table 2. Antioxidant activity

EXTRAIT	ABTS IC50(µg/mL)	FRAP Eqaa(µM/mL)	LPO(%inhib)
HEM	35,92±5,04	957,99± 10,96	58,08±4,64
AM	46,81±0,30	716,13± 48,93	60,07±4,52
AD	44,75±1,04	1092,30± 18,50	48,30±3,43
TROLOX	03,76 ± 0,41	-	-
Ascorb Ac	-	-	94,01± 0,07

CI50: Inhibitory concentration 50; AAEQ: Ascorbic acid equivalent; %inhib: Inhibition percentage; AM: Aqueous macerate; HEM: Hydroethanol macerate; AD: Aqueous decoctate.

2.1.3 Study of anti-inflammatory activities

Carrageenan anti-edema test

Figures 2, 3 and 4 show the mouse paw edema inhibition test results using *Gomphrena serrata* extracts. The results show a dose-dependent and time-dependent inhibitory effect of the three *Gomphrena serrata* extracts.

Table 3. anti-inflammatory activities

Extract (mg/Kg.p.c)		Edema volume			Inhibition percentage		
		1hour	3 hours	5 hours	1 hour	3 hours	5 hours
Control		0,47±0,02	0,53±0,02	0,69±0,02	-	-	-
AM							
	200	0,38±0,03	0,34±0,02	0,31±0,02	19,51	35,98	54,82
	400	0,35±0,03	0,31±0,04	0,26±0,02	25,73	41,21	61,09
	600	0,33±0,04	0,30±0,04	0,20±0,04	29,38	43	70,58
AD							
	200	0,38±0,02	0,37±0,01	0,31±0	20,09	30,02	54,1
	400	0,35±0,02	0,33±0,02	0,25±0,02	25,71	37,32	63,89
	600	0,31±0,03	0,26±0,02	0,17±0,01	35,49	50,42	73,07
HEM							
	200	0,33±0,02	0,29±0,02	0,26±0,02	30,67	45,71	63,1
	400	0,32±0,05	0,28±0,05	0,21±0,02	33,02	47,49	70,02
	600	0,28±0,02	0,24±0	0,17±0,01	40,07	54,08	75,56
ASA	100	0,24±0	0,16±0	0,10±0,01	47,82	69,25	85,05

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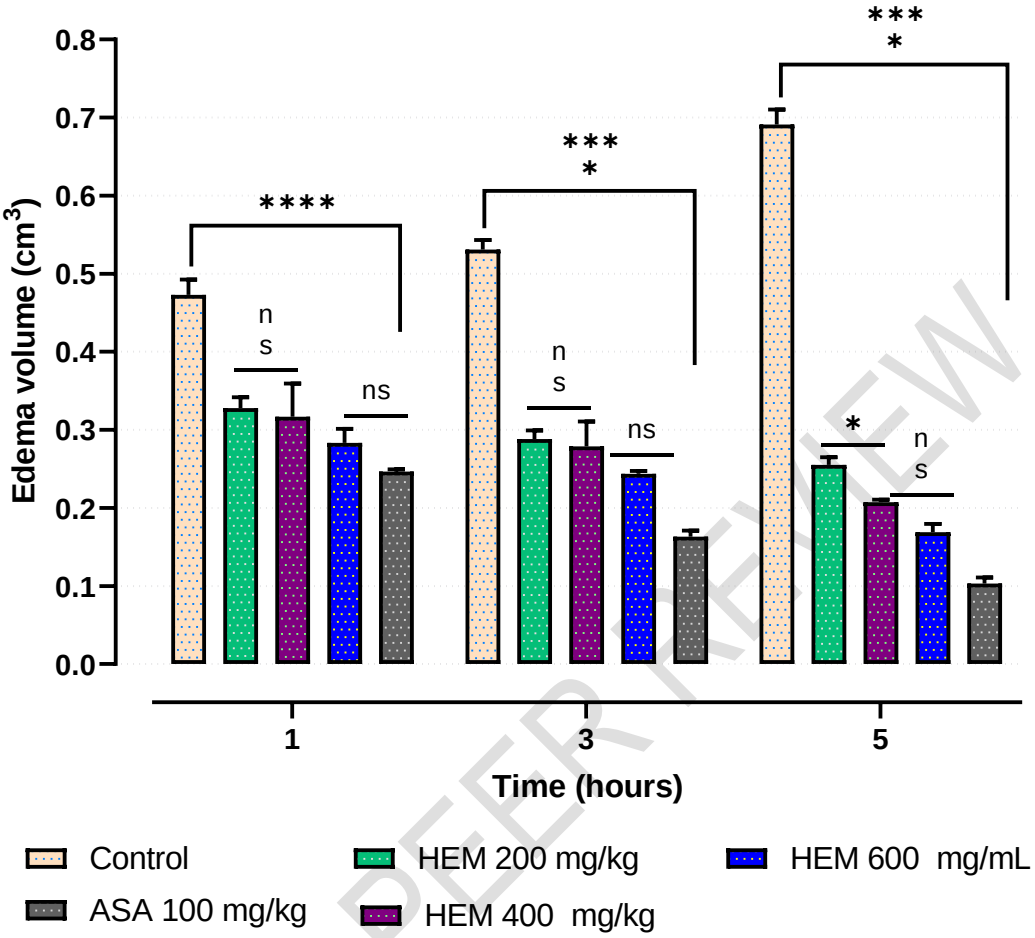

