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Research Article:

Exploring the Antimicrobial Activity of Quince Seeds Extracted by Two Different Methods

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Abstract

Background: The rise of resistance to numerous effective antimicrobic drugs and the necessity to investigate alternative agents to address this emerge are conspicuously underscored. **Aim:** The aim is to evaluate the TPC (total phenolic contents), TFC (total flavonoidal contents), and the antimicrobial activity of two quince seeds extracts and their combination with commercial antimicrobial drugs. **Methods:** In this work two ethanolic extracts (Qu-1 and Qu-2) were prepared from quince seeds using two methods, microwave (M.wv) and sequential microwave-sonication assisted extraction (MSAE). The quantification of total phenolic content TFC and total flavonoid content TFC in these extracts were conducted using Folin-Ciocalteu and $AlCl_3$ method respectively. The extracts, along with its combination with conventional antibiotics were tested for antimicrobial activity against a variety of aerobic and anaerobic bacteria using ciprofloxacin and metronidazole as references, respectively. The extracts were also tested against fungi (*C. albicans* and *A. niger*) using the standard, nystatin. **Results:** TPC and TFC for Qu-1 were 2.2578±1.82 µg GAE/g crude solid, 1.4901±1.76 µg RU/g crude solid respectively, while these values for Qu-2 were 1.0341±2.05 µg GAE/g and 0.7342±1.53 µg RU/g. Both extracts showed antimicrobial effectiveness against all examined pathogens with Qu-1 showing superior effectiveness. So, there is a positive association between TPC, TFC and antimicrobial activity. The combination of Qu-1 can enhances both the antifungal and anti-aerobic effects of NYS and CIP by reducing MFC and MBC and their corresponding potency factor. On the contrary, the Qu-2-antibiotic combination does not show any modification. Both extracts have microbicidal activity based on MBC/MIC ratio which is lower than 4. **Conclusion:** Quince seed extracts, especially Qu-1, show promise in fighting microbial infections. They could be used alone or in combination with antibiotics. Further research and development may lead to the development of new antimicrobials to address the growing issue of resistance.

1. Introduction

Several recent international health crises have revealed significant vulnerabilities in global pandemic preparedness regarding infectious diseases, which constitute one of the major contributing factors towards high mortalities worldwide (1,2). The rise of antimicrobial resistance is a paramount health challenge in the 21st century. The enclosure of antimicrobial resistance (AMR) on a global map coincides with the advancements made in the healthcare system's organizational, scientific, and technical capabilities, highlighting the immense socioeconomic vicissitudes of the last century (3). Bacterial antibiotic resistance research has been continuously expanding over the past few decades and is currently one of the microbiological sciences disciplines with the highest rate of growth. The presumption that it is urgent to address a problem with the potential to have catastrophic effects on human health is what is driving research in that direction (4).

The ability to cure common infectious diseases is jeopardized by the regular emergence and global dissemination of new resistance mechanisms. As the effectiveness of antibiotics diminishes, infections like pneumonia, tuberculosis, sepsis, gonorrhea, and foodborne diseases become increasingly challenging, and in some

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cases, even untreatable. Although the acquisition and diffusion of resistance genes require time, the speedy evolution of bacterial resistance is greatly expedited by the unnecessary use and incorrect application of antibiotics (5–7). Resistance to antimicrobial drugs was recently listed as one of the top 10 worldwide public health dangers to humanity by the WHO (3). Since there are fewer novel agents on the horizon and more bacteria are growing resistant to treatment with all known antibiotics, it is vital that new classes of antibiotics be developed to prevent significant global health tragedies (8).

The natural world serves as a major inspiration for the res earch and development of novel drugs. Within the past 4 decades, antibacterial substances with medical applications have been developed that are approximately 75% derived from natural sources (9) .

Plants include a wide range of biomolecules and metabolites that are highly relevant for the identification of novel drugs (10). These natural components are a gold mine in the fight against pathogens resilient to many medications (11,12). In the botanical realm, the collective chemical compounds referred to as phenols encompass various subclasses such as simple phenols, phenolic acids, flavonoids, coumarins, lignans, and other related entities. This expansive group of secondary metabolites, prominently found in plants, stands out as the most extensive in terms of diversity and abundance. These metabolites have demonstrated diverse attributes such as antimicrobial, anti-inflammatory, anticancer, and numerous others. In particular, the antimicrobial efficacy of these plant secondary metabolites can be attributed to the presence of hydroxyl groups attached to the phenol ring. Additionally, the aromatic nature of these compounds may further subsidize their antimicrobic effects (13).

Unlike synthetic antibiotics, which often lead to the development of resistant strains, plant bio-actives offer a multi-component approach, targeting multiple sites in microorganisms and making it difficult for them to develop resistance. Moreover, phytochemicals rich plant extracts often exhibit synergistic ramifications when combined with customary antibiotics, enhancing their efficacy and reducing the required dosage (14,15).

Quince, also known as *Cydonia oblonga* Miller, is a little deciduous tree in the *Rosaceae* family. It bears yellow fruits and their seeds; and its buds, leaves, and bark are also utilized for their healing properties (16). Cydonia oblonga contains a variety of beneficial phytonutrients such as antioxidants, hypoglycemic agents, antimicrobials, anti-inflammatories, anticarcinogens, and anti-ulceratives.

Of particular interest is the use of quince seeds in traditional panacea to address cough, throat dryness, and malignant ulcers. Utilizing quince seed powder in desserts and dairy products has pointedly increased consumer acceptance and improved the nutritional benefits of these products. This is noteworthy considering that quince seeds, which are often discarded, have been recognized as an untapped resource (17).

