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Phytochemical analysis, *in vitro* antioxidant and *in vivo* anti-inflammatory activities of a hydroethanolic extract of *Zingiber officinale* Roscoe rhizome

Análisis fitoquímico, actividad antioxidante *in vitro* y antiinflamatoria *in vivo* de un extracto hidroetanólico de rizoma de *Zingiber officinale* Roscoe

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ABSTRACT

This present study aims to evaluate the *in vitro* antioxidant and the *in vivo* anti-inflammatory activities of the ethanolic extract of *Zingiber officinale* Roscoe rhizome. The extract is obtained by maceration (80 %). The analysis of total phenolic and flavonoid contents showed that the ethanolic extract is rich in polyphenols (206.95 ± 11.78 µg equivalent gallic acid) and flavonoids (11.62 ± 0.00 µg equivalent quercetin/mg dry extract). According to the 1-diphenyl-2-picrylhydrazyl assay, the ethanolic extract exhibits an important antioxidant activity with an IC_{50} of 0.11 ± 0.00 mg/mL. Furthermore, the ethanolic extract demonstrates a higher reducing power compared to the butylated hydroxytoluene, with IC_{50} values of 0.01 ± 0.00 mg/mL. The ethanolic extract shows the highest inhibition in the β-carotene assay, with an inhibition of 82 %. The two induced inflammation tests, Xylene-Induced Ear Edema and Carrageenan-Induced Paw Edema, showed that the ethanolic extract of *Zingiber officinale* produced a remarkable anti-edematous effect, with a dose of 400 mg/kg for Xylene-Induced Ear Edema and a dose of 200 mg/kg for Carrageenan-Induced Paw Edema. In conclusion, the rhizome extract of *Zingiber officinale* has demonstrated both antioxidant and anti-inflammatory properties, as can be justified by its traditional uses.

Key words: *Zingiber officinale* Roscoe; anti-inflammatory; antioxidant; phenolic compounds.

RESUMEN

Este estudio tiene como objetivo evaluar las actividades antioxidantes *in vitro* y antiinflamatorias *in vivo* del extracto etanólico del rizoma de *Zingiber officinale* Roscoe. El extracto se obtiene por maceración (80 %). El análisis de los contenidos totales de fenólicos y flavonoides mostró que el extracto etanólico es rico en polifenoles (206.95 ± 11.78 µg de ácido gálico equivalente) y flavonoides (11.62 ± 0.00 µg de quercetina equivalente/mg de extracto seco). Según el ensayo 1-difenil-2-picrilhidrazilo, el extracto etanólico exhibe una importante actividad antioxidante con una CI_{50} de 0.11 ± 0,00 mg/mL. Además, el extracto etanólico demuestra un mayor poder reductor en comparación con el hidroxitolueno butilado, con valores de CI_{50} de 0.01 ± 0.00 mg/mL. El extracto etanólico muestra la mayor inhibición en el ensayo de β-caroteno, con una inhibición del 82 %. Las dos pruebas de inflamación inducida, Edema de Oído Inducido por Xileno y Edema de Pata Inducido por Carragenina, mostraron que el extracto etanólico de *Zingiber officinale* produjo un notable efecto antiedematoso, con una dosis de 400 mg/kg para Edema de Oído Inducido por Xileno y una dosis de 200 mg/kg para Edema de Pata Inducido por Carragenina. En conclusión, el extracto de rizoma de *Zingiber officinale* ha demostrado propiedades antioxidantes y antiinflamatorias, como lo justifican sus usos tradicionales.

Palabras clave: *Zingiber officinale* Roscoe; antiinflamatorio; antioxidante; compuestos fenólicos.

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INTRODUCTION

Oxidative stress is a proven biological phenomenon that takes place as a consequence of an overproduction of reactive oxygen species (ROS) in either cells or tissues. This leads to macromolecular damage from oxidation and the disruption of gene regulation, ultimately resulting in cell death [1 , 2 , 3].