Figure. 2. inhibition of mice right paw volume at different doses of hydroethanolic macerate after 1h; 3h; 5h time after carrageenan injection.

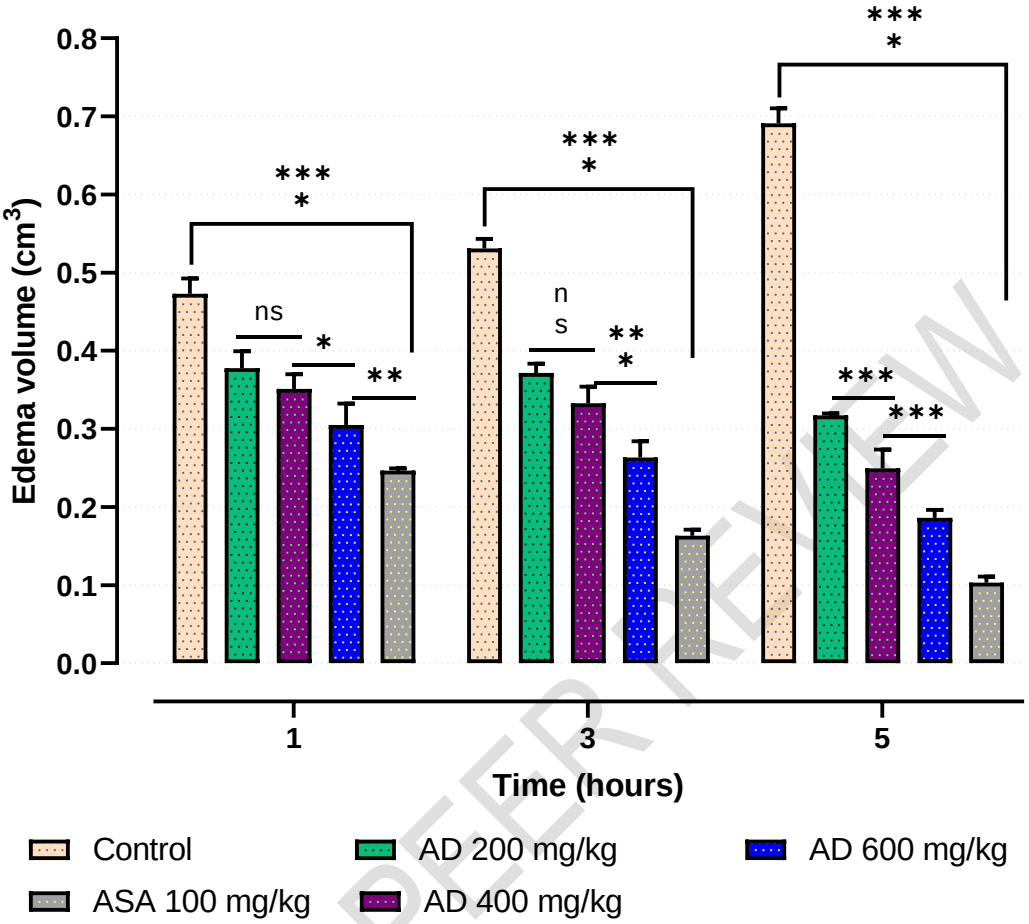

Figure 3. inhibition of mice right paw volume at different doses of aqueous decoctate after 1h; 3h, 5h after carrageenan injection.