The mucilage derived from quince seeds demonstrates several healing properties for the skin, by enhancing fibroblast proliferation. It is worth mentioning that the quince fruit contains approximately 10% seeds within its center (18).

Multiple investigations have elucidated the antimicrobial properties of various aerial components of the quince plant, revealing the efficacy of both its leaves and fruits against various pathogenic micro-organisms. Unfortunately, the extent of seed experiments is currently constrained (19). The objective of this study was to assess the antimicrobial efficacy, overall phenolic concentration, and flavonoid content of ethanolic extracts secured from quince seeds, utilizing two distinct green extraction tactics.

2. Materials and methods

The solvents, chemical compounds, and microbial cultures employed in this research were sourced from reputable suppliers, namely Sigma-Aldrich and Tokyo Chemical Industry, known for their reliability and quality. Microbiologics supplied standard strains of fungi and bacteria. The desiccated seeds of quince were obtained from a public marketplace located in Mosul.

In this study, the extraction procedure entailed employing both an ultrasonic bath and a domiciliary microwave oven. Specifically, the ultrasonic bath utilized was the Korean model Power Sonic 410, which operated at a frequency of 40 kHz and possessed a power of 350 W. In conjunction, the microwave extraction method employed the Moulinex MW Steam 23L (MW531070) from France**.**

2.1 Seed processing and extraction

After smashing, the seeds fine powder underwent two distinct extraction approaches: A combination of microwave and ultrasonic assistance in a consecutive manner and extraction enhanced by microwaves.

For microwave-enhanced extraction, the technique proposed by El Kahlout et al. was implemented with certain adaptations. Thirty grams of seeds powder was added to 300 ml ethyl alcohol in a conical flask. The flask was then meticulously positioned at the center of the microwave chamber and mounted on a turning turntable to guarantee consistent heating throughout the experiments. The sample was exposed to microwave irradiation at 180 W for a duration of 90 seconds (20).

The procedure of sequential microwave-sonication extraction followed the protocol established by Gorgani et al. research with simple modification. After microwave irradiation using the same conditions mentioned above, the sample was directly transferred to an ultrasonic bath, where it underwent sonication for a period of half an hour at a temperature of 30 degrees Celsius (21). The consistency was maintained by keeping the process parameters constant throughout two trials.

After the completion of the extraction process, the resultant suspensions (named **Qu-1** and **Qu-2** based on the 2 approaches of extraction mentioned above respectively) underwent filtration. The filtrate was then concentrated utilizing low-pressure techniques. The concentrated liquid was subsequently refrigerated in preparation for forthcoming tests (22).

2.2. Total phenolic content (TPC)

The determination of total phenolic compounds (TPC) in coded ethanol-based extracts was carried out utilizing the colorimetric method devised by Folin-Ciocalteu. To elaborate, a 200 µl volume of a mother solution containing

an aqueous crude solid of 1 mg/ml concentration was appropriately diluted with distilled water (DW) as a diluent, resulting in a final volume of 3 ml. The inquiry was inaugurated by combining this final solution with 0.5 ml of the folin-ciocalteu reagent. The amalgamation procedure persisted for a duration of 3 minutes, after which the resultant mixture was subjected to treatment with a 2 ml aliquot of 20% aqueous sodium carbonate (Na_2CO_3). Following a period of 1 hour of incubation in a light-shielded laboratory setting, the measurable absorbance at a wavelength of 650 nm was recorded. The total phenolic content (TPC) was subsequently determined by extrapolating from the calibration curve constructed using gallic acid, and in order to ensure the accuracy of the findings, three separate and independent prosecutions were undertaken (23).

2.3 Total flavonoid content (TFC)

The TFC of each categorized ethanol-based extract was calculated utilizing the colorimetric $AlCl_3$ technique. In a nutshell, 1 mg/ml aqueous crude solid solution was prepared as the mother solution. Subsequently, a 50 µl portion of the mother solution was sequentially diluted with methanol as the diluent to a final volume of 1 ml, and then further diluted up to 4 ml with distilled water (DW). The experiment commenced by combining the daughter solution with a solution of aqueous $NaNO_2$ (0.3 ml, 5%). Following an incubation period of 5 minutes, an aqueous $AlCl_3$ solution (0.3 ml, 10%) was introduced, and the resultant mixture was stored in a light-restricted area for 6 minutes. An aqueous NaOH solution (2 ml, 1N) was then added to the mixture, and the total volume was adjusted to 10 ml using distilled water as a diluent. Prior to measuring the visible absorbance at 510 nm, the mixture was incubated away from light for 15 minutes. The total flavonoid content (TFC) was determined by utilizing the rutin calibration curve, and to legalize the results, three replicates of the experiment were conducted (24,25).

2.4 Inspecting the antimicrobial attribute

In this particular investigation, the efficacy of antibacterial properties against gram-negative aerobic bacteria was assessed by means of the broth dilution method. Mueller-Hinton broth (MHB) was utilized as a medium to foster bacterial growth, ciprofloxacin (CP) was employed as a (benchmark) for comparison, and (DMSO) (methylsulfinylmethane) was employed as a negative control to determine MIC, MBC and MBC/MIC (PF) values.

In the experiment involving anaerobic bacteria, a specialized growth medium called brucella-agar enriched with sheep blood at a concentration of 5% was utilized. Metronidazole (MNZ) was employed as the benchmark compound in this context. In order to ascertain MIC, MFC and PF for fungal strains, sabouraud-dextrose broth as a conducive growth habitat, nystatin (NY) as a standard were used.

The methodologies employed in this study were adopted from the following published paper: https://www.jmchemsci.com/article_148988.html(6). Notably, a single alteration in the experimental protocol proved critical, namely, the generation of a mother solution (1.5 mg/ml) using autoclaved distilled water.

