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Research Article

Phytochemical and Antimicrobial Activity of the Extract of *Cymbopogon citratus* Plant

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Abstract

Objective: This study investigated the phytochemical and antimicrobial activity of *Cymbopogon citratus (Mahareeb)* plant leaves extract (MPLE) against two fungi (*Aspergillus niger* and *Penicillium italicum*) and two bacteria (*Escherichia coli* and *Salmonella typhi*).

Materials and Methods: To determine the phytochemical constituents in the plant extracts, we tested for phenols, tannins, alkaloids, flavonoids, terpenoids, sterols, cardiac glycosides, saponins, lipids, and fixed oils.

Results: The plant extracts were found to inhibit mycelial radial growth of both fungi. The effect was clear at the first days, although with further incubation *Aspergillus niger* was able to resist the lower concentrations and at the 8th day. The effect was not significantly better than the control treatment. Mycelial fresh and dry weights of both fungi were also greatly reduced with the *mahareeb* extracts. The higher concentration gave the maximum effect which decreased with dilution. The effect on mycelial growth was more pronounced on *A. niger* than on *P. italicum*. The effect of *mahareeb* extract on the two bacteria (*E. coli*, *S. typhi*) was evaluated by the inhibition zone and dilution methods. A clear zone of inhibition was shown by the extracts against both bacteria, although the effect was less against *E. coli*.

Conclusion: The results of this study suggest that MPLE has potential as an antibacterial, antifungal and antioxidant agent.

Keywords: *Mahareeb; Tannins; Saponins; Antifungal; Antibacterial Activity; Herbalism; Radial Growth*

Introduction

Herbal medicine, herbalism, or botanical medicine uses herbs for medicinal purposes. Herbs are medicinal, aromatic, or culinary plants or plant parts. Herb plants produce several body-affecting chemicals [1]. From 2022 to 2026, the worldwide herbal medicine market is estimated to increase at 8.1% CAGR from $135 billion. Extracting medicinal components from herbal plant leaves makes many herbal supplements. Herbal drugs and dietary supplements treat or prevent disease using whole plants or isolated plant elements. Medicinal herbs are good for treating a variety of health conditions and for usage in food [2].

Constipation, malaria, gout, jaundice, and other conditions are all treated with plants in traditional Sudanese medicine. Most of the plants on the list have medicinal properties, and they are used to treat things like spasms, nausea, bleeding, high blood pressure, and diabetes. Many of these plants have a wide variety of therapeutic and pharmacological applications. Most research has focused on the plants' antioxidant, antibacterial, antifungal, and anti-inflammatory properties [3].

Lemongrass, *Cymbopogon citratus* L., grows in warm areas. Lemongrass oil, steam-distilled from dried leaves, is a yellowish, highly fragrant liquid with significant chemical properties. Perennial herbaceous plant with rose-like smell.

The plant is used. Geraniol, like lemongrass oil, and citral, used to make vitamin A and ionone smell, are volatile oil constituents. It contains flavonoids.

Mild, it dissolves solid tumors in the stomach, liver, and kidneys as a drink and bandage; stimulates urine and menstruation; breaks up stones; strengthens the teeth and stomach columns; relieves nausea and rationalizes the stomach; and has a strong root.

C. citratus more commonly known as lemongrass, has been oughed for medicinal purposes for many years. Numerous studies have reported that lemongrass oil has antibacterial activity against a wide variety of organisms; consequently, this study was conducted to evaluate the effectiveness of lemongrass extract as an adjunct in mouthwashes against *S. mutans* [4], the most important oral pathogen, in order to inhibit the formation of plaque. Due to its antibacterial and antioxidant qualities, lemongrass (*C. citratus*) essential oil can replace synthetic food preservatives.

