Original Article

Phytochemical Analysis and Assessment of Antimicrobial Properties of Selected UAE Desert Plants and Specific Remedies Against Resistant Microorganisms

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ABSTRACT

Antimicrobial resistance is a significant global health challenge, driven in part by the overprescription of antimicrobial agents and their improper use by patients. This study aimed to compare the antimicrobial properties of five desert plants—*Indigofera intricata, Plantago psyllium, Tribulus arabicus, Fagonia indica Burm.f., and Zygophyllum shewinf*—alongside various types of honey, olive oil, and colostrum against select Gram-positive and Gram-negative bacteria and fungi. The objective was to associate their antimicrobial activity with their phenolic and triterpenoid profiles. Each plant material was extracted using solvents of different polarities, and thin-layer chromatography was employed for phytochemical analysis. The total phenolic and flavonoid contents of the extracts were also measured, and the purity of the honey samples was evaluated. Antimicrobial activity was assessed using both well diffusion and dilution methods. The findings revealed that the plant extracts, different honey varieties, olive oil, and colostrum all demonstrated varying levels of antimicrobial efficacy.

Keywords

Antimicrobial resistance (AMR); Desert medicinal plants; *Indigofera intricata*; *Plantago psyllium*; *Tribulus arabicus*; *Fagonia indica* Burm.f.; *Zygophyllum shewinf*; Phenolic compounds; Flavonoids; Triterpenoids; Colostrum milk; Honey; Olive oil; Phytochemical screening; Thin-layer chromatography (TLC); Antibacterial activity; Antifungal activity; Multidrug-resistant bacteria; MRSA; Carbapenem-resistant Enterobacteriaceae (CRE)

INTRODUCTION

Pathogenic microorganisms—such as bacteria, fungi, parasites, and viruses—are key contributors to infectious diseases transmitted among humans[1]. Antimicrobial resistance (AMR) refers to the ability of these microorganisms (e.g., bacteria, certain parasites, and viruses) to resist the action of antimicrobial agents like antibiotics, antimalarials, and antivirals. As a result, standard treatments become less effective, and infections persist and may continue to spread [2].

Human activities can exacerbate the spread of AMR, for instance, through the overprescription of antimicrobials by healthcare providers, improper use of these agents by patients, and insufficient infection prevention and control measures.

Medicinal plants, especially those long utilized in traditional remedies, are recognized as valuable sources of novel, therapeutically beneficial compounds. These plants play a crucial role in combating bacterial infections. Among these compounds, phenolic substances (a class of secondary metabolites) are abundantly found in plants and are known to possess antimicrobial properties[3]. Phenolics also exhibit a wide range of

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biological activities such as anti-inflammatory, anti-ulcer [4], anti-diabetic[5], antitumor, and antioxidant effects [6,7].

The genus *Indigofera* (*family Fabaceae*) is extensively distributed in the UAE, Nigeria, India, and tropical regions. It is traditionally employed to treat dysentery, constipation, malaria, and stomach ache. Chemical constituents include flavonoids, saponins, and quinones[8]. *Indigofera intricata,* which grows in the UAE, Iran, and tropical areas, has shown strong cytotoxic activity, with methanolic extracts displaying moderate cytotoxic effects (LC50 values of 30–50 µg/mL) against brine shrimp [9].

Another study investigating *I. intricata* formulated in chitosan nanoparticles reported that the alcoholic extract contained high levels of flavonoids (22%) and phenolics (15%). Thin-layer chromatography (TLC) and Fourier-transform infrared spectroscopy (FTIR) confirmed the interaction between chitosan and the polyphenolic compounds. The highest dose of the plant extract (250 mg) exhibited the lowest cell viability of 45.21% ± 4.8%[10]

Plantago psyllium (ispaghula), belonging to the family Plantaginaceae, is found in the UAE and Northern Africa. It is used as a laxative to treat hemorrhoids, irritable bowel syndrome, high cholesterol, and diabetes. Its seeds and husks are rich in fiber, which promotes bowel movements, and contain polyphenols and flavonoids such as rutin [11].

The genus *Tribulus* (family Zygophyllaceae), also called Sharshar, is found in the warmer regions of Asia, Africa, and Australia. Folk medicine uses it as an analgesic and diuretic. Its constituents include saponins, alkaloids, and tannins[12]. *Tribulus arabicus* is a perennial herb with large yellow flowers and grey-green leaves, known for notable antioxidant activity; however, information on its broader biological properties and chemical constituents is limited[13].

