Arabian Journal of Chemistry (2022) 15, 104308

King Saud University

Arabian Journal of Chemistry

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ORIGINAL ARTICLE

Bioactive principles, anti-diabetic, and anti-ulcer activities of *Ducrosia anethifolia* Boiss leaves from the Hail region, Saudi Arabia

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Received 30 July 2022; accepted 25 September 2022
Available online 30 September 2022

KEYWORDS

Anti diabetic;
Anti-ulcer;
Apiaceae;
Bioactive constituents;

Abstract This work aimed to identify the bioactive constituents of *Ducrosia anethifolia* Boiss eaves through cold methanolic extract. The GC–MS study of cold methanolic extract showed the presence of various pharmaceutically important bioactive compounds with unique peaks at specified retention time. The significant compounds are α-linoleic acid, α-sitosterol, *n*-hexadecanoic acid, palmitic acid β-monoglyceride, 2-methoxy-4-vinylphenol and benzoic acid, methyl ester. The FT-IR study

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Peer review under responsibility of King Saud University.

Production and hosting by Elsevier

https://doi.org/10.1016/j.arabjc.2022.104308
1878-5352

Ducrosia anethifolia Boiss;
Leaves

showed them fingerprint region at 3326.80, 2943.53, 2831.74, 1450, 1110.67 and 1020.80 cm^{-1}. The FT-IR study suggested the presence of glycosides, flavonoids, tannins, steroids, saponins, fatty acids and squalene. Oral administration of Ducrosia *anethifolia* Boiss leaves powder (DLP) (100 mg/kg body weight) was successfully reduced the blood sugar level after 14 d treatment in STZ (50 mg/kg bodyweight) induced diabetic rats significantly from 327.93 ± 24.5 to 171 0.03 ± 3.78 mg/dL. Furthermore, DLP (400 mg/kg body weight) was showed 74 ± 1.9 % inhibition of ulcer. The results of this study showed that DLP has both anti-diabetic and anti-ulcer characteristics when tested in vivo.

1. Introduction

Herbal medicine refers to the use of plant organs such as roots, leaves, flowers, bark, berries, and seeds to treat or prevent disease. The use of herbal remedies for the prevention and treatment of a variety of health conditions has been widespread for a long time worldwide (Al-Ghamdi et al., 2017). In Saudi Arabia, the use of herbal medicine is widespread (Arabsalehi et al., 2022; Shayganfar et al., 2022, Al Akeel et al., 2018). *Ducrosia anethifolia* Boiss (*D. anethifolia),* is a drought-resistant plant that grows all over the Saudi Arabian desert. It is a biennial herbaceous plant with a height range of 10 to 30 cm. The stems are hairless and branched, and they have ramified and ovate-oblong leaves. The umbel inflorescences are *D. anethifolia* composed of white florets. This plant is from the Apiaceae family and has medicinal properties (Akbar and Arezoo, 2017). In Asian countries, such as Afghanistan, Iran, Iraq, Pakistan, and Lebanon, the aerial parts of *D. anethifolia* have long been used to treat headaches, backaches, and colic discomfort (Mottaghipisheh et al., 2020; Mottaghipisheh et al., 20214; Haghi et al., 2004). The effects of *D. anethifolia* aerial parts on the central nervous system have also been documented, including its usage as an anxiolytic, antidepressant, and insomnia treatment (Mottaghipisheh et al., 2020). According to the published research, the crude extract of *D. anethifolia* displayed a wide variety of therapeutic benefits. These benefits included anti-diabetic, anti-microbial, anti-radical scavenging, anti-inflammatory, anti-cancer, ant locomotor, and anxiolytic properties (Shahabipour et al., 2013). In this study, the assessment of bifunctional activity (anti diabetic and anti-ulcer) was investigated because type 2 diabetes mellitus (T2DM) patients are more likely to have *Helicobacter pylori* infection, which can produce symptoms like bleeding and perforation, making peptic ulcer formation one of the most common consequences of T2DM (Tacheci and Bures, 2011). The purpose of this study was to investigate the chemical characterization of *Ducrosia anethifolia* leaves powder (DLP) and to evaluate its effects for antidiabetic and anti-ulcer capabilities. Since the focus of the research was on testing the hypothesis that consuming DLP could be beneficial for the treatment of diabetes and stomach ulcers, the study was designed accordingly.

