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Antioxidant and antibacterial properties of anise (Pimpinella anisumL.)- An Overview

[1]Chandra, [2]Noorulla, [3]Pravin Kumar, [4]Yokeshwaran
(1) Associate Professor, PSV College of Pharmaceutical Science and Research, Krishnagiri, India.
(2,3,4) Ug Students, PSV College of Pharmaceutical Science and Research, Krishnagiri, India.

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ABSTRACT:
The aim of the study was to evaluate the chemical composition and antibacterial effect of essential oil of Pimpinella anisum [anise] against Pseudomonas aeruginosa and Bacillus subtilis. The essential oil of anise, extracted from the dry ripe fruits of Illicium verum and Pimpinella anisum, contains anethole, an active chemical compound that showed several functional properties including antimicrobial, antioxidant, hypoglycemic, hypolipidemic and oestrogenic properties. As a screen test to detect antibacterial properties of the essential oil, agar disk and agar well diffusion methods were employed. The results indicated that the most substance found in P. anisum essential oil was Trans-anethole (89.7 %), also the essential oil of P. anisum in 0.003 and 0.007 g/ml concentrations has prevented from the growth of the P. aeruginosa and B. subtilis, respectively. Thus, the research represents the antibacterial effects of the ethnomedical herb against both of bacteria. The highest percentage of radical-scavenging activity (91.3±1.8%) was recorded for the ethanolic extract of seeds at a concentration of 0.3 mg/ml, followed by the aqueous extract of seeds (82.0±1.2%), whereas the aqueous extract of aerial parts demonstrated the lowest frequency of radical-scavenging activity (39.0±1.7%) at the same tested concentration. The largest inhibition zones were determined to be 21.0±1.2, 18.3±1.5, 9.7±1.2, and 7.0±1.2 mm for Bacillus cereus, Staphylococcus aureus, Salmonella typhimurium, and Escherichia coli, respectively. Overall, the results demonstrated the superiority of seed extracts over aerial part extracts. The results also indicated the stronger activity of ethanolic extracts compared with aqueous extracts. Also, further evaluation is necessary on potential of it as an antibacterial agent in topical or oral applications. Fractionation and characterization of active molecules will be the future work to investigate.

Keywords: Pimpinella anisum, agar well diffusion, antimicrobial activity, antioxidant property, Essential oil, Chemical composition, Antibacterial effect.

I. INTRODUCTION:

Antibiotics provide the primary basis for the treatment of microbial (bacterial and fungal) infections. Since the detection of these antibiotics and their use as chemotherapeutic agents, there was a belief in the medical fraternity that this would cause to the presumptive eradication of infectious diseases. But overuse of antibiotics has become the main factor for the emergence and dissemination of multi-drug resistant strains of different groups of microorganisms. Nowadays, multiple drug resistance has developed due to indiscriminate use of commercial antimicrobial drugs commonly used in treatment of infectious diseases. Herbs and spices are invaluable resources useful in daily life as food additives, flavours, fragrances, pharmaceuticals, colours or directly in medicine. These plants contain medicinal properties which make them potent to cure or prevent diseases. Some medicinal plants used in traditional Iranian medicine are efficient in treating diverse ailments caused by bacterial and oxidative stress. Many plants have been used because of their antimicrobial traits, which are due to compounds synthesized in the secondary metabolism of the plant. Free radical-induced oxidative damage plays a major part in the development of many chronic and degenerative ailments such as cancer, stroke, arthritis, autoimmune disorders, and cardiovascular diseases. Oxidative stress which occurs when a biological system is incapable of keeping up with the detoxification of the free radicals can damage all kinds of molecules including important macromolecules such as nucleic acids, proteins, lipids, and carbohydrates. The antioxidants neutralize the free radicals mainly through their free radical scavenging property, thereby mitigating the effect of oxidative stress. Although some antioxidants are produced naturally by the human body, the body cannot manufacture the principal micronutrient antioxidants such as vitamin E (α-tocopherol), vitamin C (ascorbic acid), and B-carotene. Therefore, they must be supplied in the diet.

Infectious diseases are another major problem worldwide. Synthetic antibacterial drugs are not only expensive and inadequate but are also often with side effects. Moreover, because of the overuse and/or misuse of antibiotics through the past decades, many pathogens have evolved resistance to them via natural selection. This problem is progressively becoming more and more intense in terms of frequency and severity. Nowadays, resistance is seen to nearly all antibiotics that have been developed.