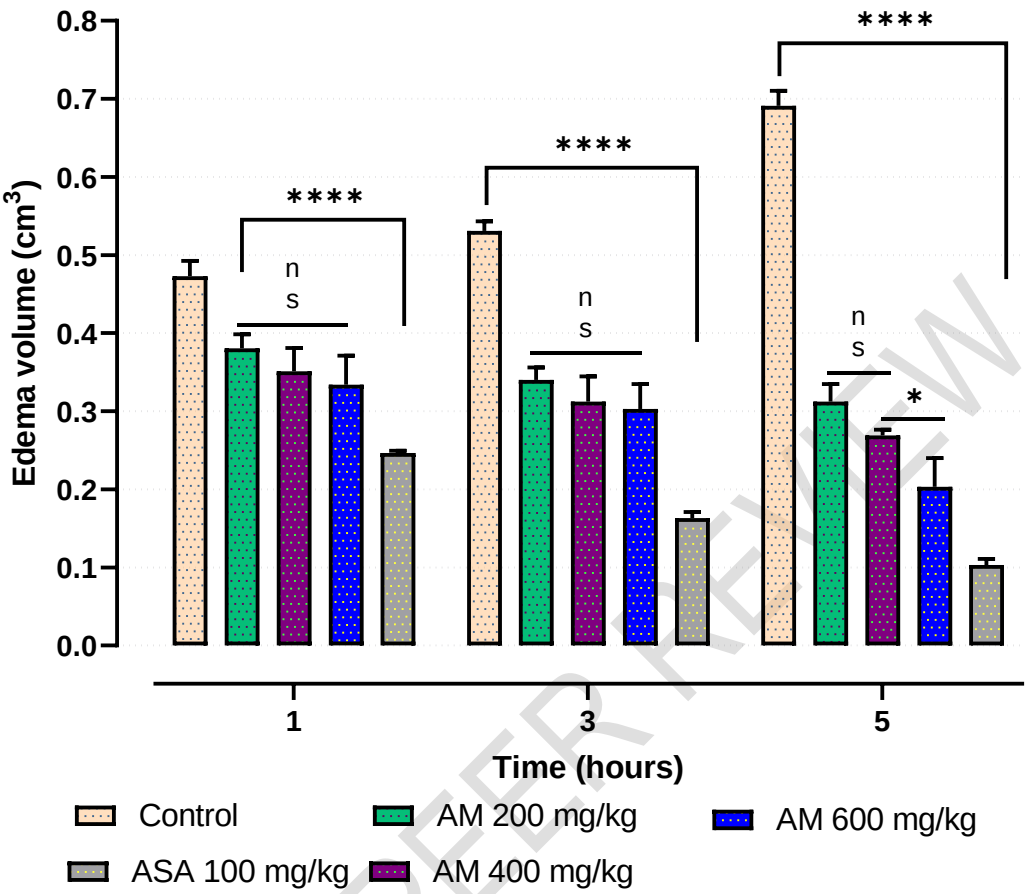

Figure. 4. inhibition of mice right paw volume at different doses of aqueous macerate after 1h; 3h; 5h time after carrageenan injection.

3.1.4 Analgesic activity

The results of an inhibition test on abdominal contortions in mice using *Gomphrena serrata* extracts are shown in figure 5. These results show a dose-dependent inhibitory effect of the extracts.

Table 4. Analgésic Test

Extracts and doses(mg/g)	Number of contortions	Percentage of inhibition
Contrôle	58,67±1,53	
AM		
200	42,33±0,58	27,84091
400	37±2	36,93182
600	27,33±2,52	53,40909

AD		
200	41±1	30,11364
400	27,67±2,3	52,84091
600	23±1,73	60,79545
HEM		
200	38,33±0,57	34,65909
400	25±0	57,38636
600	18±0	69,31818
Paracétamol 200	16±1	72,72727