2. Materials and methods

2.1. Sourcing, taxonomy, and processing of *Ducrosia anethifolia* leaves

Hail Province is in north-central Saudi Arabia which shares borders with the provinces of Madinah, Tabouk, Northern Border, Riyadh, and Qassim. It is the largest province in the country. Hail Province is located in the Waadi Hail region surrounding the Shammar mountain ranges, and it serves as the provincial capital. Jabal Aja is a huge protrusion of granite mountains with deep valleys located to the west and southwest of Ha'il city. Its latitude and longitude are (27°30′N 41°30′E). The site has been expanded north-east to the border of the Nafud desert to cover a region utilized for migrating Grus Virgo surveys (Jibal at-Tuwal and Dilan al-Jilf, 27°50′N 41°45′E). This region is distinguished by flattish semi-desert and sandstone hills punctuated by medium-altitude granitic mountains with deep valleys. Fresh leaves were collected from the plants and taken to the laboratory in plastic bags. After removal of contaminants by rinsing with running water from a tap, the leaves were rinsed with Millipore water, followed by air-drying on a neat floor for 10 days. Washed branches of tree specimens were identified by a registered taxonomist, after which the sample was deposited at the University of Hail Herbarium, with reference number UOH-COP010. The voucher specimen was also donated to the University Herbarium, where it is being kept as source of information in the future. The air-dried sample was chopped into little bits and finely ground into powder with a blender. The finely-powdered leaf specimen was stored in a sealed vessel. The leaf powder of *D. anethifolia* was designated as DLP.

2.2. Materials

All chemicals and organic solvents were purchased from Sigma, USA. Ejadah Medical Supplies Est, Riyadh, Saudi Arabia, supplied all items for this study.

2.3. Extraction procedure

Methanol (99.6 %) was utilized for extraction because to its greater volatility and extraction efficiency. Cold methanol maceration was used to extract the bioactive parts of the DLP. To create the reaction mixture (RM), 25 g of DLP were soaked in 50 mL of methanol and agitated with a magnetic stirrer on a hot plate at room temperature for 60 min. Overnight, the RM was stored in a refrigerator at 4 °C. The process was then repeated for a further week while the RM was retained in a magnetic stirrer. The macerated RM was centrifuged using a Sigma table-top centrifuge at 2000 × g for 10 min. The extract was produced by filtering the supernatant solution through Whatman No. 1 filter paper and drying it by air at 25 °C. Based on this technique 20 % yield was able to achieve soon after drying the solvents. The dried sample was gathered and kept for future use in a refrigerator at 4 °C (Rahamat et al., 2022).

2.4. FT-IR spectroscopy

FT-IR spectroscopy was used to examine the individual functional groups of bioactive compounds in the methanolic extract of DLP using a Nicolet iS10 FT-IR spectrophotometer

(Thermo Scientific, USA). The FT-IR spectrophotometer was used to obtain spectra of the pellet sample against a reference KBr pellet in the range of 400–4000 cm^{-1}, with a resolution of 4 $cm^{-1,}$ using a simple KBr pellet method (Rahamat et al., 2022).

2.5. GC–MS analysis

The presence of bioactive components in the methanolic extract of DLP was determined by GC–MS (Thermo Scientific GC–MS-AS 3000 autosampler - ISQ detector). Using a TR 5MS capillary column with helium as a carrier gas at a flow rate of 1.2 mL/min, 2 µL of the methanol diluted powdered sample was injected for partial separation of bioactive components. The injector was run at 250 °C while the oven temperature was 60 °C for 15 min, then increased gradually to 280 °C over the course of 3 min. The MS data were taken from 30 to 600 *m/z* with a 2-minute solvent cutoff. The software XCalibur was utilized for data collecting and processing. Utilizing the NIST and MAINLIB software libraries, the mass spectra were interpreted (Gomathi et al., 2015).

2.6. In vivo study

2.6.1. Experimental animals

The study was initiated after proper approval by the Institutional Animal Ethical Committee (MRC/JU/1443/SA2). Male Wistar rats were purchased from the Medical Research Centre (MRC), Jazan University. Male Wistar rats weighing 150 ± 30 g were allowed to acclimatize to the laboratory environment for two weeks at 22 ± 0 8 °C and relative humidity of 56 ± 6 % which was attained in an atmosphere with equal durations of light and darkness. Rats were kept individually in cages. Clean water and feed were provided ad libitum.

2.6.2. Diabetic model

The rats were randomly divided into four groups (5 rats per group and categorized as follows:

Group 1: **Normal control group:** The animals did not receive streptozotocin (STZ).

Group 2: **Disease control group** (diabetes induction): The STZ was administered to the rats through intraperitoneal injection at a dose of 50 mg/kg of body weight dissolved in 0.9 % w/v sodium chloride.

Group 3: Standard drug treatment group:

The rats received intraperitoneal injection of STZ (50 mg/kg bodyweight) dissolved in normal saline for diabetic induction and the animals received rosiglitazone orally as single dose (5 mg/kg/d) for 21 consecutive days.