Fagonia indica Burm.f. (family Zygophyllaceae), referred to as Shekaa, is distributed across Asia, Egypt, and warm regions of every continent except Australia. Traditionally, it is used to alleviate pain and severe itching. Its alcoholic extract has demonstrated antimicrobial and antitumor activities [14], and it also contains saponins, alkaloids, and tannins[15].

Zygophyllum shewinf (family Zygophyllaceae) thrives in the Arabian Gulf, especially in areas with high salt accumulation[16]. It is known for its anthelmintic and antidiabetic properties[17]. However, reports detailing the biological activities of *Zygophyllum hamiense* Schweinf. and its active components are scarce.

Accordingly, this study aimed to perform a comparative investigation of selected desert plants—*Indigofera intricata*, *Plantago psyllium*, *Tribulus arabicus*, *Fagonia indica* Burm.f., and *Zygophyllum shewinf*—alongside different types of honey, olive oil, and colostrum. The goal was to assess their efficacy against specific resistant bacteria and fungi, and to relate their antimicrobial capabilities to their phenolic and triterpenoid profiles.

METHODS

Chemicals and Drugs

Methanol, ethanol, hexane, and chloroform were sourced from Fisher Scientifics (UK) and Scharlam. Quercetin, rutin, α-amyrin, linoleic acid, oleic acid, culture media, and DMSO were procured from Sigma Chemical (St. Louis, Missouri, USA). Folin-Ciocalteu reagent and thin-layer chromatography (TLC) plates were obtained from Merck (Darmstadt, Germany).

Plant Material

Whole plants of *Fagonia indica*, *Plantago psyllium*, *Zygophyllum hamiense* Schweinf., *Indigofera intricata*, and *Tribulus arabica* were collected from the Muhaisnah desert, Dubai, UAE, in September 2018. Various honey types were obtained from Al-Mansoura (Egypt), Sharjah (UAE), and Sanaa (Yemen), while olive oil was sourced from Nablus (Palestine). Colostrum milk was purchased from Kordel's Advanced Nutrition. The plants were identified and authenticated by Prof. Naglaa Gamil Shehab, Department of Pharmaceutical Chemistry and Natural Products, Dubai Pharmacy College for Girls, Dubai, UAE. The plant samples were shade-dried, ground into a fine powder, and stored for further analysis.

Preparation of Plant Extracts

Each powdered plant sample (100 g) was extracted separately using cold maceration in methanol, ethanol, *n*-hexane, and chloroform (1 L × 2). The solvents were evaporated under reduced pressure at 50 °C. The dried extracts were preserved for biological evaluation and phytochemical analysis. The residual weights of the extracts are presented in Table 1.

Phytochemical Investigation

TLC analysis was conducted on all plant extracts using different solvent systems: chloroform:methanol (9.8:0.2, 9.5:0.5, 8.5:1.5) and benzene:ethyl acetate (8.4:1.6). Spots were visualized under a UV lamp (365 nm) and sprayed with *p*-anisaldehyde. Retention factor (Rf) values were recorded and compared with known standards, including quercetin, rutin, α-amyrin, linoleic acid, and oleic acid. Additionally, the extracts were screened for anthraquinones, carbohydrates or glycosides, tannins, and flavonoids.

Purity Testing of Honey

All honey samples were subjected to chemical tests to assess their purity [18].

Colorimetric Determination of Total Phenolic and Flavonoid Contents

Total phenolic and flavonoid contents were quantified spectrophotometrically (UV-1700 Pharma Spec, Shimadzu, Japan). Powdered plant samples (10 g each) were macerated in methanol and ethanol. Total phenolic content was measured using the Folin-Ciocalteu reagent, following the method described by Singleton and Rossi [19] and modified by Oktay et al. [20]. The results were expressed as mg/g gallic acid equivalent, calculated based on the dry weight of the plant material.

A standard curve was prepared using gallic acid solutions (10, 20, 30, 40, and 50 µg/mL). Each sample or standard (1 mL) was mixed with 9 mL of water in a volumetric flask, followed by the addition of 1 mL Folin-Ciocalteu reagent. The mixture was vortexed, and after 5 minutes, 10 mL of 7% sodium carbonate was added. The solution was incubated at room temperature for 90 minutes, and absorbance was measured at 750 nm against a reagent blank. All measurements were performed in triplicate.