2.5 Impact of reciprocal application

In order to evaluate the influence of applying different extracts in conjunction with ciprofloxacin (**CIP**), (**MNZ**), or (**NYS**) separately on the targeted microbes, the previously described broth microdilution methodology was employed, which encompassed both quantitative and qualitative assessments. The only alteration made to this procedure was the inclusion of each drug concentration (1 ml) along with its respective concentration of a specified extract (1 ml) (26) with minor modifications.

3. Results and discussion

The widespread utilization of various natural products for their healing properties is facilitated by their relative safety and easy accessibility. This factor contributes to their widespread adoption across diverse communities, enabling individuals to benefit from their therapeutic potential (27). An extensively employed strategy for the identification of innovative lead compounds is the evaluation of bioactivities and biologically active substances within natural products (28).

Numerous investigations have demonstrated that various phytochemicals and distinct phenolic profiles can be found within the seeds of diverse species belonging to the *Rosaceae* family. Motivated by these findings, this current study aim to explore the prospective antimicrobial properties of quince seeds in addition to their contents of phenolics and flavonoids (29). Extraction plays a pivotal role in the phytochemical processing pathway as it serves as a crucial step for the identification and isolation of bioactive compounds from plant materials (30).

A crucial aspect in the standardization of herbal products is the necessity of employing a reliable extraction method. Extensive corroborations suggests that the selection of solvents utilized in the extraction process significantly influences the type and quantity of secondary metabolites retrieved from plants. Accordingly, it is paramount to conscientiously choose both an appropriate extraction solvent and technique to achieve the desired biological activity from these extracts (31). In the present framework, and based on our comprehensive investigation carried out in 2020, the choice of employing ethanol as an extracting agent was made in this scholarly inquiry, owing to its discernible enrichment in phytoconstituent components in comparison to the other five extracts, show casing enhanced antimicrobial efficacy (6).

3.1 Total phenolic contents, TPC

Phenolic compounds can be categorized as secondary metabolites that exhibit a wide spectrum of complexity, encompassing both uncomplicated phenolic molecules and extensively polymerized compounds with different biological activities such as anti-aging, anti-oxidant and antimicrobial (32).

The results in **Table 1** indicated that the utilization of microwave-enhanced extraction (Qu-1) divulged the utmost total phenolic content (2.2578±1.82 µg GAE/g) in contrast to the extracts derived from sequential microwave-sonication extract (Qu-2), (1.0341±2.05 µg GAE/g).

Table 1. TPC and TFC values of quince seeds extracts

Extract code	TPC (µg GAE/g crude solid)±SD (n=3)	TFC (µg RU/g crude solid)±SD (n=3)
Qu-1	2.2578±1.82	1.4901±1.76
Qu-2	1.0341±2.05	0.7342±1.53

GAE and RU: gallic acid and rutin equivalents.

This finding aligns with research conducted by Solaberrieta et al, which demonstrated that the TPC in microwave extracts surpassed that achieved through alternative methodologies (33).

3.2 Total flavonoid content, TFC

Flavonoids epitomize the prevailing category of polyphenolic constituents in the human nourishment and they are omnipresent in plants. These entities portray extraordinary traits as antioxidants, thereby safeguarding plants against deleterious surroundings. Scientists have conducted comprehensive scrutiny of these compounds owing to their inherent potential and projected practical applications. As a result, the methods employed for the retrieval of flavonoids from natural sources have undergone exhaustive investigation, contemplating analytical, preparative, and industrial goals, encompassing the employment of state-of-the-art techniques like microwave and ultrasound-assisted extraction methodologies (34,35).

In this work, TFC for Qu-1 and Qu-2 was estimated to be (1.4901±1.76 µg RU/g) and (0.7342±1.53 µg RU/g) respectively. These results indicate that both extraction methods were successful in extracting flavonoids from the plant material. However, notable differences in TFC values were observed between Qu-1 and Qu-2. The microwave method resulted in a higher TFC value compared to the sequential microwave-ultrasound method. This disparity might be attributed to the different mechanisms involved in each extraction method. So microwave was more efficient and time saving for flavonoid extraction (36).

3.3 Antimicrobial activity

The earnestness for innovative therapeutic agents has reached unprecedented levels due to the advent of drug-resistant diseases caused by morbific microbes, presenting a substantial wide-reaching health predicament. In light of this, extensive exploration across various references has become imperative to identify promising bioactive compounds for the development of groundbreaking treatments. In the hunt of ecologically sustainable substitutes for synthetic chemicals and pharmaceuticals, researchers have extensively studied natural compounds, especially phyto-actives generated by plants, for their up-and-coming antimicrobic properties (37), (38). Within this particular framework, the extract of quince seeds was opted to study microbial suppressive properties.

The results suggest that both Qu-1 and Qu-2 extracts have inhibitory activity against the tested pathogens to varying degrees albeit at a lower level than standard drugs CIP, MNZ and NYS. Ethanol is high polar solvent so, its ability to extract organic and inorganic compounds such as tannins, alkaloids and phenolics with potent antimicrobial action are greater than solvent of low polarity (39). MIC values for Qu-1 ranged between (588 and 632 µg/ml) and for Qu-2 (656 – 744 µg/ml). MBC values for Qu-1 were ranged between (600 and 640 µg/ml), while for Qu-2 were (664 – 760 µg/ml) against aerobic bacteria as in Table 2 and Figure 1. It's obvious that the most sensitive aerobic gram -ve bacteria was *S. typhi*, while *P. aeruginosa* was the most resistant.