Group 4: DLP treatment group:

The rats received intraperitoneal injection of STZ (50 mg/kg bodyweight) dissolved in normal saline for diabetic induction and the animals were administered orally with DLP (100 mg/kg body weight) in distilled water as a single dose for 21 consecutive days.

The level of glucose in all groups of animals was measured on the tail-vein blood using a standard commercially available glucometer. Following the induction of diabetes, the levels of fasting glucose level in the blood were evaluated at specified time in the morning. The level of glucose was measured on the tail-vein blood using a standard commercially available glucometer, we determined the amount of glucose in the blood drawn from the tail vein.

2.6.3. Gastric ulcer model

Thirty male rats were randomly divided into six groups, with five animals/group. The ulcer model was in line with a previously published procedure, but with slight modification (Al-Wajeeh et al. 2016). The groups used, and their treatments were as follows:

Group 1: Normal control group: In this group, stomach ulcers were not produced, and as a result, the animals did not receive any treatment.

Group 2: Disease control group: Ulceration group: In overnight-fasted rats, ulceration was induced by administration of 95 % (*v/v*) ethanol (5 mL/kg body weight).

Group 3: Standard drug treatment group: These animals received omeprazole (20 mg/kg body weight in distilled water) given as a single oral dose at 2 h prior to ethanol administration.

Group 4: The animals received OLP (100 mg/kg body weight) in distilled water as a single dose orally at 2 h prior to administration of 95 % (*v/v*) ethanol.

Group 5: The animals received OLP at oral dose of 200 mg/kg body weight in distilled water as a single dose 2 h prior to administration of 95 % (*v/v*) ethanol.

Group 6: The animals received OLP at oral dose of 400 mg/kg body weight in distilled water as a single dose 2 h prior to administration of 95 % (*v/v*) ethanol. Diethyl ether was used to anesthetize the rats before they were sacrificed by having the jugular veins in their necks cut with a sharp, sterile scalpel to draw blood.

2.6.4. Determination of ulcer index and % inhibition of ulcer

We measured the overall mucosal surface area as well as the total ulcerated surface area. Ulcer index (U.I.) was determined according to the method used in previously published work (Sabiu et al., 2015), applying the following equation:

$$U.I. = \frac{Ulceratedarea}{Totalstomacharea} \times 100$$

The % inhibition of ulceration was calculated as follows:

$$\%Inhibition = \frac{(Ulcerindexofcontrol) - (ulcerindexoftest)}{Ulcerindexofcontrol}$$

2.6.5. Macroscopic and biochemical gastric assessments

The stomach's contents and tissues were removed for macroscopic and pathological analysis. Using a USB digital microscope with a magnification endoscope camera, photographs were captured. The total ulcerated area was estimated using the standard approach, with slight modifications, and the % inhibition was computed using a modest modification of the standard given in a previous publication (Njar et al., 1995). Acidity was determined with a pH meter and titration with sodium hydroxide solution, and the results are presented in milliequivalents per liter (Tan et al., 2002). A sensitive digital balance was used to determine mucus weight. The volume (mL) of the gastric juice was also measured using measuring cylinder.

2.7. Statistical analysis

Data are presented as mean ± SD. Comparison amongst groups was done with one way ANOVA and Dunnett's multi-comparison test. Values of $p < 0.05$ indicated statistically significant differences. Statistical analysis was performed by GraphPad Prism, 9 software, USA.

3. Results and discussion

Natural products are a unique and important source of bioactive substances that can potentially have therapeutic effects and come in a wide variety of molecular structures (Nishitha et al., 2018). Plants have a high concentration of biologically active chemicals that have therapeutic capabilities, which has led to their use in traditional medicine for the treatment of a wide variety of illnesses (Collins et al., 2018). In Saudi Arabia, numerous plants have an important role in the practice of traditional medicine (Aati et al., 2019). Therefore, the current study was conducted to investigate the bioactive constituents of cold methanolic extract of DLP, anti-diabetic and anti-ulcer properties of DLP. Results from GC–MS revealed that the presence of several bioactive compounds, as shown in the chromatogram in Fig. 1. The existence of several bioactive elements is depicted in Table 1, and their structures are shown in Fig. 2.