Total flavonoid content was determined using the aluminum chloride colorimetric method, with quercetin as the standard [21]. A 0.1 mL aliquot of each extract was mixed with 0.3 mL distilled water, followed by the addition of 0.03 mL of 5% $NaNO_2$ solution. After 5 minutes at room temperature, 0.03 mL of 10% aluminum chloride was added, and the mixture was incubated for another 5 minutes. Then, 0.2 mL of 1 mM NaOH was added, and the volume was adjusted to 1 mL with distilled water. The absorbance was measured at 510 nm. All experiments were performed in triplicate.

Antimicrobial Study

Antimicrobial activity was assessed following Clinical and Laboratory Standards Institute (CLSI) guidelines [22].

Microorganisms

Eight microbial strains were provided by Rashid Hospital for antimicrobial screening. These included:

- **Gram-positive bacteria**: MRSA (*Methicillin-resistant Staphylococcus aureus*), *Streptococcus pyogenes*
- **Gram-negative bacteria**: *E. coli*, *Pseudomonas aeruginosa*, *Proteus mirabilis*, Carbapenem-resistant *Enterobacteriaceae*, *Klebsiella pneumoniae*
- **Fungi**: *Candida albicans*

Microorganisms were cultured on appropriate media: nutrient agar for *S. aureus*, *P. aeruginosa*, *P. mirabilis*, and *S. pyogenes*; MacConkey agar for *E. coli* and *K. pneumoniae*; and Sabouraud dextrose agar for *C. albicans*.

Preparation of McFarland Standard

A McFarland standard solution was prepared by mixing 0.05 mL of a 1% w/v barium chloride solution with 9.95 mL of a 1% v/v sulfuric acid solution under continuous stirring. The standard was stored in sterile, airtight screw-cap tubes at room temperature, protected from sunlight [23].

Media Preparation

- **Nutrient broth** was prepared following the manufacturer's instructions (23 g in 1 L distilled water) and autoclaved at 121°C for 15 minutes at 15 lb pressure [23].
- **Mac Conkey agar** (52 g in 1 L distilled water) was prepared, autoclaved, and used for culturing *P. mirabilis*, *P. aeruginosa*, *E. coli*, *Enterobacteriaceae*, and *K. pneumoniae* [23].
- **Nutrient agar** (28 g in 1 L distilled water) was prepared and used for culturing *S. pyogenes* and MRSA and for antibiotic susceptibility testing [23].
- **Sabouraud dextrose agar** (65 g in 1 L distilled water) was prepared and used for *C. albicans* culture [23].

Inoculum Preparation

A broth suspension was prepared from colonies isolated

from 24-hour incubated agar plates. The turbidity was adjusted to match a 0.5 McFarland standard, ensuring a bacterial population of approximately 5 × 10^5 CFU/mL within 15 minutes of preparation [24].

Antimicrobial Activity Assessment

Diffusion Method

Extracts (2.5 mg) were tested for antimicrobial activity using the well diffusion method, with DMSO as the solvent. Bacterial inoculum (~5 × 10^5 CFU/mL) was spread on large nutrient agar Petri plates (150 mm). Wells (7 mm diameter) were made, and 50 µL of the extract (2.5 mg dose) was introduced. Plates were incubated at 37ºC for 24 hours. Zones of inhibition were measured using a Kirby-Bauer standard scale and compared with amoxicillin (10 mcg) and ceftazidime/clavulanic acid (30/10 mcg) as controls. Results were recorded as susceptible, intermediate, or resistant.

Dilution Method

The broth dilution method was used to determine the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC), following CLSI guidelines. Bacterial culture (0.5 mL) and nutrient broth (4 mL) were mixed with 0.5 mL of varying extract concentrations and incubated at 37°C for 24 hours. MIC was recorded as the lowest concentration inhibiting visible growth. Samples from MIC wells were sub-cultured on nutrient agar plates to determine MBC, defined as the lowest concentration preventing 99.9% bacterial growth.

RESULTS

Phytochemical Investigation

Residual Weight of Plant Extracts

The residual weights of the extracted plant materials are presented in **Table 1**.