C. citratus is a popular herb [5]. *C. citratus* extracts have diverse pharmacological actions. Its anti-infection, inflammatory, diabetes, and cancer properties are well established. *C. citratus* works alone or with additional plant compounds. Since *C. citratus* has many ethno-pharmacological uses, many studies have characterized its volatile and nonvolatile chemical components. These analyses show that *C. citratus's* essential oil and extracts' chemical composition varies by geographic origin, age, and character. *C. citratus'* isolated compounds comprise as many chemical families as the plant.

Cooking and preserving food with *mahareeb*, or lemongrass, oil is natural. Recent research has demonstrated that pure lemongrass essential oil can replace synthetic fungicides and other sanitation methods during storage and packing [6]. This study examined *mahareeb* (*C. citratus*) plant leaves extract (MPLE) for phytochemical and antibacterial and antifungal activities.

Materials and Methods

Collection of samples

C. citratus L. (*Mahareeb*) plant samples. The leaves were purchased in 2008 from vendors at the Wad Medani local market. The samples were collected from various retail locations. The leaves were cleaned of debris including rocks, sand, and dust before being stored in the lab for further study. The leaves were then sanitized with water, dried, and ground into a powder in a laboratory mill.

Different concentrations (0%, 50%, and 100% extract) were prepared using aqueous extracts of the powder (50g/500 ml).

Cultural origins

Microbiological Laboratory of the Department of Food Science and Technology, Faculty of Engineering and Technology, University of Gezira, Wad Medani, Sudan, provided the two bacterial isolates (*Escherichia coli* and *Salmonella typhi*) and the two fungal isolates (*Aspergillus niger* and *Penicillium italicum*).

Phytochemical profiles

To determine what compounds were present in the extracts, we tested for phenols, tannins, alkaloids, flavonoids, terpenoids, sterols, cardiac glycosides, saponins, lipids, and fixed oils.

Flavonoids and polyphenols

Each extract was diluted in 1 ml of distilled water, and then a few drops of ferric chloride in methanol (at a concentration of 5%) were added. The presence of phenolic compounds can be seen as a bluish-black hue [7].

Alkaloids

The presence of the alkaloids can be indicated in two ways:

1. For the Dragendorff assay, 5 mg of extracts were mixed with 2 ml of MeOH and 2 ml of 1% HCl along the side of the test tube, and then 500l of Dragendorff's reagent was added. U.C. and Nair found that a positive test result was marked by the appearance of an orange or orange-reddish brown precipitate.
2. The extract was diluted to 1 mg/ml and then tested using the Mayer's reagent. It is known that alkaloids (U.C. and Nair) are present when a white creamy precipitate forms.

Alkaline reagent test for flavonoids

The extracts (5 mg) turned a bright yellow after being mixed with 2 cc of sodium hydroxide (2%). The presence of flavonoids is shown by the bleaching out of the hue upon addition of diluted HCl acid [7].

Five milligrams of extract was combined with two milliliters of chloroform for the Terpenes (Salkowski's test). Following that, 3 ml was added. Terpenoids are present when a reddish-brown tint develops [7].

Sterols

Five milligrams of extract was mixed with two milliliters of hydrogen peroxide. A shift in hue from violet to blue or green denotes the presence of steroids [8].

Glycosides of the heart (The Keller-Kiliani procedure)

The extract was diluted with 1 ml of glacial acetic acid and a few drops of a ferric chloride solution (5%). The side of the test tube was also injected with 0.5 cc of strong sulfuric acid. If a green or blue tint develops, then cardiac glycosides are present [7].

Saponins

To 5 mg of extract, 10 ml of distilled water was added. Five minutes were spent vigorously shaking the fluid. The existence of saponins can be verified by their ability to create a stable foam [9].

Oils

To one ml of extract, we added 1 ml of a 1% copper sulphate solution and a few drops of 10% sodium hydroxide. Lipids and fixed oils (U.C. and Nair) can be confirmed to be present when a clear blue solution forms.