At controlled levels, ROS act as signaling molecules that mediate plant responses to biotic and environmental stresses by modulating physiological functions and participating in diverse metabolic pathways [4 , 5]. However, their accumulation may lead to the formation of pro-inflammatory molecules, contributing to inflammation and tissue injury, which can ultimately result in chronic inflammatory diseases [6 , 7].

In order to counteract these harmful consequences, antioxidants of natural origin, in particular those derived from medicinal plants with bioactive compounds capable of modulating oxidative stress and inflammation, have received considerable interest from scholars in recent years [8 , 9].

In fact, for a long time in the history of medical practices, plants have been used as remedies for many diseases, with secondary metabolites playing a vital role as precursors for numerous drugs and pharmaceuticals in modern medicine [10 , 11].

Under this group, *Zingiber officinale (Z. officinale)*, generally known as ginger in English or “gingembre” in French [12 , 13].

In Asian countries such as India, was used as a spice for over two thousand years and as a flavoring agent throughout antiquity. It is a seasonal, herbaceous, erect plant with a pseudo-stem growing to a height of 0.3 to 1 meter. Its leaves have hairy petioles with lengths of 2 to 4 mm, with leaves themselves measuring 15 to 23 cm in length and 8 to 15 mm in width. Its morphological characteristics include roots, stems, rhizomes, leaves, and flowers. Its rhizomes can last for long in the soil and display the capability to produce new shoot growth to compensate for shed leaves and stems [14].

Its rhizome is flat with a pale-yellow interior and is strongly fibrous. Its roots grow from the junction at the lower side of the rhizome, with shoots developing at the top. Varieties of ginger are different in shape and have differences in taste as well [14].

Ginger (*Zingiber officinale,* Roscoe) has bioactive compounds such as shogaol, gingerol, and flavonoids, making it a natural source of various secondary compounds and an antioxidant-rich plant (FIG. 1), [15 , 16].

It has traditionally been used in the prevention and treatment of stomach-upsetting dilemmas such as dyspepsia, intestinal-derived ailments, and various types of food poisoning [17].

Ginger has also been proven useful in combating arthritic diseases, aiding in transport-related complaints, and reducing pregnancy-related vomiting. Raw ginger is richest in enzymes that aid digestion and has traditionally been used as an antidote for burned skin [16 , 18 , 19].

FIGURE 1. *Zingiber officinale* rhizome

The increase in body warmth has further been linked to strengthened circulation and a possible reduction in blood pressure [20]. Zingiberene is primarily identified as the main component found in *Z. officinale* essential oil (19.71 %), accompanied by (+)-β-cedrene, farnesene, α-curcumene, and β-elemene in larger amounts [21].

The major active and stable phenolic compounds found in the active component of ginger, such as shogaols, paradols, zingerone and gingerols, have been known to exhibit antioxidant, anti-angiogenic, anti-atherosclerotic, and anticancer activities as suggested by previous studies [22 , 23].

Taking into consideration the aforementioned factors, the aim of this present study is to evaluate the *in vitro* antioxidant, *in vivo* anti-inflammatory activity, as well as the total phenolic compound content of *Z. officinale.*

MATERIALS AND METHODS

Plant material

Dried rhizomes of *Z. officinale* were purchased from a local market and ground to a fine light-yellow powder. The powder was prepared immediately before extraction.

Extraction procedure

With a few minor adjustments, the plant’s ethanolic extract was prepared [24]. In short, 85 % ethanol was used to extract the dried powder of *Z. officinale* for 72 hours (h) at room temperature. After filtering the resultant mixture, the filtrate was evaporated (Buchi rotavap R-205, Switzerland), at 45 °C. The pharmacological characteristics of the dried extract were examined.

Total phenolic contents

The total phenolic content was measured using a modified Folin-Ciocalteu (FC) assay, as described previously [25]. In summary, a 0.1 mL aliquot of plant extract or standard was mixed with a 0.5 mL aliquot of FC reagent, which was previously diluted 1:10. After a 4-min reaction time, a 0.4 mL aliquot of a 7.5 % sodium carbonate solution was added. Absorbance was then measured (Shimadzu UV1800 spectrophotometer, Germany), at 765 nm, and total phenolics were calculated as gallic acid (GA) equivalents (FIG. 2).