9,12-octadecadienoic acid (Z,Z)- is otherwise called α-linoleic acid exhibited retention time (RT) 77.07 min with a % probability of 51.89 and occupy the maximum, almost 14.02 % in the GC–MS chromatogram. Khalid et al. (2009) reported that the seed oils of *Ducrosia anethifolia* contain oleic, linolenic, and palmitic acids with trace levels of other saturated fatty acids. According to previous studies, α-linolenic acid may reduce the risk of heart disease by aiding in the preservation of a normal heart rhythm, reduces the blood clots and pumping

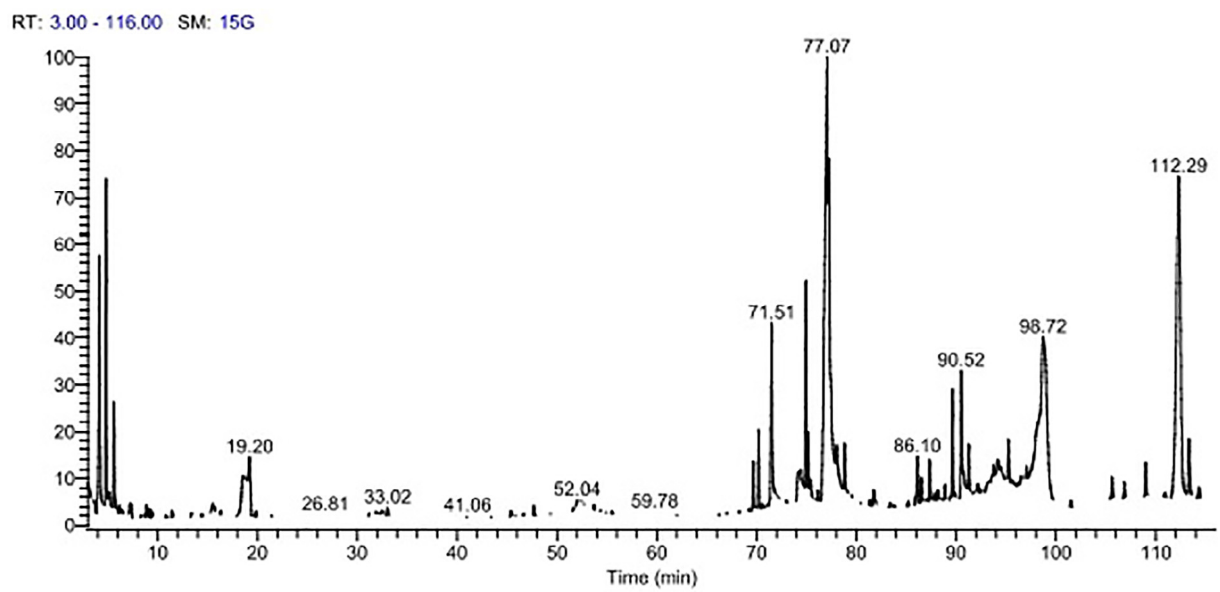
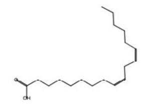


Fig. 1 GC–MS chromatogram of the cold methanolic extract of *Ducrosia anethifolia* leaves powder.

Table 1 GC–MS detection of possible bioactive compounds cold methanolic extract of *Ducrosia anethifolia* leaves powder.

S. no	Compound name	Molecular formula	Molecular weight	Retention time (Min)	Probability Index	Percent area of curve
1	9,12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	280	77.07	51.89	14.02
2	α-Sitosterol	$C_{29}H_{50}O$	414	112.29	48.34	10.80
3	*n*-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	71.51	80.67	4.30
4	n- Heptacosane	$C_{27}H_{56}$	380	98.72	51.08	3.56
5	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl	$C_{19}H_{38}O_4$	330	90.52	53.37	2.68
6	Limonene	$C_{10}H_{16}$	136	19.52	25.31	2.64
7	2-Dodecenylsuccinic anhydride	$C_{16}H_{26}O_3$	266	86.10	51.17	1.31
8	17-Octadecynoic acid	$C_{18}H_{32}O_2$,	280	52.04	40.95	0.15
9	2-Methoxy-4-vinylphenol	$C_9H_{10}O_2$	150	41.06	24.76	0.15
10	Decanal	$C_{10}H_{20}O$	156	52.05	33.03	0.15
11	Benzoic acid, methyl ester	$C_8H_8O_2$	136	25.33	59.77	0.10
12	Ethyl isoallocholate	$C_{26}H_{44}O_5$	436	26.81	57.08	0.10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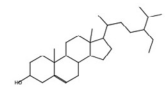
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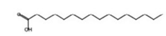
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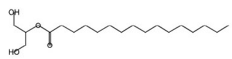
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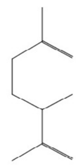
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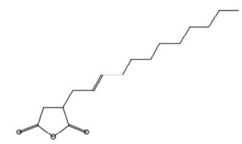
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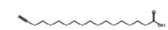
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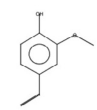
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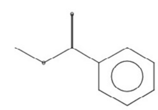
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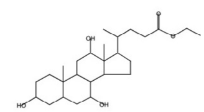
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Fig. 2 GC–MS detection of possible bioactive compounds of the cold methanolic extract of *Ducrosia anethifolia* leaves powder. (1) 9,12-octadecadienoic acid (Z,Z)- (2) α-sitosterol (3) *n*-hexadecanoic acid (4) n- heptacosane (5) Hexadecanoic acid, 2- hydroxy-1-(hydroxymethyl)ethyl (6) Limonene (7) 2-dodecenylsuccinic anhydride (8) 17-octadecynoic acid (9) 2-methoxy-4-vinylphenol (10) Decanal (11) Benzoic acid, methyl ester (12) Ethyl isoallocholate.