The bioactive natural compounds represent a remarkable source of novel antioxidants and antimicrobial agents.

Phytochemicals such as vitamins (A, C, E and K), carotenoids, terpenoids, flavonoids, polyphenols, alkaloids, tannins, saponins, pigments, enzymes and minerals that have antimicrobial and antioxidant activity. The specific function of many phytochemicals is still unclear; however, a considerable number of studies have shown that they are involved in the interaction of plants/pests/diseases. Phytochemical studies have attracted the attention of plant scientists due to the development of new and sophisticated techniques. These techniques played a significant role in the search for additional resources of raw material for pharmaceutical industry. Essential oil is a concentrated hydrophobic liquid containing volatile aroma compounds from plants. The main constituents of essential oils –mono- and sesquiterpenes including carbohydrates, phenols, alcohols, ethers, aldehydes and ketones are responsible for the biological activity of aromatic and medicinal plants as well as for their fragrance. Due to these properties, spices and herbs have been added to food since ancient time, not only as flavouring agents but also as preservatives. Plants and their essential oils are potentially useful sources of antimicrobial compounds.

Antimicrobial screening of plant essential oils and phytochemicals, then, represents a starting point for antimicrobial drug discovery. Essential oils are effective on a wide range of Gram-negative and positive bacteria such as Staphylococcus aureus, Bacillus subtilis, and Escherichia coli. P. anisum (Anise, also called aniseed), a plant belonging to the Umbelliferae family, is one of the oldest medicinal plants. It is an annual grassy herb with 30–50 cm high, white flowers, and small green to yellow seeds, which grows in the Eastern Mediterranean Region, West Asia, the Middle East, Mexico, Egypt, and Spain. This plant is primarily grown for its fruits (aniseeds) that harvested in August and September. Its flavour has similarities with some other spices, such as star anise, fennel, and licorice. It was reported that P. anisum had several therapeutic effects such as neurologic, digestive, gynecologic, fungal disease, and respiration disorders. The most substance found in P. anisum essential oil is Trans-anethole. The aim of this study was to screen the in vitro antibacterial activity of the plant essential oil against some bacteria including P. aeruginosa and B. subtilis.

II. MATERIALS AND METHODS:

Anise samples collection:

The medicine plant collected from Kermanshah. The sample was cleaned from any strange, plants, dust, or any other contaminants. The seeds were surface sterilized with 20% commercial Clorox (5% NaOCl) containing 0.1% Tween 20 for 20 min and thoroughly washed with sterilized, distilled water three times. Aseptic seeds were cultured on MS medium without growth regulators and allowed to germinate under laboratory conditions.

Essential oil extraction:

Essential oil from fresh, clean, weighed aerial part P. anisum fruits extracted by hydro-steam distillation using the Clevenger apparatus were collected and stored in sterile vials. Briefly, 100 to 150 g of plant was introduced in the distillation flask (1L), which was connected to a steam generator via a glass tube and to a condenser to retrieve the oil. This was recovered in a funnel tube. Aromatic molecules of the essential oil were released from the plant material and evaporated into hot steam. The hot steam forced the plant material to release the essential oil without burning the plant material itself. Then, steam containing the essential oil was passed through a cooling system in order to condense the steam. The steam was applied for 3h. After settling the recovered mixture, essential oil was withdrawn. The supernatant essential oil was filtered through anhydrous Na_2SO_4 to dry the yielded essential oil. Afterward, the essential oil was collected in tightened vials and stored in a refrigerator. For the antimicrobial activity test, several dilutions of the essential oil were done using dimethyl sulfoxide (DMSO).

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Preparation of extracts:

Seeds and aerial parts were dried in oven at 40°C for 24h. Afterwards, the two dried samples were ground into a fine powder. Ethanol and distilled water were used as solvents for the preparation of extracts. Ten grams of each of the two samples were soaked separately in 100ml of each solvent and kept in a shaker for 2 days. The obtained mixtures were filtered through Whatman filter paper no.1. The filtrates were evaporated to dryness and the resulting viscous powders were dissolved in the same extract solvents to get a final concentration of 50mg/ml stock solutions. These stock solutions of the four extracts were stored at 4°C until used.