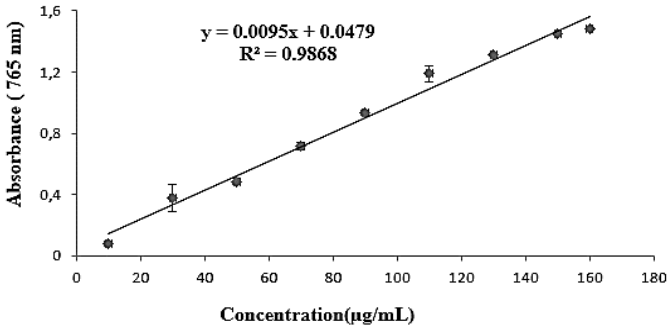

FIGURE 2. Standard curve for gallic acid.

Total flavonoid contents

Total flavonoid content in each of the plant extracts was determined using the aluminum chloride ($AlCl_3$) colorimetric assay [26]. A detailed description of this method involves mixing 1 mL of extract with 1 mL of 2 % $AlCl_3$ in methanol, which is then allowed for 10 minutes (min) of incubation, with measurements conducted at 430 nm to calculate in terms of quercetin equivalent, (FIG. 3).

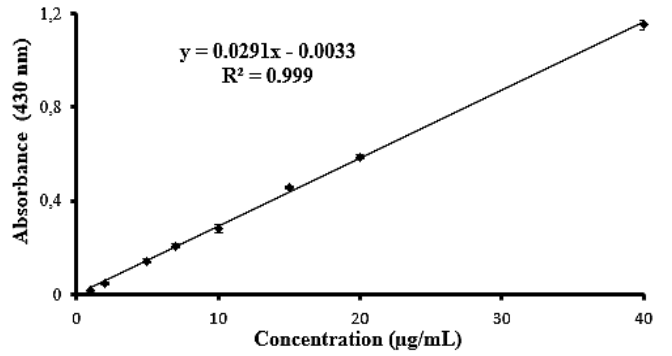

FIGURE 3. Standards curve for quercetin.

Antioxidant potentials

1-diphenyl-2-picrylhydrazyl radical scavenging assay

The scavenging capacity of extract was determined using the 1-diphenyl-2-picrylhydrazyl (DPPH) assay, according to the method of Burits and Bucar [27]. Briefly, a mixture of each extract (0.05 mL) and a solution of DPPH (1.25 mL, containing 0.004 % DPPH in methanol) was used, with butylated hydroxytoluene (BHT) serving as the positive control. After 30 min of incubation, the absorbance was measured at 517 nm. The scavenging activity was estimated from the relevant formula.

$$\text{Scavenging activity} = \frac{A_C - A_S}{A_C} \times 100$$

It expresses further the antioxidant activity as IC_{50} (mg/mL), where : is the absorbance of the control and : is the absorbance of the sample

β-carotene bleaching assay

Antioxidant activities were evaluated using the ability to suppress the accumulation of conjugated dienes and volatile oxidation products of linoleic acid, based on the method of Dapkevicius *et al.* [28]. Briefly, a mixture of 0.5 mg of β-carotene in 1 mL of Chloroform was prepared, to which were added 200 mg of Tween-40 and 0.025 mL of Linoleic acid. Oxygen-saturated distilled water (100 mL) was added after complete evaporation of the Chloroform at 40 °C to achieve the emulsion.

To each test tube pre-loaded with 0.350 mL of sample solution (2 mg/mL), 2.5 mL of the aforementioned emulsion was added. All were kept at room temperature in the darkness. Positive controls used were GA and BHT. Absorbances were recorded at 490 nm at intervals of 0, 1, 2, 3, 4, 6, 12, and 24 h. Relative antioxidant activities (A %) was estimated using the following formula:

$$\text{A\%} = \frac{\text{AS}}{\text{AC}}\ x\ 100$$

where and represent the absorbances of the control and sample at , respectively.