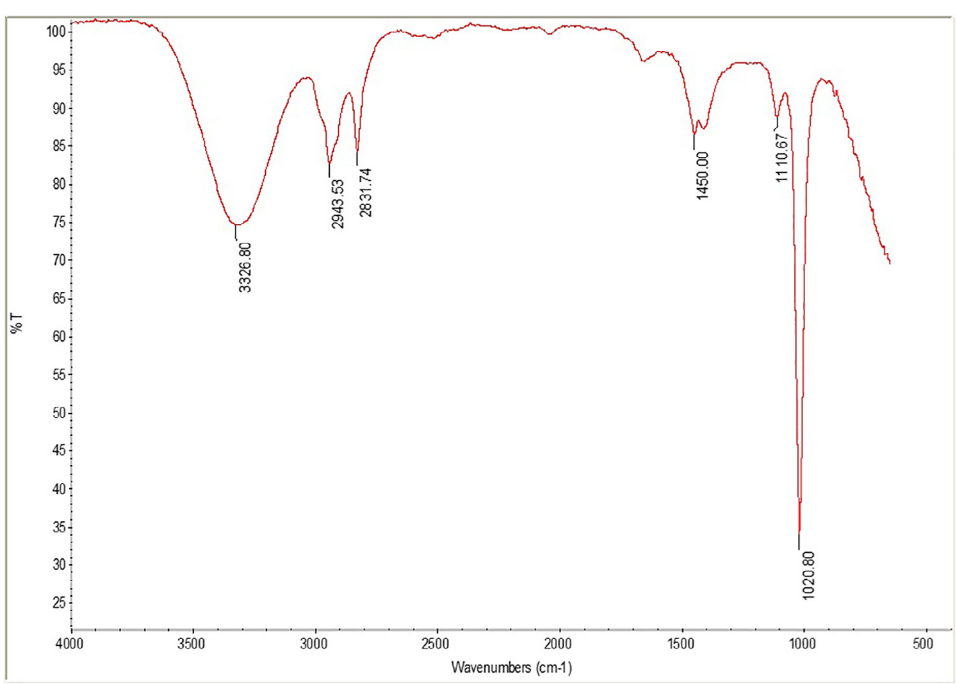

Fig. 3 The FT-IR spectra of compounds of the cold methanolic extract of *Ducrosia anethifolia* leaves powder.

motion (Sina et al., 2021; Watanabe and Tatsuno, 2017; Guasch-Ferré et al., 2022;; Yang et al., 2014; Brzosko et al., 2002). Previous research has suggested that omega-3 polyunsaturated fatty acid, α-linolenic acid influence glycemic management by the reduction in HbA1c (Jovanovski et al., 2017). Recent research suggested that α-linolenic at 300 mg/Kg body weight dosage reduced the blood glucose level by decreeing the insulin resistance (Kato et al., 2020). Thai *Perilla* *frutescens* fruit oil is rich in α-linolenic acid (ALA) reported as anti-ulcer, analgesic, and anti-inflammatory effects (Paradee et al., 2021).

α-sitosterol was determined in the cold methanolic extract of DLP at 112.29 min RT, with a % probability of 48.34 and occupied 10.8 % in GC–MS curve. α-Sitosterol otherwise called sitostanol belongs to stigmastanes and derivatives. Sitosterol have been reported inhibit the cholesterol absorption across the plasma membrane of enterocytes (Manisha and Nicola, 2004). It has been found through research that plant phytosterols have a gastroprotective effect, and a portion of an extract that contains β-sitosterol as its primary component has been shown to have effectiveness against peptic ulcers (Kumadoh et al., 2021; Noungoué et al., 2001). An earlier study reported that β-sitosterol exhibited anti diabetic and anti-oxidant properties against streptozotocin-induced diabetic model (Gupta et al., 2011).