Gas chromatography mass spectrometry (GC/MS):

P. anisum essential oil was analysed using GC/MS (Shimadzu capillary GC-quadrupole MS system QP 5000) with two fused silica capillary column DB-5 (30 µm, 0.25 mm i.d, film thickness 0.25 µm) and a flame ionization detector (FID) which was operated in EI mode at 70 eV. Injector and detector temperatures were set at 220°C and 250°C, respectively. One microliter of each solution in hexane was injected and analyzed with the column held initially at 60°C for 2 min and then increased by 3°C/min up to 300°C. Helium was employed as carrier gas (1 ml/min). The relative number of individual components of the total essential oil is expressed as percentage peak area relative to total peak area. Qualitative identification of the different constituents was performed by comparison of their relative retention times and mass spectra with those of authentic reference compounds and mass spectra.

Culture media:

Mueller-Hinton Agar (Müller-Hinton agar is a microbiological growth medium that is commonly used for antibiotic susceptibility testing) was prepared according to the manufacturer's instruction (Oxoid, UK), autoclaved and dispensed at 20 ml per plate in 12 x 12cm Petri dishes. Set plates were incubated overnight to ensure sterility before use. Evaluation of antimicrobial activities

Agar disk diffusion and agar well diffusion was used as screen tests to evaluate antibacterial property of essential oil of P. anisum based on standard protocol. The solution of the essential oil was yielded in 1g/ml from which sixfold serial dilutions (v/v) were prepared. 60 µl of each dilution was poured on each disk and well in order. After a period of 24 hours' incubation, the diameters of growth inhibition zones around the disks and wells were measured. DMSO was used as negative control whereas kanamycin and cephalexin were used as positive controls in case of E. coli and S. aureus, respectively. Minimum inhibitory concentration (MIC) means the lowest concentration of the probable antimicrobial agent which prevents growing of bacteria (regardless of killing the bacteria or stopping the growth of them). The lowest dilution which no gross microbial growth has been seen indicates MIC. Minimum bactericidal concentration (MBC) means the lowest concentration of the agent which causes death to test bacteria. The last can be revealed by pouring 60 µl of MIC tube and six dilutions before contents on agar plate. In the case, after incubation period, the lowest concentration which makes no growth indicates MBC. For determination of MIC value, macrobroth dilution method was applied. Interpretation of the results was done due to national accepted letter

Antioxidant assay:

The antioxidant capacity was studied through the evaluation of the free radical-scavenging effect on the 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical. The determination was based on the method described by Brand-Williams et al. [18] with slight modification. Briefly, 500µl of each extract (at different concentrations, that is, 0.05, 0.1, 0.2, and 0.3mg/ml) was mixed with 2.5ml of methanolic solution of DPPH (0.1mmol/l). The mixture was kept in the dark for 30min before the absorbance at 517nm was measured against a control solution of methanol and DPPH without extracts. Ascorbic acid was used as positive control. The results were expressed as percentage of the DPPH radical. Radical scavenging activity (RSC%) was calculated according to the following equation:

RSC ð Þ ¼ A control A sample ×100

where $A_{control}$ is the absorbance of DPPH without extract, while A_{sample} is the absorbance of the extracts.

Antimicrobial assay:

Numerous wild plants show antimicrobial properties, particularly in blocking the bacterial reproduction and their development. The antibacterial effects of the individual components of anise oil varied depending upon their chemical structure, functional groups and configuration as well as doses used.

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Antimicrobial activity tests were conducted by using the agar well diffusion method. Fifteen millilitres of the nutrient agar medium were added into petri dishes. The melted and tempered (40°C) agar was previously inoculated with 200µl of the target microorganism cell suspension. The freshly grown suspensions were prepared by diluting microbial cultures of the target strain to achieve a microbial concentration of 10^8 CFU/ml. The agar plates were solidified for 1h and then, using a sterile cylinder, wells of 8-mm diameter were made and filled up with 100µl of the diluted stock solutions (plant extracts at a concentration of 1.25, 2.5, and 5mg/ml). Wells containing solvents (100µl) were used as a negative control, while wells containing tetracycline (a standard antimicrobial) served as a positive control (50µg/ml). The plates were incubated for 48h at 28°C. The antimicrobial activities of the anise extracts were evaluated by measuring the inhibition zones around the wells. The inhibition zones were measured with a ruler and were determined by a clear zone of at least 2mm around the well.

Statistical analysis:

Statistical analysis was performed using IBM SPSS statistics subscription. One-way analysis of variance was used for statistical analysis. Five samples were used for each treatment and each experiment was repeated three times. Mean ± SE were obtained from the analysis for each treatment. Data were presented as mean ± SE and were compared with Tukey's test at a 5% probability level.