Reducing power assay

The previously published approach was used to determine the extract's reducing power [29]. Briefly, 0.1 mL volumes of the methanol-facilitated extracts, $K_3Fe(CN)_6$, and 0.2 M phosphate buffer solution, pH 6.6, were allowed to react. After a 20-min incubation period at 50 °C, the reactions were stopped through the addition of 0.25 mL volumes of 1 % trichloroacetic acid. About 0.25 mL volumes were mixed with 0.25 mL volumes of distilled water and 0.5 mL volumes of 0.1 % $FeCl_3$ solution. The absorbance was measured (Shimadzu UV1800 spectrophotometer, Germany) at 700 nm.

Anti-inflammatory capacities

Xylene induced ear edema

As outlined by the process designed by Atta and Alkohafi [30], the Xylene-induced edema of the ears was performed using Xylene. The study involved four groups with six mice (*Mus musculus*) each. Edema was induced by the topical application of 40 µL Xylene to the outer part of the ears.

The first group was the standard treatment and was given 100 mg/kg of indomethacin. The second group was the negative control and given NaCl solution (0.9 %) as treatment, while the third and the fourth groups received the plant extract with a dose of 200 and 400 mg/kg, respectively. The ears were measured using the digital caliper (caliper to DIN 862, Germany) before and two hours after the induction of the inflammation. The percentage inhibition of the inflammation or the edema was measured using the formula:

$$I\% = \left(\frac{\text{DControl} - \text{DTreated}}{\text{DControl}}\right) x\ 100$$

Where $D_{Control}$ is the difference in edema thickness in the control group, and $D_{Treated}$ is the difference in edema thickness for the treated group.

Carrageenan-induced rat paw edema

In this experiment, four groups of adult mice (each consisting of n = 6) were administered diclofenac at doses of 50 mg/kg, ginger extract at doses of 200 and 400 mg/kg, and the negative control, which included the administration of 0.9 % NaCl solution. Before the treatment, the mice had free access to drinking water. The animals were made to fast for 12 h, after which the test subjects received compounds orally via gavage. Acute inflammation was elicited by the subplantar injection of 0.02 mL of 1 % w/v carrageenan in normal saline into the right hind paw of the test subjects, thirty min following treatment, as done in the research study conducted by Belay and Makonnen [31] on the model mentioned above. The measurement of the paw thickness for each test subject involved using the digital caliper. The percentage of paw swelling was estimated as follow:

$$I\% = \left(\frac{Dt - D0}{D0}\right) x\ 100$$

D0 is the paw thickness, and Dt is the paw thickness after carrageenan injection at a given time point.

Statistical analysis

GraphPad Prism, ver. 6.01 for Windows, was applied for statistical analysis. Values are estimated as mean ± SD. For the antioxidant test, we used t-test cause wa have only two samples, the rest we used anova because we have more then two samples.

RESULTS AND DISCUSSION

Yield, total phenolic and flavonoid contents

The focus of this study is the determination of the phytochemical constitution of a hydroethanolic extract (HEE) of the rhizome of *Z. officinale*, specifically in terms of the total phenolic and flavonoid contents using the spectrophotometric method. The estimate of yield in relation to the total powder weight (100 g) of plant, shows that the HEE present a 7.4 % of yield (TABLE I). The total phenolic and flavonoid contents of the sample were 203.12 ± 06.95 µg GAE/mg extract and 11.62 ± 0.00 µg QE/mg extract, respectively. In Comparison, Lukiati *et al.* [32] reported a phenolic content of 155.78 µg GAE/mg extract, indicating a higher phenolic content in the current study. The results are presented in TABLE I. As for flavonoid content, the results obtained are similar to the findings of Bekkouch *et al*. [33]. Previous investigations have revealed that *Z. officinale* is rich with various phenolic compounds, including 6-paradol, 6-shogaol, 6-gingerol, methyl 6-gingerol, 8-gingerol, 5-gingerol, 1-dehydro-6-gingerol and 10-gingerol, [34]. In general, Variations in total phenolic content (TPC) and total flavonoid content (TFC) values in plant extracts may be attributed to the presence of non-flavonoid polyphenols like phenolic acids and tannins. Rhizomes of *Z. officinale* contain major bioactive compounds gingerols and shogaols; they are polyphenols but not flavonoids. This may be a plausible reason for the higher values of TPC over TFC in the HEE. Variations in the type of solvents used for the extraction process may affect the polyphenol and flavonoid contents. Hydroethanolic solvents tend to increase the recovery of various polyphenolic compounds.