Mahareeb extracts and fungal growth

Mycelial weight

PDB medium was made and distributed in 100 ml conical flasks (250 ml). Each flask was diluted with the extracted solution (0.0, 50, and 100%). Before inoculation, each flask was sterilized in an autoclave at 121°C (I5-lb/in12) for 15 minutes. Each flask was infected with three 5.0 mm discs from an edge of an actively developing culture of *A. niger* or *P. italicum* on solidified PDA media. Inoculated flasks were cultured at room temperature (28 - 30°C) for 8 days. After incubation, mycelia were filtered using Whatman No.1 filter paper, weighed, and dried at 80°C for 24 hours before being weighed. Treatments were tripled.

Radial growth impact

The Potato Dextrose Agar (PDA) medium was made and dispensed in 100 ml conical flasks (250 ml). Extract solutions were added to each flask to obtain multiple dilutions (0.0, 0.50, and 100%). Finally, the media were autoclaved at 121°C (15 Ib/In2) for 15 minutes.

Each flask's medium+extract solution was placed into sterile Petri dishes and permitted to harden at room temperature (28 - 30°C) for 24 - 40 hours after sterilization. A sterile cork-borer (No.5) cut a fungal growth disc from an edge of an actively developing culture of *A. niger* or *P. italicum* cultivated on PDA and infected each solidified media centrally. Radial growth was measured every two days for eight days in inoculated Petri dishes at room temperature. Treatments were tripled.

Calculation

The average of two crossing dimensions for each Petri dish disc measured growth diameter every 48 hours. The glass Petri dish diameter was used to compute the radial growth percentage.

Mahareeb extract and bacterial growth

Two tests were performed.

Cup-plate (Inhibition zone) agar diffusion

This approach employed Nutrient Agar (NA). In this approach, 2 ml of a standardized bacterial cell suspension (10 X 10^5) of *E. coli* or *S. typhi* was well mixed with 200 ml of sterile molten nutrient agar, then spread into sterile Petri plates and permitted to solidify at room temperature for 24 hours. Sterile Whatman glass fiber discs (No. 5) were saturated with MPLE, dried, and placed centrally on the solidified medium in each plate. Barry., *et al*. [10] and Cruickshank., *et al*. [11] examined the inhibitory zones after 72 hours of room-temperature incubation. Treatments had three duplicates.

Dilution plate method

Mahareeb aqueous extracts were tested for bacterial growth using the Dilution Plate Method. Each 9-ml McCartney bottle contained nutrient broth medium with varying MPLE concentrations. Each bottle received 1 cc of the above bacterial suspension after the medium was autoclaved. The dilution plate method assessed bacterial growth in three inoculated bottles incubated at room temperature. One ml was collected from each bottle and placed in a tube cooling to 9 ml sterile distilled water and serially diluted.

A sterile pipette deposited 0.1 ml of each dilution over the surface of solidified Nutrition Agar (NA) in Petri dishes and spread it with a glass rod. At room temperature, inoculated plates were counted for colonies at various times. The number was log colonies per milliliter. Number logs were plotted versus time.

Phytochemical constituents

MPLE's phytochemical makeup is summarized in table 1. According to the data collected, the extract in question contains a wide variety of beneficial chemicals, including antioxidants, saponins, terpenes, flavonoids, phenols, tannins, and cardiac glycosides.

Saponins	Terpenes	Flavonoids	Phenols	Tannins	Alkaloids (Dragendorff)	Alkaloids (Mayer)	Cardiac glycosides
+	-	+	+	+	-	-	+

Table 1: *Phytochemical profile of MPLE.*

Antifungal properties

A. niger, P. italicum, and *E. coli,* and *Salmonella typhi* were all tested for their susceptibility to (MPLE) inhibition in the current study.

Using PDA and PDB media, the extracts were evaluated for their effects on mycelial growth (mycelial radial growth and mycelial fresh and dry weights). Extracts of MPLE had a significant effect on *A. niger's* mycelial radial growth, as seen in table 2. This effect was most noticeable on the first few days of incubation, but by the eighth day the fungus had adapted to the reduced concentrations and the effect was no longer distinguishable from the control.