According to the illustrated data presented in **Table 3** and graphed in **Figure 2**, it is palpable that the anaerobic strain known as *C. perfringens* exhibited slightly greater resilience towards the Qu-1 and Qu-2 extracts in comparison to the *B. fragilis* strain. This conclusion is supported by the higher MIC and MBC values associated with the former (6).

The safety of the 2 extracts and the standard drug against normal flora *E. coli* BAA-1427 was evaluated. The findings depicted in **Table 2** and **Figure 3** indicate that Qu-1 and Qu-2 exhibit reduced levels of harmful effects compared to CIP based on their higher MIC and MBC values. It is crucial to remember that when tested against disease-causing strains, their safety is relatively reduced.

In relation to the antifungal properties, two disease-causing fungi were employed for inspecting the impact of two extracts compared to the drug nystatin, specifically *C. albicans* ATCC 10231 and *A. niger* ATCC 16888. The outcomes presented in **Table 4** and **Figure 4** demonstrated that both extracts possess a certain level of fungus-restraining capability, albeit inferior to that of NYS. In accordance with MIC and MFC values, both strains were about equally impacted by the 2 extracts.

Table 2. Inhibitory activity of **Qu-1** and **Qu-2** toward aerobic bacteria

Microbmer and its ATCC value	Antimicrobial-related factor	Codes of the reference antimicrobial drug and the extracts used		
		CIP	**Qu-1**	**Qu-2**
Pseudomonas aeruginosa ATCC 27853	MIC	0.75	608	744
	MBC	0.85	620	760
	PF	1.13	1.02	1.02
Klebsiella pneumonia ATCC 700603	MIC	0.40	632	720
	MBC	0.45	640	736
	PF	1.13	1.01	1.02
Haemophilus influenzae ATCC 49247	MIC	0.60	592	684
	MBC	0.65	608	696
	PF	1.08	1.03	1.02
Escherichia coli ATCC 25922	MIC	0.85	612	688
	MBC	0.95	620	692
	PF	1.31	1.01	1.01
Salmonella typhi ATCC 6539	MIC	0.80	588	656
	MBC	0.95	600	664
	PF	1.19	1.02	1.01
Shigella dysenteriae ATCC 13313	MIC	0.55	612	672
	MBC	0.70	616	684
	PF	1.27	1.01	1.02
Escherichia coli BAA-1427	MIC	0.85	448	428
	MBC	0.95	452	440
	PF	1.31	1.01	1.03

The results are expressed in the unite of µg/ml

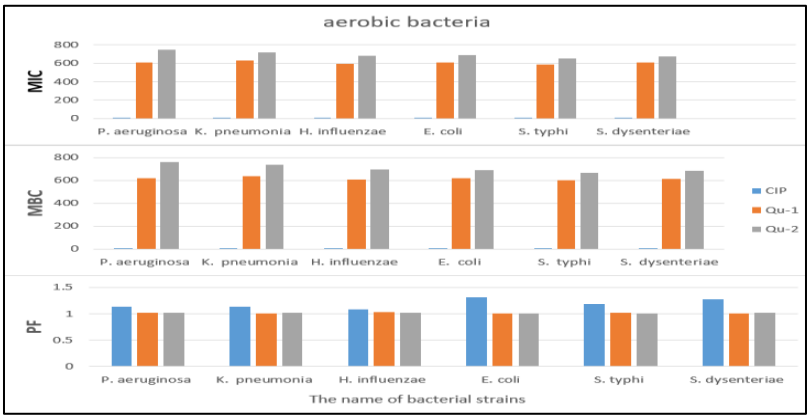

Figure 1. The results of evaluating microbial inhibitory activity against aerobic bacteria

Table 3. Bacterial inhibitory activity of **Qu-1** and **Qu-2** toward anaerobic bacteria

Microbmer and its ATCC value	Antimicrobial-related factor	Codes of the reference antimicrobial drugs and the extracts used		
		MNZ	**Qu-1**	**Qu-2**
Bacteroides fragilis ATCC 25285	MIC	3.0	204	236
	MBC	3.5	220	248
	PF	1.17	1.08	1.05
Clostridium perfringens ATCC 13124	MIC	0.80	216	244
	MBC	0.95	228	252
	PF	1.19	1.06	1.03

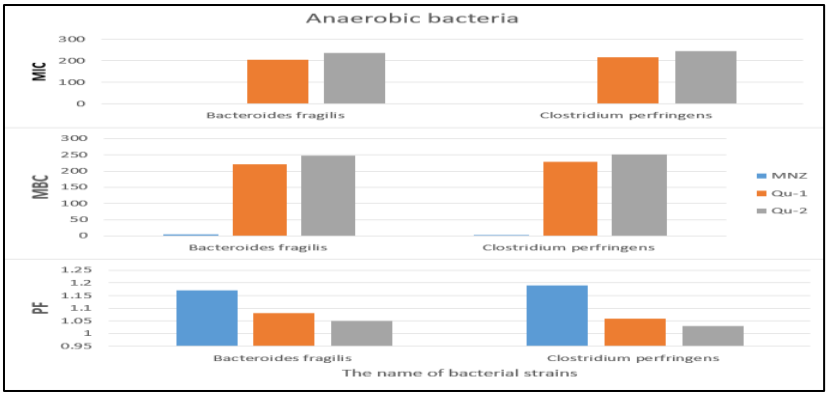

Figure 2. The results of evaluating microbial inhibitory activity of **Qu-1**, **Qu-2** and standard against anaerobic bacteria