n-Hexadecanoic acid commonly known as palmitic acid exhibited a RT of 71.51 min, with a % probability of 80.67 and with a % occupancy in the chromatogram was 4.30. A

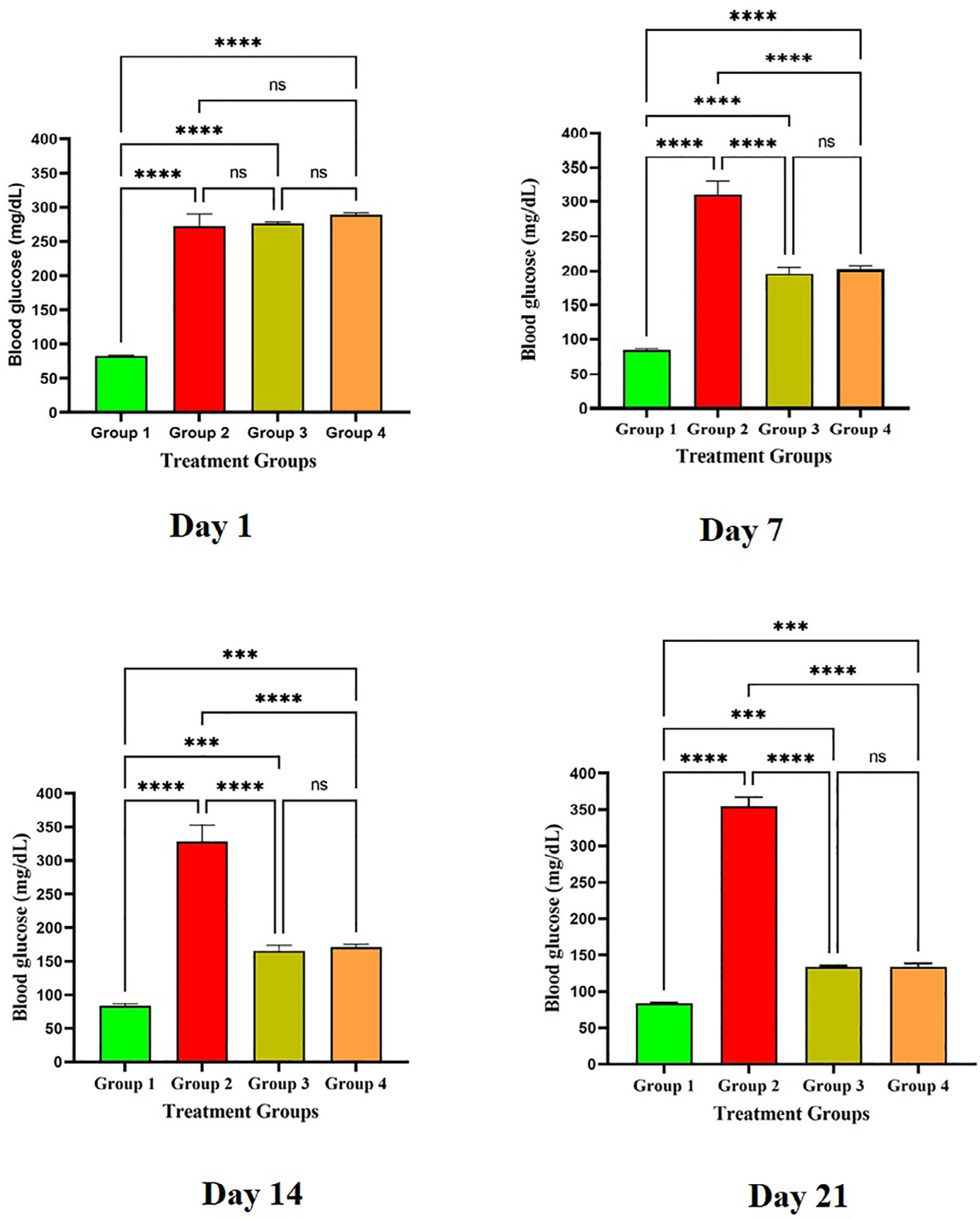

Fig. 4 The fasting blood glucose level in various treatment groups during treatment phase. **** The values are very high significant at $p < 0.05$ level. *** Significant at $p < 0.05$ level. ns: nonsignificant at $p < 0.05$ level.

recent study found that palmitic acid greatly reduced the growth of prostate cancer cells both *in vitro* and *in vivo* (Zhu et al., 2021). An earlier study demonstrated the anti-inflammatory effects of palmitic acid. Additionally, it had a strong antibacterial effect on bacteria that formed biofilms (Bakar et al., 2017; Aparna et al., 2012). An earlier study found that *n*-hexane crude extract of *Artocarpus camansi* peels has anti-diabetic action (Rosnani et al., 2018). According to Pu et al. (2011), skeletal muscle cells' Akt and ERK1/2 are activated by palmitic acid, which increases glucose absorption.