Chemical composition:

The essential oil (anisi aetheroleum) extracted from steam distillation of ripe fruits of both I. verum and P. anisum contains trans-anethole from 80% to 95% or more (responsible for its characteristic taste and smell, as well as for its medicinal properties, followed by chavicol methyl ether (estragole), anisaldehyde and cis-anethole.

P. anisum composition includes, in addition, coumarins (umbelliferon, umbelliprenine, bergapten, and scopoletin), lipids (fatty acids, beta-amyrin, stigmasterol and its salts), flavonoids (flavanol, flavone, glycosides, rutin, isoorientin, and isovitexin), proteins and carbohydrates. P. anisum is well known as a carminative and an expectorant, and it is also used to decrease bloating, especially in pediatric patients. At higher doses, it is used as an antispasmodic and antiseptic, and in vitro study have also reported an antimicrobial action.

The fruits of I. verum contain, besides volatile oil, resin, fat, tannin, pectin and mucilage, making the plant efficacious in the treatment of dyspeptic complaints, catarrhs of the respiratory tract, rheumatism and otalgia. The fruits are also used for their antiseptic, digestive, diuretic, and deodorant properties. I. verum is also used as aromatic spice for food and cosmetic preparations, representing an ingredient of the traditional five-spice powder (a mixture of star anise, clove, cinnamon, pepper and fennel) of Chinese cooking, especially for meat and soup; the seeds are used in baked goods and confections and in the West, I. verum is added in fruit compotes and jams and in the manufacture of anise-flavoured liqueurs (anisette).Composition of the plant in RI (Retention Index) including α-pinene (0.1%), sabinene (0.01%), myrcene (0.01), α-phellandrene (0.01%), p-cymene (0.1%), limonene (0.8%), 1,8-cineole (0.1%), cis-b-ocimene (0.01%), fenchone (4.62%), camphor (0.23%), methyl chavicol (2.15%), endo-fenchyl acetate (0.1%), cisanethole (0.43%), p-anisaldehyde (0.41%), trans-anethole (89.7%), respectively. The results indicate that the most substance found in P. anisum essential oil is transanethole, in contrast, sabinene, myrcene, α-phellandrene, and cis-b-ocimene are the least constituents discovered in the essential oil.

Antibacterial Activity:

Numerous wild plants show antimicrobial properties, particularly blocking the bacterial reproduction and their development. The antibacterial effects of the individual components of anise oil varied depending upon their chemical structure, functional groups and configuration as well as doses used. Singh et al. showed that the antibacterial activity of P. anisum can exploited against Staphylococci aureus responsible for bases, sepces and skin infection; Streptococcus haemolyticus causing infection of the throat and nose; and Bacillus subtilis responsible for infection in immune compromised patients. The oil can also be used to control Pseudomonas aeruginosa which causes hospital acquired infection; Escherichia coli, responsible for urinogenital tract infections and diarrhoea; Klebsella spp and Proteus vulgaris. These results are confirmed by Abu-Darwish, showing the antibacterial activity of anise by demonstrating that the plant has a relevant role against clinical and standard strains of S. aureus and E. coli. Anise plant, thanks to its active

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compounds, could be used in pharmaceutical preparations as natural antibiotics.

Other Effects:

The effects of volatile oil of this plant on the relaxation of the tracheal isolated muscles of guinea pig and after them, the antispasmodic effect of anise's compounds was evaluated in several other studies. An example of a latest study on antispasmodic effect was conducted by Tirapelli and coworkers, which demonstrated the reduction of anococcygeus smooth muscle contraction in rat induced by acetylcholine, due to three hidroalcoholic extract of P. anisum at different concentration (40%, 60% and 80%). The effect demonstrated by this study justifies its use in the folk medicine as an antispasmodic agent. Despite the healthy effect of P. anisum and I. verum demonstrated by scientific literature, that conferred to the plants the characteristic of "harmless medicine", some cases of intoxication were registered.

A major part of these cases, among infants and child, were reported because the I. verum tea is often used in various cultures for the treatment of infant colic pains. In particular, the toxicity is due to the contamination of I. verum by Japanese star anise or Illicium anisatum L, which contains several neurotoxins (anisatin, neoanisatin, and pseudoanisatin) causing serious neurologic and gastrointestinal symptoms. Furthermore, the effect of I. anisatum on people exposed to the substance contained in the contaminated beverage was serious, consisting of signs of acute-onset irritability, jitteriness, clonus or myoclonus, increased deep tendon reflexes, nystagmus, vomiting, and seizures. In this perspective, the knowledge of the effect of eventual contamination of herbal teas with Japanese star anise is a cardinal point to prevent such events.