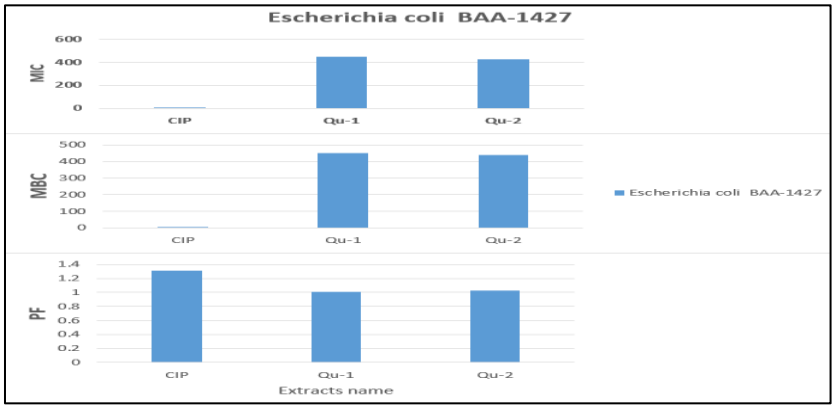

Figure 3. The results of evaluation the two extracts and the standard toward normal bacterial flora

Table 4. Fungal inhibitory activity of the extracts and **NYS**

Fungi and its ATCC value	Antifungal-related factor	Codes of the reference antimicrobial drugs and the extracts used		
		NYS	**Qu-1**	**Qu-2**
Candida albicans ATCC 10231	MIC	4	128	188
	MBC	6	136	200
	PF	1.50	1.06	1.06
Aspergillus niger ATCC 16888	MIC	8	128	192
	MBC	12	140	196
	PF	1.5	1.09	1.02

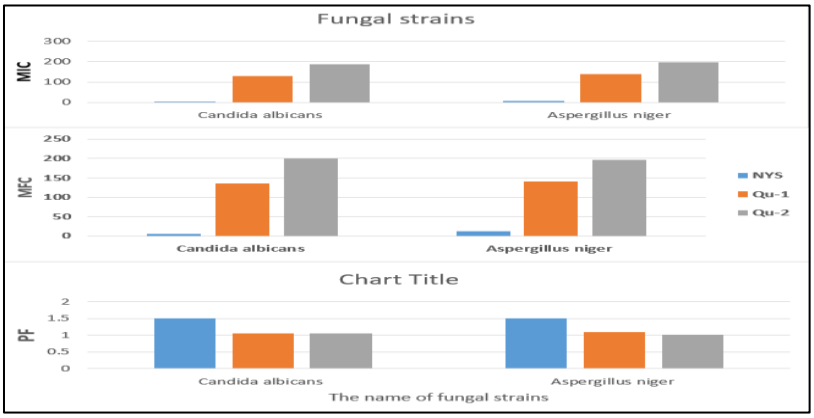

Figure 4. The results of evaluating fungal inhibitory activity of **Qu-1**, **Qu-2** and standard

The efficacy of our seed extracts was additionally substantiated by calculating the ratio (MBC) or (MFC) to (MIC), which enabled the determination of potency factor (PF). Based on a PF value which is below 4, both extracts exhibited bactericidal and fungicidal properties.

Additionally, although both Qu-1 and Qu-2 display antimicrobial inhibitory effects against all tested strains, Qu-1 exhibits more potent activity compared to Qu-2. This enhanced antimicrobial efficacy is directly linked to the higher total flavonoid content (TFC) and total phenolic content (TPC) observed in Qu-1 (40). Flavonoids possess the capability to establish intricate associations with cellular membrane and extracellular proteins (41). Phenolic compounds have the ability to modify the structure of the cell membrane, reducing its lipid content and ultimately hindering microbial growth. These constituents are presumed to inflict harm upon or permeate the lipid structures by saturating the cell membrane (42).

3.4 Reciprocal application

The strategic employment of harmonious combinations of varied antimicrobial substances is perceived as an optimal arsenal in the fight against infections. The utilization of botanical treatments alongside pharmaceutical medications entails specific herb-drug interactions, wherein the potential consequences of these interactions encompass a mutually reinforcing augmentation of antimicrobial efficacy and a concomitant decrease in the unfavorable side effects associated with synthetic drugs. Undoubtedly, the combination of these effects has substantially reduced the likelihood of diminished potency of solitary drug usage in the prolonged treatment of microbial infections (43), (44). Henceforth, a study to evaluate the inhibitory activity of our distinct extracts in conjunction with CIP, MNZ, or NYS against the test microbes was conducted.

Within the domain of aerobic bacterial species, the concurrent utilization of Qu-1 has the potential to magnify the anti-aerobic impact of CIP through the mitigation of minimum bactericidal concentration (MBC) and potency factor (PF) values but not MIC, as in **Table 5** and **Figure 5** and 6. On the other hand, Qu-2 do not have this effect. This variation may be attributed to higher phenolic and flavonoidal contents of Qu-1 extract.

Table 5: The impact of mixing two plant extracts with **CIP** on aerobic bacterial growth.

Microbmer and its ATCC value	Antimicrobial-related factor	Codes of the reference antimicrobial drugs and the extracts used		
		CIP	**CIP + Qu-1**	**CIP + Qu-2**
Pseudomonas aeruginosa ATCC 27853	MIC	0.75	0.75	0.75
	MBC	0.85	0.80	0.85
	PF	1.13	1.07	1.13
Klebsiella pneumonia ATCC 700603	MIC	0.40	0.40	0.40
	MBC	0.45	0.40	0.45
	PF	1.13	1.00	1.13
Haemophilus influenzae ATCC 49247	MIC	0.60	0.60	0.60
	MBC	0.65	0.60	0.65
	PF	1.08	1.00	1.08
Escherichia coli ATCC 25922	MIC	0.85	0.85	0.85
	MBC	0.95	0.85	0.95
	PF	1.31	1.00	1.31
Salmonella typhi ATCC 6539	MIC	0.80	0.80	0.80
	MBC	0.95	0.85	0.95
	PF	1.19	1.06	1.19
Shigella dysenteriae ATCC 13313	MIC	0.55	0.55	0.55
	MBC	0.70	0.60	0.70
	PF	1.27	1.09	1.27
Escherichia coli BAA-1427	MIC	0.85	0.85	0.85
	MBC	0.95	0.85	0.95
	PF	1.31	1.00	1.31

