Phytochemical Study and Evaluation of Iraqi Fennel Seed Oil as Antibacterial of Urinary Tract Infection

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Abstract

Objective: This study were designed by investigation of active compounds of the fennel seeds in Iraqi plant and determined the terpenes compounds by gas chromatography analysis and finally shown the biological activity of the volatile oil of fennel seeds upon different pathogenic bacteria which isolated from patients suffered from urinary tract infection.

Methods: in this study we used different methods and different solvent for extract the active compounds such as clavenger for volatile oil, decoction and Soxhlet for main other active compounds.

Results: The results of this study had shown that bacteria such as Pseudomonas aeruginosa, Streptococcus spp and Salmonella spp. were more resistant to The volatile oil of fennel seed than bacteria such as., Staphylococcus aureus and E. coli. five compounds were identified from the n-hexane extract of volatile oil of fennel seeds. Anethole was in maximum content (43.13 %) in retention time 13.762, followed by Estragole (40.84 %) in retention time 12.062, 1,6, octadien-3-ol,3,7-dimethyl (3.71%) in retention time 9.937, 1-fenchone compounds (3.49%) in retention time 9.645 and the minimum one is d-limonene (1.28%) in retention time 8.435.

Keywords: fennel seeds, antibacterial activity, phytochemical screening, GC analysis

Introduction

Medicinal plants are nature's gift to human beings to make healthy lifestyle free from diseases, and play an important role to look after our health ¹. They are considered to be much safer and certain cure in the treatment of several conditions. Iraq is one of the countries that consider traditional medicine is very important, Iraq, like to any other country in the world, the use of medicinal plants betake back thousands of years^(2,3). The strategic location, the climatic state of affairs and geographical diversity make it suitable place for cultivate of many medicinal herbs as source of treatment or protection of many diseases. The *Foeniculum vulgare* consists one of those plants and consider as one of the most important aromatic plant^(4,5).

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There are many traditional medicine for part used (fennel seeds) of F. vulgare consider as carminative and its mainly used with purgatives . there are similarity between fennel water and dill water both used to correct flatulence of infants (gripe water) ⁶. The volatile oil or called essential oil one of the most active constituents in fennel seed it's made as flavoring agent in additional used as medicine which reported it have been many components like estragol, trans-anethole, fenchone, and α -phellandrene^(7,8). the major active compounds of F. vulgare are some of the most important active constituents such as phenols and phenolic glycosides 9 . The phenolic acids structures like 3-O-Caffeoylquinic acid, 4-O-caffeoylquinic acid, 5-O-caffeoylquinic acid, and the flavonoids like eriodictyol-7-rutinoside and quercetin-3-rutinoside, also isolated others constituents from F. vulgare were Quercetin-3-O-galactoside, kaempferol-3-O-rutinoside and kaempferol-3-Oglucoside 10 .The phenolic compounds which isolated from F. vulgare were associated with the prevention

of some of dangerous diseases because they are caused by oxidative stress like cancer and inflammation 11 . In many studies reported the volatile oil which isolated from fennel seeds of F. vulgare act as antibacterial effect against many pathogens bacteria such as Escherichia coli, Bacillus megaterium and Staphylococcus aureus , E. coli , Listeria monocytogenes and S. aureus $^{(12,13)}$.

Material and Method

Collection of plant material

The fennel seeds plant was authenticated by National Iraqi Herbarium, Botany Directorate at Abu-Ghraib. The seeds of the plant was washed and dried in the shade at room temperature at Pharmacognosy and medicinal plants department-college of pharmacy-Mustansiriyah University, for (7 days) until crisp and then was grinded by mechanical mills and weighed according to previous studies ¹⁴.

Extraction

The essential oil content of fennel seeds was extracted by hydro distillation method by the use Clevenger apparatus (100 g) of the plant material was hydrodistilled by adding (500 mL) of distilled water D.W. in round flask bottom, the plant was left boiling for three hours; the volatile oil was collected after observing that there is no increase in volatile oil was achieved ¹⁵.

Preliminary phytochemical investigation

The extracts of fennel seeds from Baghdad plants was screened according to standard procedures of qualitative investigation to identify the major classes of natural secondary metabolites such as Tannins, Saponins , Flavonoids, Terpenes , Alkaloids , Anthraquinone glycoside and Carbohydrate (16,17).

Antimicrobial screening

The essential oil of fennel seeds was tested for antimicrobial activity of six different pathogenic bacteria which were **Staphylococcus aureus**, **Streptococcus spp.**, **Pseudomonas aeruginosa**, **Salmonella spp.**, **Serratia** and **E. coli** which isolated from patients suffered urinary tract infections by using diffusion well agar method ¹⁸.