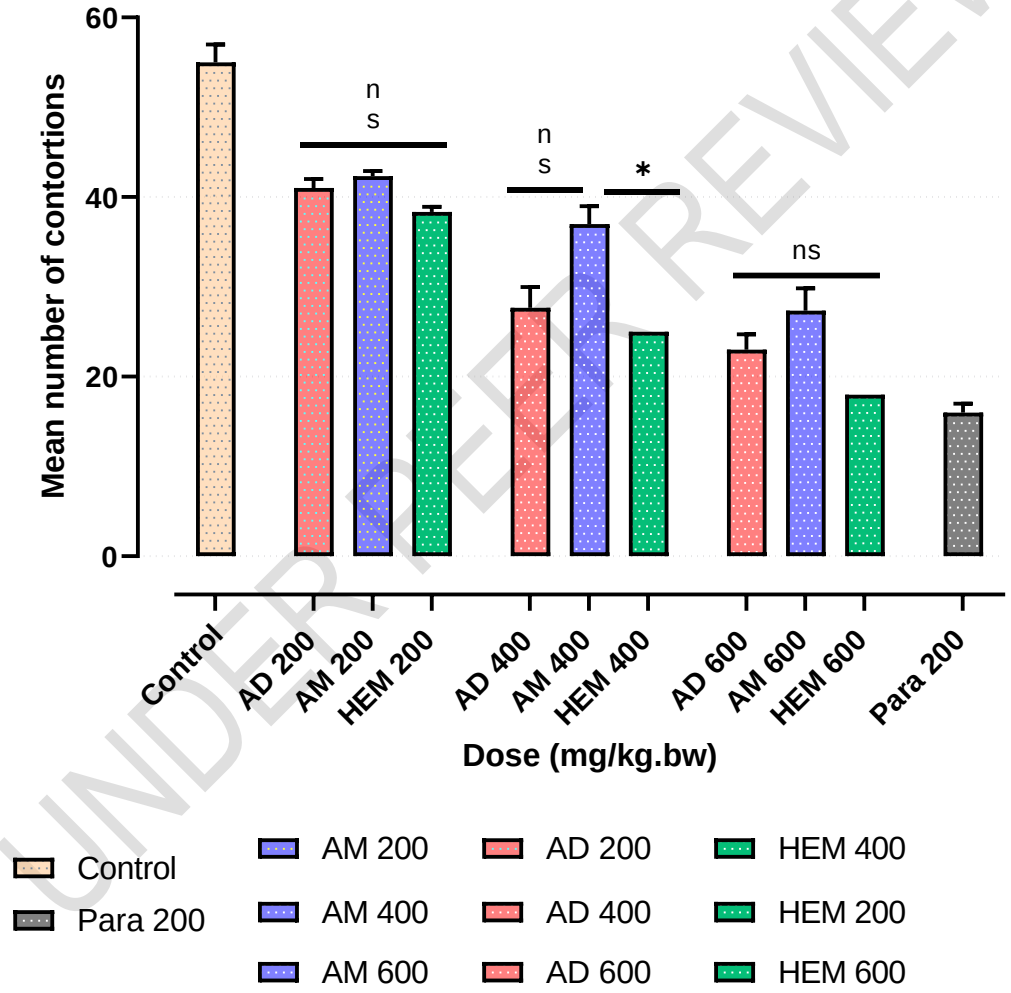

Figure. 5. Number of contortions in mice at different doses of *G. serrata* extracts.

3.2 Discussion

Thin-layer chromatography revealed the presence of bioactive secondary metabolites of interest, including coumarins, saponosides, sterols and triterpenes, phenolic compounds such as tannins and flavonoids. These results corroborate those reported by (Ouedraogo et al., 2024) on *Gomphrena serrata*.

Concerning ABTS antioxidant activity, the results showed that the HEM of *Gomphrena serrata* has a lower IC50 than those of AD and AM. However, the free radical scavenging activity of all three *Gomphrena serrata* extracts using the ABTS method was well below that of the reference compound, Trolox (03.76 ± 0.41 µg/mL). As far as inhibition of lipid peroxidation is concerned, only AM and HEM have an inhibition percentage greater than 50%, although these are much lower than that of the reference substance, ascorbic acid. Hydroethanol macerate and aqueous macerate, therefore have a proven capacity to inhibit lipid peroxidation. Based on these 3 tests, we can confirm that Gomphrena serrata has evident antioxidant activity across all three extracts. This could be explained by the presence of phenolic compounds in the plant (Oracz and Nebesny, 2016). Polyphenols (flavonoids and tannins) are powerful antioxidants capable of preventing and regulating free radical damage while acting as free radical garbage cans (Bene et al., 2017; Ipona et al., 2023). To prevent oxidation, chelators form complexes with metals, inhibiting the metal's redox cycle (Cillard and Cillard, 2006). Flavonoids such as anthocyanins, catechins, flavones, flavonols, isoflavones and proanthocyanidins are metal chelators, superoxide anion scavengers and hydrogen donors. Inhibition of lipoperoxidation by *Gomphrena serrata* extracts prevents alteration of the functionality of cell membranes, which are particularly rich in polyunsaturated fatty acids (30-50%) present in phospholipids, sphingolipids and cardiolipins (Nakagawa, 2004).