TABLE I
Yield, total polyphenols and flavonoids contents of *Z. officinale* extract.

Extract	Yield (%)	Total phenolic content (a)	Total flavonoids content (b)
GEE	7.4	203.12 ± 06.95	11.62 ± 0.00

(a): µg gallic acid equivalent (GA)/mg dried extract (DE), (b): µg quercetin equivalent (QE)/ mg dried extract (DE), GEE: ginger ethanol extract

Generally, 6-gingerol is known to be more prevalent in fresh forms, whereas 6-shogaol is more prevalent in dried forms of ginger, attributed to the dehydrating properties of gingerols. The disparity in concentrations of 8-gingerol and 10-gingerol may also be attributed to variations in extraction and stability. These compounds are known to be the main bioactive markers for ginger and are believed to be responsible for its antioxidant and anti-inflammatory properties. Thus, the quantitative analysis of these compounds is essential for standardization, quality, and efficacy of ginger-based pharmaceuticals for uniform therapeutic effects.

Antioxidant activity

2,2-Diphenyl-1-picrylhydrazyl free radical scavenging assay

In vitro antioxidant tests are used extensively as a first step to screen plant extracts for the ability to scavenge free radicals. Among these tests, the DPPH radical-scavenging assay is the most frequently used technique for its simplicity, speed, and reproducibility. The DPPH free radical has been shown to be a stable nitrogen-centered radicals, which changes color to yellow upon reduction by an antioxidant, hence permitting quantitative measure of the radical-scavenging ability. The amount of extract needed to scavenge 50 % of the DPPH radicals has been shown to serve as an indicator for the potency of the antioxidative effect. The extract showed a clear dose-dependent ability to scavenge DPPH radicals, with an IC_{50} of 0.12 ± 0.00 mg/mL. The results are presented in TABLE II. While this level was significantly lower than the standard antioxidant: BHT, ($P < 0.0001$), which had an IC_{50} of 0.01 ± 0.00 mg/mL, the extract still proved its capacity to neutralize free radicals. The IC_{50} value observed is considerably lower than that reported by [35] (IC_{50} = 4.25 mg/mL).

TABLE II
Antioxidant activity of hydroethanolic *Z. officinale* extract

	Antioxidant activity/Inhibition concentration (IC_{50})	
Extract/ standards	**DPPH radical scavenging**	**Reducing power**
GEE	26.06 ± 0.00***	39.43 ± 0.00***
BHT	87.26 ± 0.00	ND
Vitamin C	ND	21. 91 ± 0.48

GEE: ginger ethanol extract. BHT: butylated hydroxytoluene

The HEE of *Z. officinale* rhizomes had a strong concentration-dependent ability to scavenge the DPPH radicals. The higher the concentration of the extract, the higher the ability to scavenge the radicals, thereby indicating a strong ability to donate hydrogen or electron density to scavenge the radicals. The low IC_{50} value obtained depicts that only a low concentration of the

extracts is required to scavenge 50 % of the DPPH radicals, thus signifying a high ability to scavenge free radicals.

This remarkable activity is likely linked to the extract's high phenolic content, consistent with previous studies highlighting the pivotal role of phenolic compounds in free radical neutralization [35].