Hexadecenoic acid, 2- hydroxy-1- (hydroxymethyl)ethyl is otherwise called palmitic acid β -monoglyceride was exhibited at the retention time of 90.52 min with 53.37 probability index and depicting 2.68 % in chromatogram. Limonene is a cyclic monoterpene is the one of the constituents of in the cold methanolic extract of DLP detected at 19.20 min with % probability of 25.31 and occupies 2.64 % in chromatogram. According to the findings of a recent study, limonene may aid in weight loss by reducing blood sugar and cholesterol levels which are related to metabolic syndrome. Limonene may potentially aid in the prevention of peptic ulcers and other inflammatory gastrointestinal illnesses (Li et al., 2013; Thiago et al., 2009). 2-Methoxy-4-vinylphenol is a natural aromatic substance belongs to methoxyphenols group have been identified in the cold methanolic extract of DLP. The compound, also known as 4-vinylguaiacol, is employed as a flavoring agent in the food, cosmetic, and pharmaceutical industries. It is also frequently utilized in industrial food processing (Li et al., 2019). Benzoic acid, methyl ester is otherwise called methyl benzoate and decanal were identified in the cold methanolic extract of DLP which has pleasant smell and used perfumery. n- Heptacosane, 2-dodecenylsuccinic anhydride, and ethyl isoallocholate have been identified at specified retention time in the cold methanolic extract of DLP.

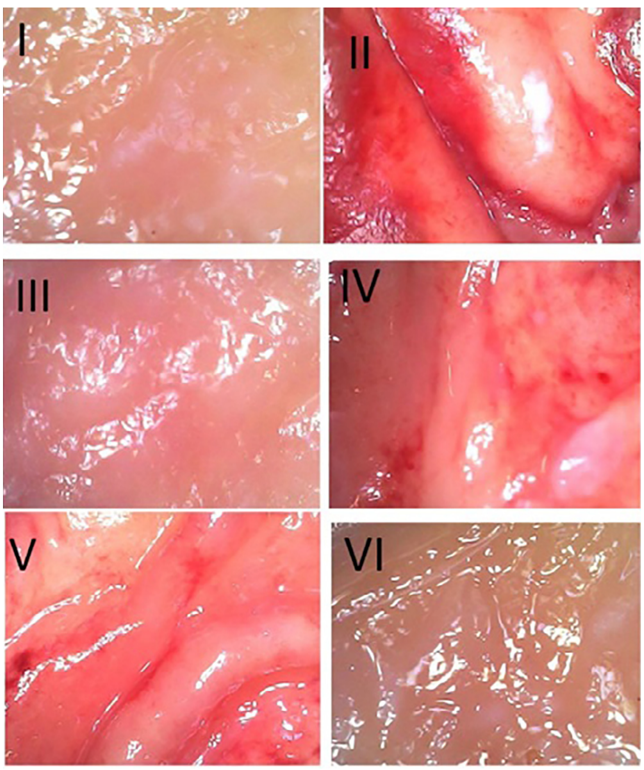

Fig. 5 Examination of hemorrhagic lesions macroscopically on the glandular part of the rat stomach. I, II, III, IV, V and VI are representative photos from groups 1, 2, 3, 4, 5 and 6 respectively. Photo II showed the most severe hemorrhagic lesions on the glandular part of the stomach obtained from Group 2.

The chemical groups in the cold methanolic extract of DLP were determined using FT-IR spectra by recording in the fingerprint region of 400–4000 cm^{-1} (Fig. 3). The FT-IR spectroscopy showed the presence of many peaks at various fingerprint regions. The investigation of cold methanolic extract of DLP with spectroscopy revealed distinct peaks, which verified the presence of functional groups and the molecules that correspond to them. The bell-shaped peak at 3326.80 cm^{-1} shown by FT-IR spectrum measurements contained a variety of functional groups, including stretching of the carboxylic acid with stretching frequencies that indicated the presence of glycosides, flavonoids, and saponins. The frequencies at 2943.53 and 2831.74 cm^{-1} represents the aliphatic compounds with CH2 stretching vibrations and depicting the presence of steroids, tannins, and saponins. The stretching vibrations in the fingerprint region at 1450, 1110.67 and 1020.80 cm^{-1} also suggest the presence of fatty acids, and squalene (Rahamat et al., 2022).