Table 1. The residual weight of the plants

Solvent Name / Plant Name	Ethanol	Methanol	Hexane	Chloroform
F.indica	2.989g	16.064g	0.237g	0.586g
Z.shewinf	3.142g	5.778g	0.318g	1.06g
T.arabica	7.673g	4.327g	0.258g	0.952g
P. psyllium	3.818g	4.702g	0.888g	0.646g
I. intricata	11.479g	10.832g	0.582g	1.276g

3.1.2. Chemical Analysis

Anthraquinone was exclusively detected in the methanolic extract of *I. intricata* (**Table 2**). In contrast, tannins were present in all plant extracts, except for the hexane and chloroform extracts of *Z. shewinf* and the hexane extract of *T. arabica* (**Table 2**). Flavonoid glycosides were identified in the methanolic and ethanolic extracts of all investigated plants (**Table 2**). Additionally, all extracts of *F. indica* contained saponin glycosides, while saponin glycosides were also found in the ethanolic extracts of *Z. shewinf*, *I. intricata*, and *T. arabica* (**Table 2**).

Table 2. Phytochemical screening of different extracts

Extract	Saponin test	Tannin test	Flavonoids test	Anthraquinone Test
F. indica Hexane	+	+	-	-
F. indica Chloroform	+	+	+	-
F. indica Ethanol	+	+	+	-
F. indica Methanol	+	+	+	-
Z.shewinf Hexane	-	-	-	-
Z.shewinf Chloroform	-	-	+	-
Z.shewinf Ethanol	+	+	+	-
Z.shewinf Methanol	-	+	+	-
P. psyllium Hexane	+	+	-	-
P. psyllium Chloroform	-	+	-	-
P. psyllium Ethanol	-	+	+	-
P. psyllium Methanol	-	+	+	-
T.arabica Hexane	-	-	-	-
T.arabica Chloroform	-	+	+	-

Extract	Saponin test	Tannin test	Flavonoids test	Anthraquinone Test
T.arabica Ethanol	+	+	+	-
T.arabica Methanol	+	+	+	-
I. intricata Hexane	+	+	-	-
I. intricata Chloroform	-	+	+	-
I. intricata Ethanol	+	+	+	-
I. intricata Methanol	-	+	+	+

3.1.3. TLC Analysis

Thin-layer chromatography (TLC) analysis revealed the presence of α-amyrin and linoleic acid in the hexane and chloroform extracts of all plant samples (**Table 3**). However, oleic acid was detected in the hexane and chloroform extracts of all plants except *I. intricata* and *T. arabica* (**Table 3**). Quercetin was identified exclusively in the ethanolic and methanolic extracts of *I. intricata* and *P. psyllium*, whereas rutin was found only in the ethanolic extract of these two plants (**Table 3**).

3.1.4. Total Phenolic and Flavonoid Content

The methanolic and ethanolic extracts of *I. intricata* and the ethanolic extract of *T. arabica* exhibited the highest flavonoid content (**Table 4**). Similarly, the highest total phenolic content was observed in the methanolic and ethanolic extracts of both *P. psyllium* and *I. intricata* (**Table 4**).

Table 4. Total phenolic and Flavonoids' contents

Plants' name	Flavonoids %		phenolic %	
	Methanol	Ethanol	Methanol	Ethanol
F.indica	1.312	1.12	0.824	1.05
Z.shewinf	0.512	0.832	0.656	0.584
T.arabica	0.992	1.968	0.864	1.088
P. psyllium	0.672	1.104	1.36	1.44
I. intricate	1.92	2.24	1.44	1.44

3.2. Purity of Honey

All tested honey samples were confirmed to be pure.

Table 3. TLC investigation of different extracts

	α-Amyrin				Rutin				Quercetin				Linoleic acid				Oleic acid			
Extracts' Name	Meth.	Eth.	Ch.	H.	Meth.	Eth.	Ch.	H.	Meth.	Eth.	Ch.	H.	Meth.	Eth.	Ch.	H.	Meth.	Eth.	Ch.	H.
F.indica	-	-	+	+	-	-	-	-	-	-	-	-	-	-	+	+	-	-	+	+
Z.shewinf	-	-	+	+	-	-	-	-	-	-	-	-	-	-	+	+	-	-	+	+
T.arabica	-	-	+	+	-	-	-	-	-	-	-	-	-	-	+	+	-	-	-	-
P. psyllium	-	-	+	+	+	+	-	-	+	+	-	-	-	-	+	+	-	-	+	+
I. intricate	-	-	+	+	-	+	-	-	+	+	-	-	-	-	+	+	-	-	-	-

3.3. Antimicrobial Activity

All plant extracts, various honey samples, colostrum milk, and olive oil demonstrated antibacterial activity against *Pseudomonas aeruginosa* (**Table 5**).