This strong antioxidative property can be attributed to the high phenolic content presented in the extract. Phenolics, such as the gingerols present in *Z. officinale*, are known to tame the free radicals by the phenomenon of resonance. This is because the phenolics prevent the oxidation reaction by the free radicals. This concentration-dependent property also confirms the presence of the bioactive compounds, as the concentration increases the amount of the antioxidative molecules that can interact with the DPPH molecules [36]. Taken together, the high DPPH scavenging capacity shown by the HEE of the *Z. officinale* rhizome reported in the present study supports the traditional use and provides further rationale to the reported findings demonstrating the antioxidant role of ginger phenolic compounds. Such antioxidant activity may be shown to be responsible for the anti-inflammatory activity exhibited by the HEE.

Reducing power assay

The extract exhibited a strong reducing power, with an IC_{50} of 0.013 ± 0.00 mg/ml, a result comparable to that of the standard BHT, measured at 0.0012 ± 0.00 mg/ml. This suggests that the extract has a strong ability to donate electrons and can effectively reduce Fe^{3+} to Fe^{2+}. Comparative study on different ginger plant parts noted that ethanolic extracts excel in electron transfer-based antioxidant assays, such as ferric reducing power, which highlights the contribution of phenolic compounds to reducing capacity [37 , 38].

Overall, the DPPH, β-carotene-linoleic, and reducing power results show that Zingiber officinale's ethanolic extract has potent antioxidant activity. This effect largely stems from its high levels of phenolic and flavonoid compounds such as shogaol, gingerol, zingerone, and paradol. These compounds work synergistically to reduce the production of ROS. Ginger helps boost glutathione levels, stops lipid peroxidation, inhibits the generation of nitric oxide, scavenges hydroxyl radicals, promotes the activity of antioxidant enzymes like superoxide dismutase and catalase, and lowers the expression of inducible nitric oxide synthase, further supporting its role as a powerful antioxidant [39].

β-carotene/linoleic acid bleaching assay

In the β-carotene–linoleic acid assay, antioxidants inhibit the formation of volatile compounds and conjugated diene hydroperoxides that result from linoleic acid oxidation [40]. The ethanolic extract of ginger showed a strong result in preventing lipid peroxidation, with 82 % inhibition. This result is comparable with BHT, which reached an inhibition of 99, and is significantly higher than GA with 46 % inhibition (FIG. 4).

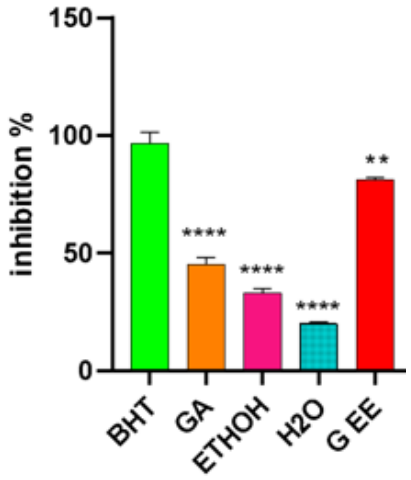

FIGURE 4. The inhibition percentage of *Z. officinale* ethanolic extract was evaluated by β-carotene-linoleic acid assay. Samples were analyzed after 24 hours, Positive controls used were GA and BHT. Data are presented as mean ± SD (n = 3). *P < 0.05 statistically significant, ***P < 0.0001. GEE: *Z. officinale* ethanol extract.

The β-carotene-linoleic acid assay is one common method that has been widely accepted for the determination of the antioxidant activity of plant extracts using their ability to block the oxidation of lipids. In the β-carotene-linoleic acid assay, the linoleic acid undergoes an oxidative breakdown when heated in the presence of oxygen, forming free radicals and conjugated diene hydroperoxides. The highly unsaturated molecules of β-carotene are attacked by the reactive oxidation product. The results obtained align with previous studies reporting that ethanolic plant extracts protect tissues against lipid peroxidation *in vivo* and effectively scavenge reactive oxygen species in lipid rich environments [41 , 42].