III. RESULTS:

Agar disk diffusion test:

P. aeruginosa and B. subtilis were sensitive to P. anisum essential oil. In case of P. anisum, the most sensitive bacterium was P. aeruginosa and B. subtilis by developing the halo around which in 19 mm in diameter in dilution 0.031 g/ml. There was inhibition zone in P. aeruginosa due to dilution 0.002 g/ml whereas there wasn't inhibition zone in B. subtilis. No inhibition zone was observed due to DMSO. Growth inhibition zones due to different dilutions are listed in table.

Dilution(g/ml)	Inhibition zone in disk diffusion (mm)	
Microorganism	P. aeruginosa	B. subtilis
Positive control	22	22
1/32 (0.031)	19	19
1/64 (0.015)	19	17
1/128 (0.007)	12	12
1/256 (0.003)	9	8
1/512 (0.002)	8	0
1/1024 (0.001)	0	0
Negative control	0	0

Agar well diffusion test:

Regarding P. anisum essential oil, the widest zone was seen in 0.031 g/ml, due to P. aeruginosa and B. subtilis (16 mm). It was no growth inhibition in 0.001 g/ml and less for both of bacteria. The data are discoverable in table.

Dilution(g/ml)	Inhibition zone in well diffusion (mm)	
Microorganism	P. aeruginosa	B. subtilis
1/32 (0.031)	16	16
1/64 (0.015)	11	11
1/128 (0.007)	9	8
1/256 (0.003)	8	8
1/512 (0.002)	8	0
1/1024 (0.001)	0	0
Negative control	0	0

MIC and MBC ascertaining:

The values of MIC are 0.007 g/ml and 0.003 for B. subtilis and P. aeruginosa, respectively. But the values of MBC are same for B. subtilis and P. aeruginosa and they are 0.015 g/ml. As the table showed, P. anisum essential oil have prevented the growth of P. aeruginosa and B. subtilis. Also, by increasing the concentration of P. anisum essential oil, the inhibition zone increased ($p \leq 0.001$). The results determined that in tested bacteria, there was a significant difference ($p \leq 0.001$) in terms of sensitivity to the essential oil.

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Microorganism	P. aeruginosa	B. subtilis
MIC	1/256(0.003)	1/128 (0.007)
MBC	1/64 (0.015)	1/64 (0.015)

IV. DISCUSSION:

In spite of the current interest in drug discovery by molecular modelling, combinatorial chemistry and other synthetic chemistry methods, plant-derived compounds are still substantiating to be an important source of medicines for human being. The significance and uses of plants in modern drug discovery has been recounted in recent reports. Plant essential oils have been used for many thousands of years, in food preservation, pharmaceuticals, alternative medicine and natural therapies. In recent years, much attention has been devoted to natural bioactive compounds and their health benefits. Plant extracts are commonly rich in phenolic compounds such as flavonoids, phenolic acids, and tannins that have multiple biological effects including antioxidant and antimicrobial properties. In vitro studies in the work showed that the essential oils inhibited bacterial growth, but their effectiveness varied. The antimicrobial activities of many essential oils have been previously reviewed and classified as strong, medium or weak.

Phenolic compounds are hydrogen donors capable of directly scavenging free radicals and reducing oxidative damage, which makes them potent antioxidants. In addition to the antioxidant activity, phenolic compounds act as antimicrobial agents via several mechanisms including the disruption of microbial membranes. In the present study, the antioxidant and antimicrobial activities of anise extracts may be attributed to their phenolic contents since numerous phytochemical studies indicated the presence of noticeable amounts of phenolic compounds in anise\

In this investigation, solvent extracts prepared from the seeds of anise exhibited a significantly higher activity than those prepared from the aerial parts and of anise. The varied effects of extracts from different parts of anise could be attributed to the differences in their phytochemical constituents. Different parts contain different bioactive compounds at different levels. This result is in consistent with that of Embong and his colleagues, who investigated the components of the whole plants and the seeds of Pimpinella anisum and reported that the major oil constituent (the flavonide trans-anethole) − which is widely known to have strong biological activities − was 57.4% of whole plant and 75.2% of seed oil. A similar trend was observed also in other medicinal plants such as chicory. Jurgonski and his colleagues investigated the chemical composition of the seed, peel, leaf, and root extracts and found that the seed extract was the richest source of minerals, fat, protein, and most importantly, phenolic compounds