- Colostrum milk, all honey types, and the methanolic extract of *I. intricata* exhibited the strongest antibacterial activity against *Klebsiella pneumoniae*. Other plant extracts displayed moderate activity, while olive oil showed no antibacterial effect (**Table 5**).
- The highest antibacterial activity against *Proteus mirabilis* was observed in the chloroform extract of *F. indica*, the ethanolic extract of *T. arabica*, and the hexane extracts of *Indigofera* and *P. psyllium* (**Table 5**).
- The methanolic extracts of *F. indica* and *I. intricata*, along with all honey types, showed the highest antibacterial activity against *Escherichia coli* (**Table 5**).

Table 5: Antibiotic diffusion method for gram negative microorganisms. **(R indicates Resistance)**

Gram Negative Microorganisms								
Plant extract	Zone of inhibition (mm) against *Pseudomonas aeruginosa* (mean ± SD)		Zone of inhibition (mm) against *Klebsiella* (mean ± SD)		Zone of inhibition (mm) against *E-coli* (mean ± SD)		Zone of inhibition (mm) against *Proteus* (mean ± SD)	
	18 hrs	24 hrs	18 hrs	24 hrs	18 hrs	24 hrs	18 hrs	24 hrs
F. indica Hexane	29.8± 0.21	30± 0.30	17.8±0.20	18±0.1	17.8±0.03	18±0.071	R	R
F. indica Chloroform	29.8± 0.24	30.0	16.8±0.21	17±50	R	R	34.7±0.05	35±0.01
F. indica Ethanol	28.8± 0.15	29± 0.21	19.8±0.61	20±0.12	R	R	R	R
F. indica Methanol	19.8± 0.01	20± 0.222	17.8±0.90	18±0.21	34.8±0.09	35±0.08	R	R
Z.shewinf Hexane	R	R	12.8±0.27	13±0.66	R	R	R	R
Z.shewinf Chloroform	R	R	14.8±0.65	15± 0.71	R	R	R	R
Z.shewinf Ethanol	19.8± 0.02	20± 0.24	14.8±0.42	15±0.68	R	R	R	R
Z.shewinf Methanol	24.8± 0.5	25.0	15.8±0.28	16± 0.54	R	R	R	R
P. psyllium Hexane	17.8± 0.24	18± 0.24	12.8±0.81	13±0.05	R	R	21.7±0.18	22±0.11
P. psyllium Chloroform	R	R	12.8±0.70	13±0.07	12.8±0.51	13±0.12	R	R
P. psyllium Ethanol	17.8± 0.41	18± 0.60	14.8±0.30	15± 0.70	R	R	R	R
P. psyllium Methanol	24.8± 0.36	25± 0.85	14.8±0.23	15± 0.02	R	R	R	R
T.arabica Hexane	17.8± 0.24	18± 0.22	12.8±0.85	13±0.71	R	R	R	R
T.arabica Chloroform	R	R	R	R	R	R	R	R
T.arabica Ethanol	R	R	R	R	R	R	29.7±0.22	30±0.71
T.arabica Methanol	12.8± 0.18	13± 0.59	R	R	R	R	21.7±0.20	22±0.87
I. intricata Hexane	19.8± 0.69	20± 0.10	R	R	R	R	24.7±0.72	25±0.21
I. intricata Chloroform	29.8± 0.52	30± 0.22	12.8±0.31	130.±06	R	R	R	R
I. intricata Ethanol	22.8± 0.11	23± 0.52	R	R	R	R	R	R

Gram Negative Microorganisms								
Plant extract	Zone of inhibition (mm) against *Pseudomonas aeruginosa* (mean ± SD)		Zone of inhibition (mm) against *Klebsiella* (mean ± SD)		Zone of inhibition (mm) against *E-coli* (mean ± SD)		Zone of inhibition (mm) against *Proteus* (mean ± SD)	
	18 hrs	24 hrs	18 hrs	24 hrs	18 hrs	24 hrs	18 hrs	24 hrs
I. intricata Methanol	29.8± 0.24	30± 0.67	24.8±0.61	25±0.30	29.8±0.23	30±0.12	18.7±0.21	19±0.81
Colostrum Milk	27.8± 0.33	28± 0.49	29.8±0.72	30±0.15	24.8±0.14	25±0.23	R	R
UAE Sidr Honey	14.8± 0.70	15± 0.27	24.8±0.31	25±0.11	29.8±0.32	30±0.21	R	R
Yemeni Honey	19.8± 0.12	20± 0.26	24.8±0.27	25±0.21	31.8±0.15	32±0.02	R	R
Egyptian Honey	14.8± 0.09	15± 0.56	17.8±11	18±0.37	29.8±0.78	30±0.90	R	R
Olive oil	39.8± 0.6	40± 0.71	0.0	0.1±0.02	R	R	R	R
Ceftazidime	22.0	22.0	22.0	22.0	14.8±0.21	15±0.21	16.7±0.02	17±0.61