Xylene induced ear edema

Inflammation serves as the body's defense system to get rid of infections, burns, and toxins, as well as to initiate the healing process. According to Ayustaningwarno *et al*. [43], it results in discomfort, swelling, heat, redness, and disruptions of physiological processes. Unchecked acute inflammation develops into chronic inflammation, which leads to chronic inflammatory disorders [44]. This leads to the considerable production of inflammatory mediators, including NO, PGE2, TNF-, IL-1, IL-6, and COX-2, which are linked to the pathophysiology of tissue damage [45].

Many inflammatory diseases are characterized by excessive activity of macrophages and the formation of free radicals that cause tissue destruction [46].

Topical administration of xylene induces acute edema in mice's ears, which serves as a widely accepted model for evaluating anti-inflammatory activity [6 , 47]. Xylene is known to trigger the release of inflammatory mediators, leading to vasodilation and increased vascular permeability, resulting in edema formation [47]. In the present study, ear edema induced using xylene and treated with ethanolic extract of *Z. officinale* at doses of 200 and 400 mg/kg resulted in significant edema reduction measured at 57 and 54 %, respectively (FIG. 5). These results are comparable

with those obtained using the well-known anti-inflammatory drug, indomethacin, with 76% reduction of edema. Zhang *et al*. [20] also demonstrated the anti-inflammatory efficiency of *Z. Officinale*'s oils compared to ibuprofen. According to previous investigations, the beneficial effects are largely attributed to bioactive compounds like 6-shogaol, which play a role in modulating inflammatory mediators such as COX-2, INOS, NF-κB, and MAPK signaling pathways [48].

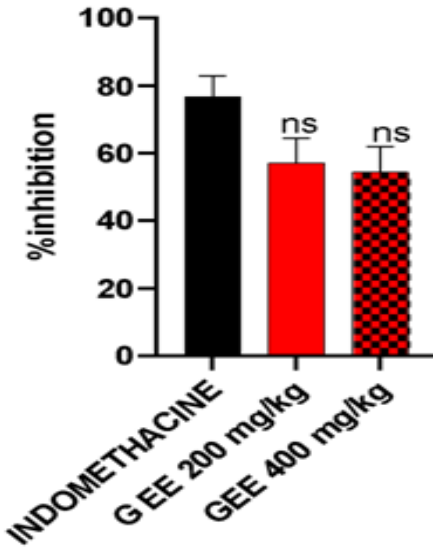

FIGURE 5. Effect of Z. *officinale* ethanolic extract on xylene-induced ear edema in mice compared to indomethacin at 100 mg/kg. GEE: Ethanol extract, values represent means ± SEM (n=6), ns: not significant.

Carrageenan-induced acute inflammation in mice

Carrageenan, a linear sulfated polysaccharide, induces local edema and serves as a standard model for evaluating anti-inflammatory activity [49]. Early-phase mediators of carrageenan-induced edema include bradykinin, histamine, and endothelial-derived nitric oxide, which increase vascular permeability through specific signaling pathways [50 , 51]. In this study, *Z. officinale* extracts effectively inhibited carrageenan-induced paw edema during both the first and second phases of inflammation.

The result of the anti-edematous activity of the oral administration of *Z. officinale* extract in carrageenan-induced paw edema in mice is given in FIG.6. The subplantar injection of carrageenan in the negative control group increased paw edema progressively in the first hour post-injection (31.61 %), attaining the maximum at the fourth hour (54.26 %).

Treatment of the animals with diclofenac resulted in a significant reduction in paw edema two h after sub plantar injection of carrageenan (27.16 %), with the effect persisting for up to 4 h (20.23 %), compared to the control group. Oral administration of the ethanolic extract appears to be more effective at 400 mg/kg, compared to the 200 mg/kg dose.