Time (days)	Concentrations		
	0.0	0.5	1.0
2	7.0	4.5	3.43
4	8.5	5.8	3.9
6	8.6	5.7	5.2
8	9	7.8	5.3

Table 2: *The effects of different concentration of Mahareeb on radial growth of A. niger.*

Similar to the effect on radial growth, the effect of MPLE extract on the mycelial weight of the fungus *A. niger* decreased with dilution (Table 3). The maximal suppression was seen at the highest dose.

Concentration	Fresh weight (g)	Dry weight (g)
0.0	8.95	4.50
0.0	8.85	4.66
1.0	8.35	4.70

Table 3: *The effects of the different concentration of Mahareeb extracts on mycelial fresh and dry weight of A. niger.*

The effect of (MPLE) on *P. italicum's* radial growth was depicted in table 4. Results showed that a greater dose of MPLE significantly inhibited radial development. The impact was greater on *P. italicum* than on *A. niger*. The greater concentration showed a decrease of more than 50%. Using the MPLE significantly decreased both the wet and dry weights, as seen in table 5.

Time	Concentrations		
	0.0	0.5	1.0
2	4.30	3.0	2.65
4	8.82	6.84	3.12
6	8.80	5.35	3.25
8	9	7.22	3.66

Table 4: *The effects of different concentration of Mahareeb extracts on radial growth of P. italicum.*

Concentration	Fresh weight (g)	Dry weight (g)
0.0	2.42	1.81
0.0	3.24	2.33
1.0	4.70	2.74

Table 5: *The effects of different concentration of Mahareeb extracts on fresh and dry weight of P. italicum.*

Incubation time	Concentrations		
	0.0	0.5	1.0
0	2.75	2.68	2.45
6	2.95	3.21	2.75
24	3.43	2.72	2.54
48	3.58	2.78	1.90
72	3.70	2.44	1.30

Table 6: *The effects of different concentration of Mahareeb extracts on log number of the bacterium E. coli.*

Incubation time	Concentrations		
	0.0	0.5	1.0
0	1.35	1.26	1.28
6	1.43	1.21	1.05
24	4.32	2.80	2.43
48	4.48	3.56	2.36
72	4.64	3.64	0.30

Table 7: *The effects of different concentration of Mahareeb extracts on log number of the bacterium Salmonella type.*

Mycelial growth (radial expansion, fresh and dried weights) was measured on PDA and PDB media to determine the effectiveness of the extracts. As can be shown in table 4, MPLE extracts significantly impacted the radial mycelial development of *A. niger*. The first few days of incubation showed the greatest effect, but by day eight, the fungus had adapted to the lower concentrations, and the difference between the treated and control groups was no longer statistically significant.

Mycelial weight of *A. niger* also reduced with increasing dilution of MPLE extract as with radial growth. The greatest dose resulted in the most effective suppression.

Table 8 shows that the radial growth of *P. italicum* after being exposed to (MPLE). A higher MPLE dose was found to considerably reduce radial growth inhibition. More inhibition was done to *P. italicum* than *A. niger*. There was a drop of almost 50% in the larger concentration. As can be indicated in table 5, the MPLE led to a notable reduction in both the wet and dry weights.

Incubation time (days)	*A. niger*			*P. italicum*		
	0.0	0.5	1.0	0.0	0.5	1.0
2	7.5	4.5	3.25	4.25	3	2.125
4	8.5	6	3.9	8.58	6.65	2.88
6	8.5	5.5	5	9	6.25	3.25
8	9	7.5	5	9	6.75	3.5

Table 8: *Comparison of the effects of MPLE on the A. niger and P. italicum radial mycelial growth.*

Concentration (%)	Fresh weight		Dry weight	
	A	P	A	P
0.0	8.95	4.12	4.70	2.64
50	8.85	3.66	4.66	2.24
100	8.35	2.22	4.50	1.81