P. anisum is primarily grown for its fruits (aniseeds) that harvested in August and September. Aniseeds contain 1.5–5% essential oil and used as flavouring, digestive, carminative, and relief of gastrointestinal spasms. Consumption of aniseed in lactating women increases milk and reliefs their infants from gastrointestinal problems P. anisum is one of the medicinal plants which have been used for different purposes in traditional medicine of Iran. So far, different studies were performed on the extracts and essential oil of P. anisum to identify the chemical compounds and pharmacological properties of the plant, and various properties such as antimicrobial, antifungal, and antibacterial. The medicinal use of aniseed is largely due to antispasmodic, secretolytic, secretomotor and antibacterial effects of its essential oil.

P. anisum extracts and oil as well as some oil components, exhibited in vitro strong inhibitory activities against the growth of a wide spectrum of bacteria and fungi known to be pathogenic for man and other species[10]. Concerning the method of essential oils and preventing from using high temperature to decrease the rate of destruction of effective herbal compound. 15 compounds representing 98.78% of the total essential oil composition of P. anisum were identified using mass gas-chromatograph, these compounds including α-pinene, sabinene, myrcene, αphellandrene, p-cymene, limonene, 1,8-cineole, cis-bocimene, fenchone, camphor, methyl chavicol, endofenchyl acetate, cis-anethole, p-anisaldehyde and trans-anethole. The most substance found in P. anisum essential oil was trans-anethole with 89.7 %. Transanethole is an alkyl alkyl-phenol ether. Both the cis and trans isomers of trans-anethole occur in nature with the trans isomer always being the more abundant.

Natural anethole occurs in P. anisum essential oils and in star P. anisum essential oils. It has been shown to block grow of inflammation and carcinogenesis. Anethole has potent antimicrobial properties, against bacteria, yeast, and fungi. Reported antibacterial properties include both bacteriostatic and bactericidal action against

Salmonella enteric, but not when used against Salmonella via a fumigation method.

Findings from the current study revealed that essential oil of P. anisum has potential inhibitory effects on P. aeruginosa and B. subtilis. In agar disk diffusion test, the maximum activity of P. anisum essential oil against P. aeruginosa and B. subtilis was 19 mm, which is comparable with a zone of inhibition exhibited by kanamycin (22 mm) and cephalexin (16 mm). Also, these results indicated that P. anisum essential oil in 0.003 and 0.007 g/ml concentrations has prevented from the growth of the P. aeruginosa and B. subtilis, respectively. In the study, the levels of MBC were same in both of bacteria. Thus, the research represents the antibacterial effects of the medical herb on Gram-negative and Gram-positive pathogenic bacteria. Several authors have mentioned the antimicrobial activity of P. anisum.

The antibacterial activities of the aqueous, 50% (v/v) methanol, acetone and petroleum ether extracts of P. anisum fruits were tested against 4 pathogenic bacteria (Staphylococcus aureus, Streptococcus pyogenes, Escherichia coli, and Klebsiella pneumoniae) by disk diffusion method.

Solvent types may also affect the biological activities of the extracts. Solvents differ in the extraction capabilities depending on their polarity and on the solute's chemical structure. Different solvent extracts have different soluble phytoconstituents in different amounts and hence, they have varying degrees of biological activities.

In the present study, ethanol extracts demonstrated higher activity compared with the corresponding water extracts. This result suggests that most of the bioactive constituents in anise are soluble in ethanol. For example, anethole (the principal active constituent in anise essential oil) is known to be slightly soluble in water whereas it exhibited high solubility in ethanol. Our result is in contrast with the report of Galkin et al.

V. CONCLUSION:

Anise extracts show a satisfactory antioxidant and antimicrobial powers suggesting their potential use to treat infectious diseases and to fight free radicals.

The results demonstrate the superiority of seed extracts over aerial part extracts. The results also indicate the stronger activity of ethanolic extracts compared with aqueous extracts. This research provides scientific insights into the antioxidant and the antimicrobial potency of anise. Further phytochemical and pharmacological studies are needed to determine the specific component(s) responsible for the biological activity and address the safety and toxicity issues.

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