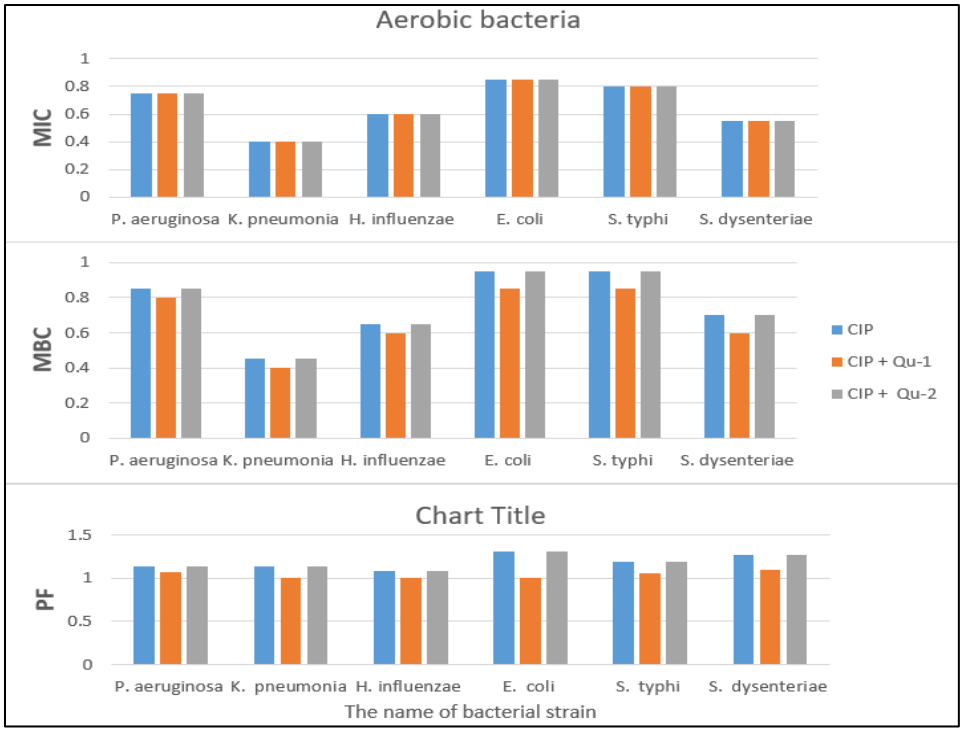

Figure 5. The results of evaluating the impact of mixing **Qu-1** and **Qu-2** with **CIP** against aerobic bacteria

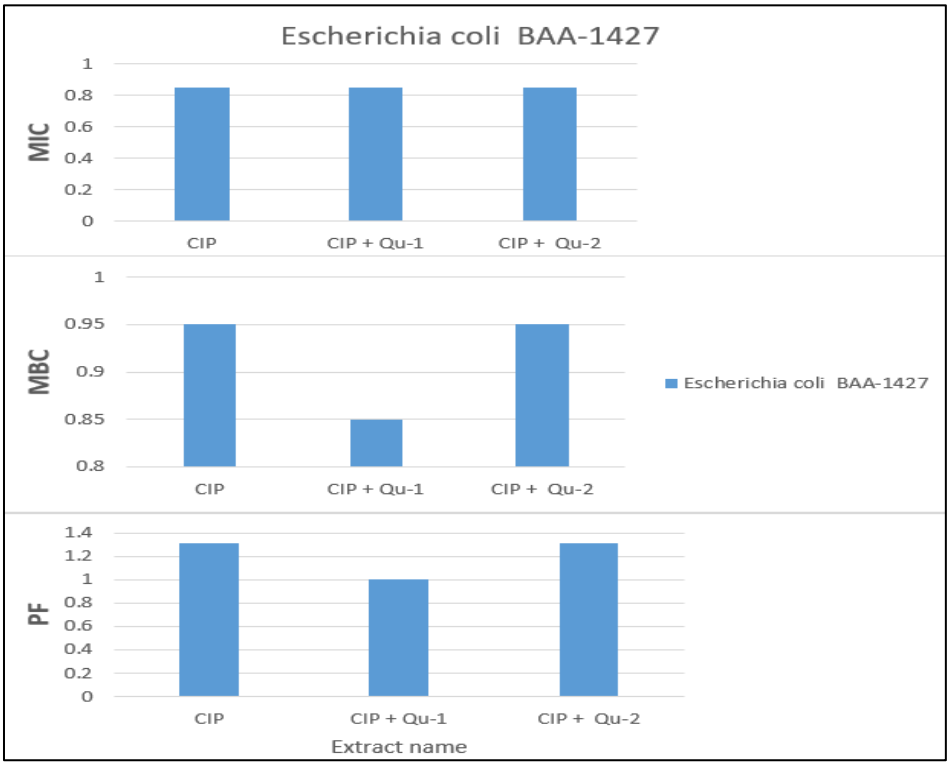

Figure 6. The results of evaluating the impact of mixing **Qu-1** and **Qu-2** with **CIP** against normal bacterial flora

The simultaneous utilization of Qu-1 or Qu-2, however, did not augment the efficacy of MNZ against anaerobic bacteria as depicted in **Table 6** and **Figure 7**.