- Strong antibacterial activity against *Streptococcus pyogenes* was detected in the methanolic and ethanolic extracts of *F. indica*, the methanolic extract of *Z. shewinf*, the ethanolic extract of *T. arabica*, and both the methanolic and ethanolic extracts of *I. intricata* (**Table 6**).
- Colostrum milk exhibited the strongest antifungal activity against *Candida albicans* (**Table 6**).

Table 6: Antibiotic diffusion method for gram positive microorganisms. **(R indicates Resistance)**

Gram Positive Microorganisms				
Plant extract	Zone of inhibition (mm) againts *Streptococcus* (mean ± SD)		Zone of inhibition (mm) againts *Candida Albicans* (mean ± SD)	
	18 hrs	24 hrs	18 hrs	24 hrs
F. indica Hexane	R	R	R	R
F. indica Chloroform	R	R	R	R
F. indica Ethanol	21.6±0.12	22±0.32	R	R
F. indica Methanol	19.6±0.52	20±0.22	R	R
Z.shewinf Hexane	R	R	15.8±0.51	16±0.25
Z.shewinf Chloroform	R	R	16.8±0.11	17±0.51
Z.shewinf Ethanol	R	R	14.8±0.31	15±0.87
Z.shewinf Methanol	19.6±0.01	20±0.5	13.8±0.22	14±0.72
P. psyllium Hexane	14.6±0.11	15±0.10	R	R
P. psyllium Chloroform	R	R	R	R
P. psyllium Ethanol	17.6±0.33	18±0.15	R	R
P. psyllium Methanol	15.6±0.51	16±0.21	R	R

Gram Positive Microorganisms				
Plant extract	Zone of inhibition (mm) againts *Streptococcus* (mean ± SD)		Zone of inhibition (mm) againts *Candida Albicans* (mean ± SD)	
	18 hrs	24 hrs	18 hrs	24 hrs
T.arabica Hexane	19.6±0.02	20±0.10	R	R
T.arabica Chloroform	19.6±0.21	20±0.21	R	R
T.arabica Ethanol	23.6±0.25	24±0.11	R	R
T.arabica Methanol	20.6±0.21	21±0.72	R	R
I. intricata Hexane	15.6±0.28	16±0.32	R	R
I. intricata Chloroform	19.6±0.12	20±0.33	R	R
I. intricata Ethanol	22.60±0.08	23.0	R	R
I. intricata Methanol	23.6±0.73	24±0.52	R	R
Colostrum Milk	R	R	44.8±0.21	45±0.22
UAE Sidr Honey	R	R	14.8±0.52	15±0.34
Yemeni Honey	R	R	12.8±0.64	13±0.44
Egyptian Honey	R	R	14.8±0.24	15±0.22
Olive oil	R	R	19.8±0.97	20±0.71
Ceftazidime	9.6±0.89	10±0.21	R	R

- The highest antibacterial activity against carbapenem-resistant *Enterobacteriaceae* was found in the ethanolic extract of *F. indica*, colostrum milk, and Yemeni honey (**Table 7**).
- The strongest antibacterial effects against MRSA were recorded for the methanolic extract of *I. intricata*, UAE Yemeni honey, and the hexane and methanolic extracts of *F. indica* (**Table 7**).

Table 7: Antibiotic diffusion method for antibiotic resistant microorganisms.