Fig. 4 depicting the fasting blood glucose level on 1st, 7th, 14th and 21st day before and after treatment with DLP which was well compared to the activity of standard drug rosiglitazone. The potential of DLP was improved after 14 days treatment. An earlier study showed the treatment with *Ducrosia anethifolia* extract exhibited hypoglycemic effect because of the presence of linear furanocoumarins that induces carbohydrate metabolizing enzymes (Nagwa et al., 2014). The cold methanolic extract of DLP demonstrated the presence of -linoleic acid, palmitic acid, and limonene, all of which have been linked to the induction of hypoglycemia effects. Therefore, successful reduction of blood glucose level from 14 d

Table 2 Effects of *Ducrosia anethifolia* leaf powder and omeprazole, administered intraesophageally, on the biochemical parameters of gastric juice obtained from rats.

Groups	Treatment	Ulcer area (mm^2)	% Inhibition	Mucus weight	pH
1	Normal control	0.00	0.00	2.8 ± 0.23	3.61 ± 0.14
2	Ulcer Control	550 ± 44	NA	0.95 ± 0.3	3.61 ± 0.14
3	Omeprazole	120 ± 6.7	79.0 ± 1.2	1.45 ± 0.2	6.54 ± 0.021
4	DLP (100 mg/Kg)	230 ± 13.6	59 ± 3.6	1.30 ± 0.4	4.92 ± 0.092
5	DLP (200 mg/Kg)	174.4 ± 9.7	68.2 ± 2.2	1.9 ± 0.1	5.4 ± 0.041
6	DLP (400 mg/Kg)	146 ± 10.3	74 ± 1.9	1.6 ± 0.2	6.29 ± 0.074

treatment with DLP on comparing with rosiglitazone treatment and the levels were non-significant at $p < 0.05$ level (Fig. 4). Interestingly, the DLP was also exhibited anti-ulcer effect and the results were depicted in Table 2.

Pre-treatment with DLP at doses of 100, 200, 400 mg/kg body weight and omeprazole at 20 mg/kg significantly ($p < 0.05$) reduced the ulcer area formation by 59 ± 3.6, 68.2 ± 2.2, 74 ± 1.9, and 79 ± 1.2 %, respectively, compared to the ulcer control (Fig. 5). In the animal model using ethanol as an ulcerogenic agent, the pre-administration with DLP (100, 200, 400 mg/kg) and omeprazole (20 mg/kg), respectively, decreased the volume of gastric juice, total acidity and raised gastric pH considerably ($p < 0.05$) compared to the control group (Table 2). The presence of α-linoleic acid, sitosterol, palmitic acid, and limonene in the cold methanolic extract of DLP showed that these compounds can induce antiulcer effects. Despite the demonstrated advantages, there are several drawbacks that need to be taken into consideration as well. The relatively high separation cost could result in overpriced products, which could be partially offset using a single solvent to create a high extract yield. Furthermore, it is critical to emphasize that these are natural extracts with a unique composition that varies depending on the season, growing conditions, and extraction procedure. Therefore, isolation of disease specific bioactive compounds is highly challenging and noted worth to say that antidiabetic and anti-ulcer bioactive compounds can be isolated from the leaves of *Ducrosia anethifolia*.

4. Conclusion

Natural products are essential for keeping excellent health. Medicinal plants have traditionally been the primary source of treatment for numerous diseases around the world. The current study provides scientific evidence for the bioactive components, anti-diabetic, and anti-ulcer activity of *Ducrosia anethifolia* leaf powder (DLP) from the Hail region of Saudi Arabia. The study found that consuming *Ducrosia anethifolia* leaves can help in the management of diabetes and gastric ulcers. Bioactive compounds were distinct in their chemical structure and long retention time. The findings of this study indicate the presence of several bioactive chemicals that may be useful in the future development of anti-diabetic and anti-ulcer medicines.

CRediT authorship contribution statement

Rahamat Unissa Syed: Conceptualization, Resources. **Sivakumar S. Moni:** Conceptualization, Investigation. **Bader Huwaimel:** . **Ahmed Alobaida:** . **Afnan Abdulkareem Almarshdi:** . **Amr S.Abouzied:** . **Amr S.Abu Lila:** . **Marwa H. Abdallah:** . **Humera Banu:** . **Mohd Abdul Hadi:** . **Hemat El-Horany:** . **Siddig Ibrahim Abdelwahab:** . **Manal Thaha:** .

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

This research has been funded by the Scientific Research Deanship at the University of Hail -Saudi Arabia through project number RG-21 129. The authors would like to thank Dr. Zia ur Rehman and Md Shamsher Alam from the Department of Pharmaceutical Chemistry at Jazan University for performing the GC-MS and FT-IR studies.

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