Table 6. The impact of mixing two plant extracts with **MNZ** on anaerobic bacterial growth

Microbmer and its ATCC value	Antimicrobial-related factor	Codes of the reference antimicrobial drugs and the extracts used		
		MNZ	**MNZ+ Qu-1**	**MNZ + Qu-2**
Bacteroides fragilis ATCC 25285	MIC	3.0	3.0	3.0
	MBC	3.5	3.5	3.5
	PF	1.17	1.17	1.17
Clostridium perfringens ATCC 13124	MIC	0.80	0.80	0.80
	MBC	0.95	0.95	0.95
	PF	1.19	1.19	1.19

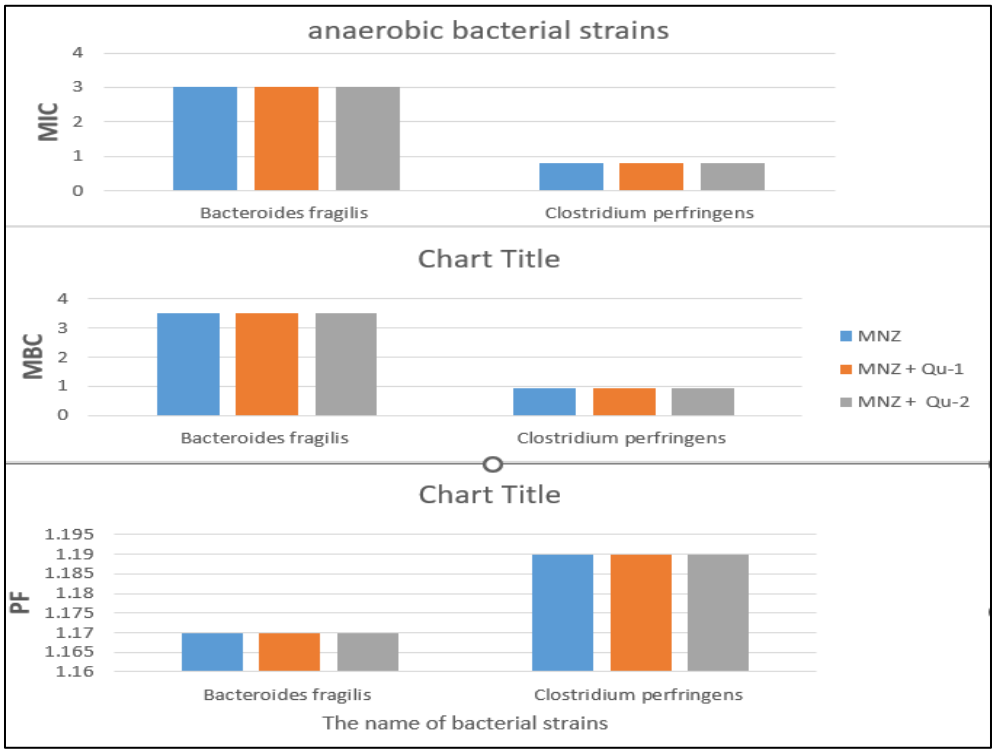

Figure 7. The results of evaluating the impact of mixing **Qu-1** and **Qu-2** with **MNZ** against anaerobic bacteria

Finally, the co-application of Qu-1 can enhance the antifungal potential of NYS by reducing MFC and PF values as seen in **Table 7** and **Figure 8**. Qu-2 on the other hand do not has this effect. This variation may be attributed to higher phenolic and flavonoidal contents of Qu-1 extract.

Table 7. The impact of mixing two plant extracts with **NYS** on pathogenic fungi

Fungi and its ATCC value	Antimicrobial-related factor	Codes of the reference antimicrobial drugs and the extracts used		
		NYS	**NYS +Qu-1**	**NYS + Qu-2**
Candida albicans ATCC 10231	MIC	4	4	4
	MFC	6	5	6
	PF	1.50	1.25	1.50
Aspergillus niger ATCC 16888	MIC	8	8	8
	MFC	12	9	12
	PF	1.5	1.13	1.5