Antibiotic Resistant Microorganisms				
Plant extract	Zone of inhibition (mm) against *Methicillin Resistant Staphylococcus Aureus (MRSA)* (mean ± SD)		Zone of inhibition (mm) against Carbapenem *Resistant Enterobacteriaceae* (mean ± SD)	
	18 hrs	24 hrs	18 hrs	24 hrs
F. indica Hexane	19.8±0.22	20±0.80	18.8±0.21	19±0.12
F. indica Chloroform	14.8±0.51	15±0.31	R	R
F. indica Ethanol	12.8±0.16	13±0.87	29.8±0.07	30±0.09
F. indica Methanol	18.8±0.61	19±0.22	19.8±0.38	20±0.29
Z.shewinf Hexane	R	R	R	R
Z.shewinf Chloroform	R	R	R	R
Z.shewinf Ethanol	R	R	R	R

Antibiotic Resistant Microorganisms				
Plant extract	Zone of inhibition (mm) against *Methicillin Resistant Staphylococcus Aureus (MRSA)* (mean ± SD)		Zone of inhibition (mm) against Carbapenem *Resistant Enterobacteriaceae* (mean ± SD)	
	18 hrs	24 hrs	18 hrs	24 hrs
Z.shewinf Methanol	R	R	R	R
P. psyllium Hexane	R	R	R	R
P. psyllium Chloroform	R	R	R	R
P. psyllium Ethanol	R	R	16.8±0.51	17±0.21
P. psyllium Methanol	R	R	R	R
T.arabica Hexane	R	R	R	R
T.arabica Chloroform	R	R	R	R
T.arabica Ethanol	R	R	R	R
T.arabica Methanol	R	R	R	R
I. intricata Hexane	R	R	R	R
I. intricata Chloroform	R	R	R	R
I. intricata Ethanol	R	R	R	R
I. intricata Methanol	29.8±0.60	30±0.33	R	R
Colostrum Milk	29.8±0.76	30±0.01	29.8±0.67	30±0.43
UAE Sidr Honey	8.8±0.45	9±0.70	19.8±0.52	20±0.23
Yemeni Honey	19.8±0.40	20±0.72	22.8±0.78	23±0.50
Egyptian Honey	13.8±0.04	14±0.27	R	R
Olive oil	9.8±0.12	10±0.20	R	R
Ceftazidime	R	R	10.8±0.37	11±0.19

- The minimum inhibitory concentration (MIC) for MRSA was determined to be **1000 ± 0.82 µg/mL** for the methanolic extract of *I. intricata* (**Table 8**).

Table 8: Antibiotic dilution method for MRSA.

Plant extract	*Methicillin Resistant Staphylococcus Aureus (MRSA)* (mean ± SD) (ug/ml)	
	MIC	MBC
F. indica Hexane	1500 ±0.87	2000 ±0.57
I. intricata Methanol	1000 ± 0.82	1500 ±0.78
Yemeni Honey	2500 ±0.29	3000 ±0.42
Vancomycin	2.0	8.0

DISCUSSION

Antimicrobial resistance has become a major global health concern, as various microorganisms have developed resistance to antimicrobial drugs. This resistance often results from the overuse of antimicrobial agents by healthcare providers or their misuse by patients. To mitigate the emergence and spread of resistant strains, it is essential to ensure the appropriate use of antimicrobial medications [1].

In this study, the antimicrobial properties of hexane, chloroform, ethanol, and methanol extracts from five plant species were evaluated against a range of bacterial and fungal strains. The plant extracts exhibited

varying levels of antimicrobial activity, which can be attributed to their diverse phytochemical compositions, particularly flavonoids [24] (notably rutin and quercetin), tannins [25], and triterpenes such as α-amyrin, oleic acid, and linoleic acid [26,27]. The synergistic effect of these compounds likely contributes to the observed antimicrobial activities.

Although the chloroform extract of *P. psyllium* contained tannins, α-amyrin, oleic acid, and linoleic acid, it did not exhibit antibacterial activity, likely due to the absence of flavonoids (**Table 2**). Conversely, the methanolic extract of *I. intricata* demonstrated greater inhibition against *Streptococcus* compared to its ethanolic counterpart, which may be attributed to its anthraquinone content. The hexane extract, which lacked flavonoids and anthraquinones, showed only mild antibacterial activity against *Streptococcus* (**Table 2** and **Table 6**).

The methanolic extract of *I. intricata* also exhibited significant antibacterial activity against MRSA, likely due to its high flavonoid content compared to other plants (**Table 2** and **Table 7**) [28]. This was further confirmed through dilution assays, where *I. intricata* demonstrated the lowest MIC and MBC values (**Table 8**).

Similarly, the methanolic extract of *F. indica* showed strong activity against *E. coli*, which can be attributed to its high flavonoid content (**Table 2** and **Table 5**).