Results and Discussion

Phytochemical study:

The preliminary investigation results of active compounds were referred to different active compounds presented according different solvents were used in this study. Ethanol 70% extract was contained Carbohydrate, tannin and flavonoids. while terpene investigated with hexane extract [table1]. Also used Decoction method with water as a solvent and Ethanol 70% by Soxhlet apparatus extract was investigated the others active compounds.

The GC-MS analysis of the individual extracts indicated a complex mixture of constituents of different chemical classes. five compounds were identified from the n-hexane extract of volatile oil of fennel seeds . [table 2 , figure 1]. Anethole was in maximum content (43.13 %) in retention time 13.762, followed by Estragole (40.84 %) in retention time 12.062 , , 1,6, octadien-3-ol ,3,7-dimethyl (3.71%) in retention time 9.937 , l-fenchone compounds (3.49%) in retention time 9.645 and the minimum one is d-limonene (1.28%) in retention time 8.435 .[Table 2] .

The volatile oil of fennel seed possessed antimicrobial activity 0.5,0.25, 0.125, 0.06 and 0.03concentration against all microorganisms were used in this study table (3) and the value of the concentration activity was reach to 20,18,18,24,20 and 12 as inhibition zone a reach at 0.5 concentrated against Staphylococcus aureus, Streptococcus spp., Salmonella spp., Serratia, E. coli and Pseudomonas aeruginosa compared with ciprofloxacin as control which gave the higher activity and reach to 24 at 0.5 concentrated [table 3]. The MIC values for The volatile oil can vary depending on factors such as: chemical compositions differences between herbs collected in different countries (differences in the climate, soil composition, age and vegetative cycle stage), as well as differences in strains of microorganisms used (standardized or clinical isolates). The results of this study had shown that bacteria such as Pseudomonas aeruginosa, Streptococcus spp and Salmonella spp. were more resistant to The volatile oil of fennel seed than bacteria such as., Staphylococcus aureus and E. coli.

Table 1: Qualitative Phytochemical Analysis of the Extracts of

Constituents	Results
Tannins	+ve
Saponins	-ve
Flavonoids	+ve
Terpenes	+ve
Alkaloids	-ve
Anthraquinone glycoside	-ve
Carbohydrate	+ve

GC-MS analysis

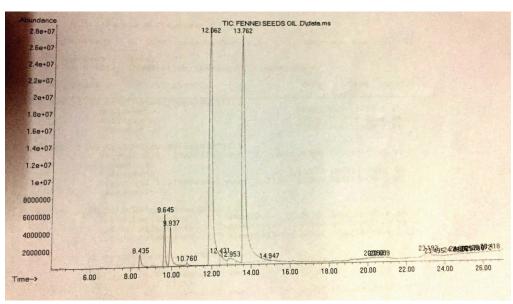


Figure (1) GC-MS chromatographic profile of fennel seeds oil

Table2: Chemical composition of fennel seeds oil

Retention time	Content %	Compounds name	Chemical structure	Molecular weight	Formula	Similarity
8.435	1.28	d-limonene	CH ₃ H ₃ C C CH ₂	136.24 g/mol	C10H16	99
9.645	3.49	l-fenchone	0	152.23 g/mol	C10H16O	91

Cont.. Table2: Chemical composition of fennel seeds oil

9.937	3.71	1,6, octadien-3-ol ,3,7-dimethyl	HO	154.25 g·mol−1	C10H18O	97
12.062	40.84	Estragole	CH ₃ O	148.2 g/mol	C10H12O	98
13.762	43.13	Anethole	H ₃ CO	148.2 g/mol	C10H12O	98

Table 3: Antibacterial activity of volatile oil of fennel seed

	Concentration of inhibition zone(mm)					
Bacterial isolation	1/2	1/4	1/8	1/16	1/32	ciprofloxacin (control)
Staphylococcus aureus	20	17	15	12	10	22
Streptococcus spp.	18	13	13	10	8	20
Salmonella spp.	18	14	10	8		20
Serratia	24	20	18	14	10	24
E. coli	20	16	8			20
pseudomonas aeruginosa	12	10				20

Conclusion

The natural extract from medicinal plants which proved to be potentially effective as (volatile oil of fennel seeds) can be used as alternative preventives naturally against many pathogenic bacterial specially in urinary tract infections

Financial Disclosure: There is no financial disclosure.

Conflict of Interest: None to declare.

Ethical Clearance: All experimental protocols were approved under the College Of Pharmacy , Mustansiriyah University, Iraq and all experiments were carried out in accordance with approved guidelines.

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