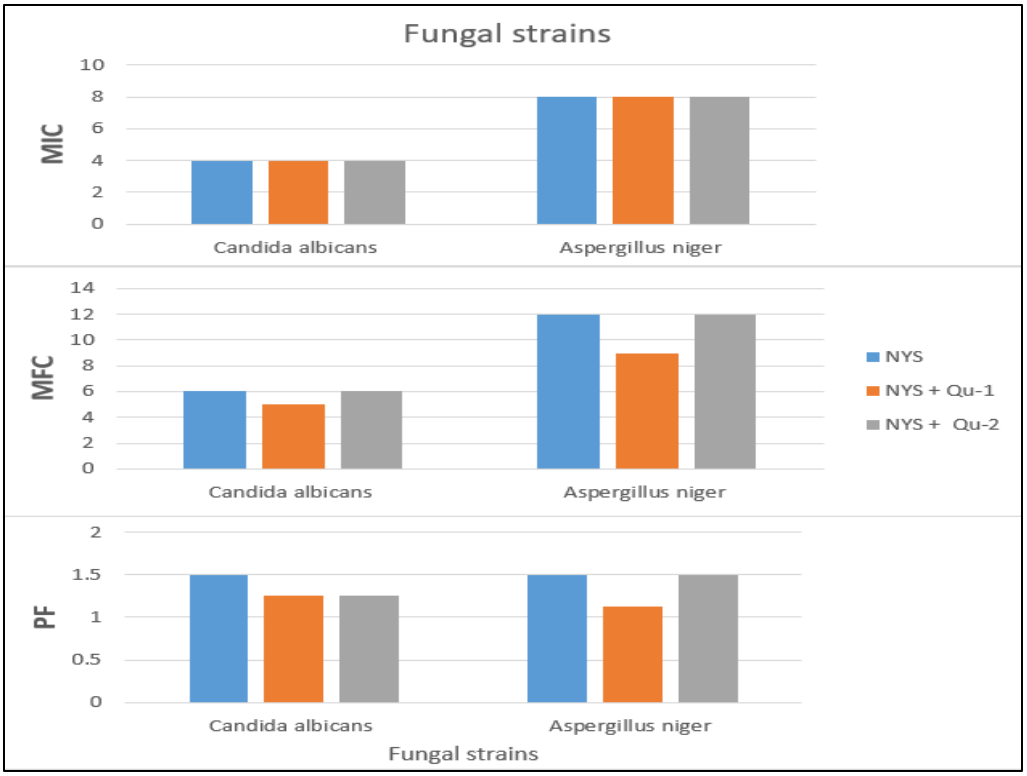

Figure 8. The results of evaluating the impact of mixing **Qu-1** and **Qu-2** with **NYS** against fungal strains.

The observed variability in outcomes implies that diverse techniques yield varying effects; specifically, microwave extraction induces significant heat production within plant cells, facilitating uniform and efficient heating. Consequently, the rapid diffusion of polyphenol compounds from the cells leads to an augmented antimicrobial effect (45).

4. Conclusion

The utilization of M.wv and MUAE approaches enabled the successful extraction of quince seeds utilizing ethyl alcohol. Although these techniques yielded an extract abundant in compounds with phenolic properties and possessed significant antimicrobial attributes, M.wv exhibited superior outcomes in terms of (TPC), (TFC), and antimicrobial activity. Total phenolic contents (TPC) and total flavonoidal contents (TFC) have a positive connection with antimicrobial activity, and this finding may imply that these substances are in part to blame. Nystatin and ciprofloxacin both displayed improved microbial inhibitory activity against the studied fungi and aerobic bacteria when combined with Qu-1. In order to lessen the toxicity of traditional antimicrobials or to boost their efficacy, the extract is suggested to be taken in conjunction. Both extracts exhibited microbicidal activity against all tested strains. It is evident that M.wv and MUAE serve as effective environmentally-friendly techniques for extracting bioactive molecules with antimicrobial properties, with M.wv displaying greater efficiency and requiring shorter treatment times with less intensity. Quince seeds extracts hold promise as a potential source of natural antimicrobials. However, further detailed studies are required to explore their safety, pharmacokinetics, mechanism of action, as well as optimization methods to reduce time, energy, and costs.

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فحص النشاط المضاد للميكروبات لبذور السفرجل المستخلصة باستخدام طريقتين مختلفتين

الخلاصة

المقدمة: تم التأكيد بشكل واضح على ظهور المقاومة للعديد من الأدوية الفعالة المضادة للميكروبات وضرورة البحث عن عوامل بديلة لمعالجة هذا ذلك. **الهدف**: الهدف هو تقييم TFC (إجمالي محتويات الفينول)، وTFC (إجمالي محتويات الفلافونويد)، والنشاط المضاد للميكروبات لمستخلصي بذور السفرجل ودمجهما مع الأدوية المضادة للميكروبات التجارية. **المواد وطرق العمل**: في هذا العمل تم تحضير مستخلصين إيثانوليين (Qu-1) و (Qu-2) من بذور السفرجل باستخدام طريقتين، الميكروويف (M.wv) والاستخلاص المتسلسل بمساعدة الموجات الدقيقة (MSAE). .تم إجراء التقدير الكمي للمحتوى الفينولي الكلي TPC ومحتوى الفلافونويد الكلي TFC في هذه المستخلصات باستخدام طريقة Folin-Ciocalteu و $AlCl_3$على التوالي. تم اختبار النشاط المضاد للميكروبات للمستخلصات، الى جانب دمجها مع المضادات الحيوية التقليدية، للتأكد من نشاطها المضاد للميكروبات ضد مجموعة متنوعة من البكتريا الهوائية واللاهوائية باستخدام سيبروفلوكساسين وميترونيدازول كمراجع على التوالي. تم اختبار المستخلصات أيضًا ضد الفطريات (C. albicans و A. niger) باستخدام النيستاتين كمرجع. **النتائج**: كانت TPC و TFCلـ (Qu-1) 1.82 ± 2.2578 و1.4901 ± 1.76 على التوالي، في حين كانت هذه القيم ل (Qu-2)2.05 ± 1.0341 و0.7342 ± 1.53. أظهر كلا المستخلصين فعالية مضادة للميكروبات ضد جميع مسببات الأمراض التي تم فحصها، حيث أظهر Qu-1 فعالية فائقة. لذلك، هناك علاقة إيجابية بينTPC ، TFCوالنشاط المضاد للميكروبات. يمكن لمزيج Qu-1 أن يعزز كلاً من التأثيرات المضادة للفطريات والمضادة للهوائية لـ NYS و CIPعن طريق تقليل MFC و MBCوعامل الفاعلية المقابل لهما. على العكس من ذلك، فإن مجموعة المضادات الحيوية Qu-2 لا تظهر أي تعديل. يمتلك كلا المستخلصين نشاطًا مبيدًا للميكروبات استنادًا إلى نسبة MBC/MIC التي تقل عن 4. **الاستنتاج**: مستخلصات بذور السفرجل، وخاصة Qu-1، تظهر نتائج واعدة في مكافحة الالتهابات الميكروبية. ويمكن استخدامها بمفردها أو بالاشتراك مع المضادات الحيوية. قد يؤدي المزيد من البحث والتطوير إلى تطوير مضادات ميكروبات جديدة لمعالجة مشكلة المقاومة المتزايدة.

الكلمات المفتاحية: مضادات الميكروبات، الاستخلاص، بذور السفرجل