The absence of rutin, quercetin, and anthraquinones in all extracts of *Z. shewinf* and *T. arabica* likely explains their lack of antibacterial activity (**Table 3**). However, the hexane extract of *Z. shewinf* exhibited antibacterial activity, possibly due to the presence of α-amyrin, linoleic acid, and oleic acid (**Table 3**).

The chloroform extract of *P. psyllium* displayed strong antibacterial activity against *E. coli*, which can be attributed to its tannin content (**Table 2** and **Table 5**).

The inhibition of carbapenem-resistant *Enterobacteriaceae* by the hexane extract of *F. indica* is likely due to the presence of triterpenoids (**Table 2** and **Table 7**). Additionally, the chloroform extract of *F. indica* demonstrated high activity against *Proteus mirabilis*, which may be due to its flavonoid and tannin content (**Table 2** and **Table 5**).

Among the tested natural agents, colostrum milk proved to be the most effective antifungal and antibacterial agent, exhibiting strong activity against most bacterial strains except *Proteus mirabilis*. The effectiveness of colostrum is likely attributed to its high antibody content (**Table 5-7**) [29].

Palestinian olive oil exhibited antimicrobial activity against *Pseudomonas aeruginosa* and *Candida albicans* but showed no effect against other tested microorganisms. This lack of activity may be attributed to bacterial resistance (**Table 5-7**). Olive oil is known for its high oleic acid content and richness in phenolic compounds, which contribute to its antimicrobial properties [28].

The variation in the antimicrobial effectiveness of different honey types is influenced by seasonal, botanical, and geographical factors. Differences in pH, osmotic pressure, and microbial susceptibility to hydrogen peroxide also play a role in inhibiting bacterial growth, which is a key antimicrobial mechanism of honey [29].

UAE Sidr honey and Yemeni honey demonstrated effectiveness against all tested microorganisms except *Streptococcus pyogenes* and *Proteus mirabilis*. However, Yemeni honey was more effective against carbapenem-resistant *Enterobacteriaceae* (CRE) than against MRSA (**Table 5-7**).

Colostrum milk, honey, and olive oil are widely recognized as immune system boosters and are primarily used for infection prevention rather than as primary therapeutic agents.

CONCLUSION

The present study underscores the growing importance of exploring natural products as alternative or complementary agents to combat antimicrobial resistance (AMR), a global health threat exacerbated by the misuse and overuse of conventional antimicrobials. Through comparative evaluation, five desert plants—*Indigofera intricata*, *Plantago psyllium*, *Tribulus arabicus*, *Fagonia indica Burm.f.,* and *Zygophyllum shewinf*—as well as various types of honey, olive oil, and colostrum, demonstrated variable but significant antimicrobial activity against a spectrum of Gram-positive and Gram-negative bacteria, including resistant strains such as MRSA and carbapenem-resistant Enterobacteriaceae, as well as fungi like *Candida albicans*.

The methanolic and ethanolic extracts of *I. intricata* and *F. indica* showed the most promising antibacterial effects, particularly against MRSA and *E. coli*, which

correlates with their high flavonoid and phenolic content. The antimicrobial efficacy of these extracts was supported by phytochemical screening and TLC analysis, revealing the presence of key bioactive compounds such as quercetin, rutin, α-amyrin, and linoleic acid. Among non-plant natural products, colostrum milk emerged as a potent antimicrobial agent, especially against *Klebsiella pneumoniae* and *C. albicans*, likely due to its rich immunoglobulin content.

These findings suggest that the antimicrobial activity of natural substances is strongly influenced by their phytochemical profiles, especially the presence of flavonoids and triterpenoids. The study highlights the potential of desert plants and other natural products as viable candidates for developing novel antimicrobial therapies, which could serve as a sustainable approach to address the escalating challenge of drug-resistant infections. Further in vivo studies and compound isolation are recommended to validate their therapeutic applications and elucidate mechanisms of action.

AUTHOR CONTRIBUTIONS

We confirm that Prof. Naglaa G. Shehab conceptualized and supervised the study. Prof. Naglaa G. Shehab, Ola Alkhalil, Aya Alkhirat, Majd Almohammed, Noor Alatwan, Dr. Gazala A. Khan, and Kholoud Y. Abushawish contributed to data collection, analysis, and interpretation. Prof. Naglaa G. Shehab, Dr. Gazala A. Khan, and Kholoud Y. Abushawish were involved in statistical analysis, manuscript drafting, and critical revision.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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