Table 9: *Comparison of the effects of MPLE on mycelial fresh and dry weight of the A. niger and P. italicum.*

Discussion

C. citratus is one of the most widely used plants in the world. *C. citratus* extracts have varied pharmacological effects. Antimicrobial, anti-inflammatory, antidiabetic, and anticancer properties are well known. *C. citratus* works alone or with other plant ingredients. Some research have characterized volatile and nonvolatile chemical compositions of *C. citratus* due to its ethno-pharmacological uses. *C. citratus* essential oil and extracts vary by geographical origin, age, and nature. *C. citratus* isolates tannins, sterols, terpenoids, phenols, ketone, flavonoids, and sugars.

Results from a Phytochemical study showed that many bioactive compounds, including flavonoids, glycosides, tannins, terpenoids, saponins, and steroids, were present. In several regions of the Sudan, the *Mahareeb* plant is used to make traditional medicines for treating a wide range of diseases (personal communication), which is supported by the presence of certain phytochemicals. The antioxidant properties of *Mahareeb* may be bolstered by the biological activity of these combinations. Plant extracts that contain flavonoids and terpenoids may have cell antioxidant properties. Antioxidant flavonoids work by scavenging and chelating free radicals [12]. Reducing peroxide arrangement *in vivo* and preventing lipid oxidation *in vitro* are two benefits of scavenging reactive oxygen species [13]. Providing hydrogen to radicals, terpenoids prevent rapid lipid oxidation [14].

The phenolic acid family is a powerful class of antioxidants. Free radicals are neutralized when encased by these compounds because they include at least one aromatic ring with a hydroxyl group [15]. As a result of their ability to scavenge free radicals, donate hydrogen, chelate metals, halt chain reactions in radical reactions, and eliminate singlet oxygen both *in vitro* and *in vivo*, polyphenols have strong antioxidant properties [16].

In addition to functioning as scavengers, hydrogen donors, and singlet oxygen quenchers, flavonoids are also important because of the high redox potential they possess. Metals can also be chelated by them. Flavonoids protect plants from a wide variety of threats, including UV light, fungal parasites, herbivores, diseases, and oxidative cell harm. Flavonoids have been linked repeatedly to a reduced risk of developing cancer and cardiovascular disease [17].

Two types of fungus (*A. niger* and *P. italicum*) and two types of bacteria (*E. coli* and *S. typhi*) had their growth inhibited by MPLE in this investigation. Both fungi's mycelial radial growth as well as their mycelial dry and fresh weights were drastically lowered by MPLE. Bullerman [18], Abdel-Rahim [19], and Al-Jali [20] all cite antifungal activity in plant extracts from a variety of plant species. Many plant extracts were discovered to suppress A growth. fungal species, including *A. niger*, *A. flavus*, *A. parasiticus* and others. However, Zainal., *et al*. [21] found that Mesquite (*Prosopis juliflora*) leaf litter extracts had a significant effect on A. yeasts like *C. niger* and *C. albicans.* Plant extracts of Cupress, Juniper, and Rose marry were found to be extremely efficient against *A. flavus* and *A. norvegicus*. According to Lai [22], MPLE is just one of several herbs and spices that can be used to extract useful chemicals. Humans have used spices and herbs to improve the flavor and aroma of their food for thousands of years.

The impact of MPLE on the development of two bacteria (*E. coli* and *S. typhi*) was also examined in this study. The test used both the dilution plate method and the Cup plate method (inhibition zone). Findings indicated that MPLE were extremely efficient against both

bacteria, with the impact being more pronounced in the case of *S. typhi*. Hassan., *et al*. [23] reported that *C. citratus* produced 1.5%v/w essential oil, indicating a significant concentration of this compound. When compared to the crude plant extract, this volatile oil had an effect on both Gram-negative and Gram-positive strains of the selected bacterial cultures, with the most antibacterial activity (as evaluated by the well diffusion method) shown against the Gram-positive strain *S. aureus*. Maryan [5] added that aqueous extract can be used as a long-term herbal antiplaque agent in maintenance phase, as an option for patients seeking natural product mouth rinses, or as a treatment for patients who experience negative effects from Chlorhexidine use. Our results were consistent with those of Valková., *et al*. (2022) [24], who found that LGEO was effective in preventing the development of a variety of tested bacteria, and that its effectiveness mostly depended on its concentration. We found that it inhibited growth of bacteria, even those that had developed resistance.