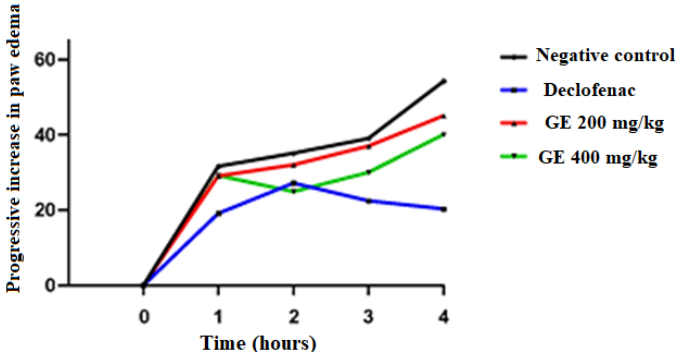

FIGURE 6. Inhibition Kinetics of GE extract vs. Control Standards Over 4 hours. GE: ginger hydroethanolic extract

The anti-edematous effect may be mediated by inhibition of prostaglandin production and antagonism of serotonin, consistent with previous reports [52]. Additionally, the presence of polyphenols and gingerols in the extract might help reduce inflammation by inhibiting cyclooxygenase and lipoxygenase pathways, reducing neutrophil and macrophage activation, and ultimately leading to limitations in monocyte and leukocyte migration [34 , 53]. It is important to point out the clear dose-dependent effect where higher concentrations lead to a greater reduction in pro-inflammatory cytokines and an increase in antioxidant capacities, which further supports the role of secondary metabolites in the observed anti-inflammatory effects [54].

The anti-inflammatory activity and the capacity to stop the release of cytokines that cause inflammation are also part of the *Z. officinale* extract [55]. The plant has been known to have pharmacological activity, especially anti-inflammatory activity and antioxidant activity. The major bioactive compounds found in ginger are gingerols, shogaols, and paradols, which play a vital role in the pharmacological activity of ginger. The anti-inflammatory activity of ginger can be explained by its capacity to inhibit key enzymes such as cyclooxygenase-2 (COX-2) and lipoxygenase (LOX) that are involved in the inflammatory process. This helps to alleviate the inflammation process in diseases such as rheumatoid arthritis and osteoarthritis [56 , 57].

Hence, the role of plant-based phytochemicals, which have ethnopharmacological potential, is to act as the main source of medicine for early drug discovery and to establish the structure for structure-activity relationship studies. Phytochemicals are potent sources of antioxidants, and this property helps in the reduction of various health risks associated with oxidative damage. This is due to the presence of various secondary metabolites such as alkaloids, glycosides, flavonoids, and saponins in the plant, and this property of scavenging free radicals helps in amelioration [58 , 59].

CONCLUSION

The study investigates the phytochemical contents, the antioxidant capacities and the *in vivo* anti-inflammatory activity of *Z. officinale* HEE. The collective outcome revealed ta notable

antioxidant and anti-inflammatory capacities when compared to the reference compounds used in the study. While the synthetic compounds demonstrated a stronger potency, the consistency of the activity displayed by the extract cannot be overlooked. This highlights the unique advantage of the plant, acting on multiple pathways simultaneously, in contrast to the one-component pharmacology of the synthetic compounds. This multi-target approach positions ginger as a valuable addition to complementary and preventive medicine for dealing with issues related to oxidative stress and inflammation. However, the findings call for more rigorous studies, emphasizing the requirement of the extract being biochemically standardized, the characterization of the active compounds, and the mechanistic evaluation at the molecular level.

Future research should be directed towards conducting *in vivo* bioavailability studies to assess the absorption of these compounds, as well as their clinical efficacy for long-term use. This strategy should aid in the scientific validation of *Zingiber officinale* based preparations, particularly for the treatment of oxidative stress-related diseases.

ACKNOWLEDGEMENTS

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Ethical approvals

The experimental assays were approved only after clearance from the Algerian Association of Sciences in Animal Experimentation (AASEA) Committee (http://aasea.asso.dz/articles/), in compliance with Law Number 88-08 of 1988, regulating veterinary medicine, and published in Official Journal Number 004/1988.

In addition, these experiments were carried out in accordance with the International Standards on the care and use of experimental animals (Directive, 2010/63/EU of the European Parliament and of the Council).

Declaration of competing interest

The authors have no known conflict of interest.

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