With regard to inhibition of carrageenan-induced edema (1%), all three Gomphrena serrata extracts proved effective at a dose of 600mg/Kg b.w., especially at the fifth hour, with inhibition percentages of over 70%, with a better inhibition percentage obtained with the hydroethanolic macerate (75.56%). However, these inhibition percentages are lower than those of acetylsalicylic acid (85.05% at a dose of 100mg/Kg b.w.), a non-steroidal anti-inflammatory effective against carrageenan oedema. The anti-inflammatory effect is due to polyphenols, which neutralize pro-inflammatory substances such as histamine, serotonin, bradykinin and prostaglandins.

The inflammatory reaction induced by carrageenan is biphasic. The first or initial phase, which occurs between 0 and 2.5 hours after injection of the phlogogenic agent, is due to the action of histamine, serotonin and bradykinin on vascular permeability (Linardi et al., 2000).

The second or late phase, which occurs after the 3rd hour and can last beyond the 6th hour, is also a complement-dependent reaction and results from an overproduction of prostaglandins in the tissues (Di Rosa, 1972). Our three extracts significantly inhibited both phases of inflammation. This suggests that *Gomphrena serrata* extracts contain flavonoids, which can inhibit arachidonic acid-metabolizing enzymes such as phospholipase, cyclooxygenase, and lipoxygenase. This, in turn, blocks the production of various chemical mediators of inflammation, including histamine, serotonin, bradykinin, and prostaglandins (Emeraux, 2021; Ouédraogo et al., 2012). Regarding analgesic effect, all three Gomphrena serrata extracts significantly reduced abdominal contortions induced by i.p. injection of acetic acid (0.6%) in a dose-dependent manner. The percentage inhibition of contortions at a dose of 600 mg/Kg body weight was 69.32% for HEM, higher than for AD (60.80%) and AM (53.41%). However, these inhibition rates are significantly lower than those of the reference substance, paracetamol (72.73%), at a dose of 200 mg/kg bw. Painful contractions are due to the release of chemical mediators that stimulate peripheral nociceptive neurons and induce increased vascular permeability. The chemical mediators responsible for nociception are serotonin, histamine, bradykinin and prostaglandins (PGE2α, PGF2α), which stimulate peritoneal nociceptive receptors (Deraedt et al., 1980; Negus et al., 2006; Reanmongkol et al., 2009; Vanderlinde et al., 2009). This analgesic effect of *Gomphrena serrata* extracts could be linked to the action of flavonoids, tanins and saponosides in inhibiting the release of certain nociceptive chemical mediators such as prostaglandins (Ior et al., 2011). The reduction in the number of contortions could be explained by the plant's peripheral analgesic effect through inhibition of prostaglandin synthesis(Asrafuzzaman et al., 2024).

4. CONCLUSION

Our study first confirmed the richness of *Gomphrena serrata* in metabolites such as tannins, saponosides, reducing compounds, coumarins and coumarin derivatives, anthocyanosides, steroids, triterpenes and flavonoids. It then showed that Gomphrena serrata has antioxidant, anti-inflammatory and analgesic properties. *Gomphrena serrata* hydroalcoholic macerate proved more effective than aqueous macerate and aqueous decoctate in inhibiting inflammation and pain. The traditional use of Gomphrena serrata leaves as an antioxidant, anti-inflammatory, and analgesic seems justified.

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COMPETING INTERESTS

The authors declare no conflicts of interest regarding the publication of this paper.

AUTHORS' CONTRIBUTIONS

CONSENT

It's not applicable

ETHICAL APPROVAL

All protocols in this study were approved by the institutional committee on the ethics of animal experiments of the institut de recherche en sciences de la sante (IRSS/CNRST), ouagadougou, burkina faso.

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APPENDIX