Spices and other plants have been shown to have antibacterial properties Sulieman., *et al*. [25]. About a hundred medicinal herbs utilized by traditional healers in Rwanda were examined by Vlietinek., *et al*. [26] for their antibacterial, antifungal, and antiviral activities. 45% were effective against *Staph aureus*, but only 2% were effective against *E. coli*. The bacteria *E. coli* and *Pseudomonas aerogenes* by 16% and the yeast *Candida albicans* by 7%. However, roughly 27% of the plant species evaluated showed antiviral activity. Extracts from 114 plant species from the Indonesian island of Sumatra were tested for antibacterial efficacy [27]. *Staph aureus* was the target of over 82% of the extracts, while only 35% were effective against *E. coli*. *E. coli*, while the growth of both *Saccharomyces cerevisiae* and *Fusarium oxysporum* was suppressed by roughly 20% of the extracts.

The antibacterial efficacy of many medicinal plants has been the subject of extensive research in Sudan. Ten Sudanese medicinal plants were examined by Ahmed [28], with their extracts put through tests against gram-positive and gram-negative bacteria and the yeast *Candida albicans*. *Staph aureus*, a gram-positive bacterium, was found to be most affected by the antibiotic treatment, followed by *E. coli* and *Candida albicans*. Many Sudanese herbs have been found to possess antimicrobial properties.

Conclusion

Antimicrobial activity of aqueous MPLE extracts against the examined species was demonstrated in the present investigation. The research also showed that the MPLE was most effective at inhibiting *P. italicum*. When applied at higher concentrations, the extract showed greater inhibitory action against the tested species. The results from this study suggest that the MPLE has phytochemicals that can neutralize free radicals through hydrogen donation, protecting against the negative effects that would otherwise occur.

The results of this study suggest that MPLE has potential as an antibacterial antifungal an antioxidant agents. It can also be used to add flavor to food and reduce microbial contamination in the food sector.

Bibliography

1. Shelef LA. "Antimicrobial effects of spices". *Journal of Food Safety* (1983): 227-232.
2. Sulieman AE., *et al*. "Evaluation of the antimicrobial and synergistic effect of selected medicinal plants in Hail area with antibiotic drugs". *Bioscience Biotechnology Research Communications* 10 (2017).
3. Nadeem A., *et al*. "Phytochemical profiling of antimicrobial and potential antioxidant plant: Nepeta cataria". *Frontiers in Plant Science* (2022).
4. Marya CM. "Antibacterial Efficacy of Lemongrass (*Cymbopogon citratus*). Extract against *S. mutans*". *Annals of Medical and Health Sciences Research* 12 (2022): 1-5.

5. Basera P., *et al*. "Analytical Investigation of *Cymbopogon citratus* and Exploiting the Potential of Developed Silver Nanoparticle Against the Dominating Species of Pathogenic Bacteria". *Frontiers in Microbiology* 10 (2019): 282.

6. Aluyor Emmanuel and Oboh Innocent. Preservatives: Traditional Preservatives - Vegetable Oils (2014).

7. Raaman N. Phytochemical techniques (2006).

8. Basano and Adeyemo S. "Phytochemical screening and antimicrobial assessment of *Abutilon mauritianum, Bacopa monnifera* and *Datura stramonium*". *Biokemistri* (2006): 18.

9. Chakraborty K and Paulraj R. "Sesquiterpenoids with free-radical-scavenging properties from marine macroalga *Ulva fasciata* Delile". *Food Chemistry* 122 (2010): 31-41.

10. Barry M., *et al*. "Quantitative measurement of iron stores with diethylenetriamine penta-acetic acid". *Gut* 11 (1970): 891-898.

11. Cruickshank R. "Medical Microbiology: A Guide to Diagnosis and Control of Infection". E and S Livingston Ltd., Edinburgh and London (1975): 888.

12. Zou Z., *et al*. "Antioxidant activity of Citrus fruits". *Food Chemistry* 196.1 (2016): 885-896.

13. Fawole OA., *et al*. "Anti-inflammatory and phytochemical properties of twelve medicinal plants used for treating gastro-intestinal ailments in South Africa". *Journal of Ethnopharmacology* 123.2 (2009): 237-243.

14. Lu Y., *et al*. "Antioxidant activity determination of citronellal and crude extracts of *Cymbopogon citratus* by 3 different methods". *Journal of Pharmacy and Pharmacology* 5.4 (2014): 395-400.

15. Gnanamani K., *et al*. "Antibacterial activity of two plant extracts on eight burn pathogens". *Journal of Ethnopharmacology* 86.1 (2003): 59-61.

16. Carro RT., *et al*. "Antioxidant and anti-inflammatory activities of *Frankenia triandra* (J. Rémy) extracts". *South African Journal of Botany* 104 (2016): 208-214.

17. López-Cobo A., *et al*. "Determination of phenolic compounds and antioxidant activity of a Mediterranean plant: the case of *Satureja montana subsp. kitaibelii*". *Journal of Functional Foods* 18 (2015): 167-1178.

18. Bullerman LB. "Inhibition of aflatoxin production by Cinnamoun". *Journal of Food Science* 39 (1974): 1163-1165.

19. Abdel-Rahim AM., *et al*. "Survey of some cereal grains and legume seeds for aflatoxin contamination in the Sudan". *Zentralbl Bakteriol Mikrobiol Hyg A* 89 (1989): 75-79.

20. Al-Jali ZI., *et al*. "Contamination of seeds of some crops with alfa-toxins in the Jabel Al-Akhdar region". Proceeding of the 6th. Arab Congress of Plant Protection, Beirut, Lebanon (1997): 294.

21. Zainal AS., *et al*. "Antimicrobial Substance(s) in the leaf litter of the xerophyte *Prosopis juliflora*". *Zentralblatt FÜr Mikrobiologie* 143 (1988): 375-381.

22. Lai PK. "Antimicrobial and chemopreventive properties of herbs and spices". *Current Medicinal Chemistry* (2004): 1451-1460.

23. Hassan ZY., *et al*. "Antimicrobial of lemongrass (*Cymbopogon citratus* L.) volatile oil and cytotoxic effects against L20B and MCF-7cellline". *Biodiversitas* 23.10 (2022): 5298-5301.

24. Valková V., *et al*. "Cymbopogon citratus Essential Oil: Its Application as an Antimicrobial Agent in Food Preservation". *Agronomy* 12 (2022): 155.

25. Sulieman AME Abdallah., *et al*. "Spices as Sustainable Food Preservatives: A Comprehensive Review of Their Antimicrobial Potential". *Pharmaceuticals* 16 (2023): 1451.

26. Vlietinek JA., *et al*. "Screening of hundred Rwandes medicinal plants for antimicrobial and antiviral properties". *Journal of Ethnopharmacology* 46 (1995): 31-47.

27. Ahmed MM. "Molluscicidal and antimicrobial activity of certain Sudanese cucurbitacea plants". (Cited from Ahmed S.A., 2007) (2002).

28. Ahmed MM. "Phytochemical, antimalarial, molluscicidal and antimicrobial activity of selected Sudanese Medicinal plants with Emphasis on: *Nigella sativa* L. seeds". Ph.D. Thesis, University of Gezira (2004): 75-78.

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