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Article

Unveiling the antibacterial and antioxidant potential of *Hedera helix* leaf extracts: recent findings

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Abstract

Hedera helix L., a member of the Araliaceae family, is a commonly known decorative plant with recognized medicinal activities. In this study, the ethanolic extract from *H. helix* leaves was investigated for its total polyphenolic and flavonoid contents, as well as its antioxidant and antibacterial properties. The aim was to evaluate its potential for controlling certain infections by screening its antibacterial activity against selected pathogenic bacteria. The total phenolic and flavonoid contents of the extract were determined using colorimetric methods. The antioxidant activity was assessed through two assay methods: the 1, 1-diphenyl-2-picryl hydrazyl (DPPH) free radical scavenging activity and the reducing power ferric reducing/antioxidant power (FRAP). The antibacterial activity against different pathogenic bacteria, including *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumonia*, and *Pseudomonas aeruginosa*, was evaluated using the well diffusion method. The total phenolic and flavonoid contents of the *H. helix* extract were found to be 134.3 ± 4.9 mg gallic acid/g and 42.4 ± 3.6 mg catechin/g, respectively. The extract exhibited antioxidant activity, with a reducing power represented by an FRAP value of 9.5 ± 0.9 mmol Fe^{+2}/g DW and a percentage inhibition of DPPH of 64.7 ± 3.8 at 80 μg/mL. The extract demonstrated antibacterial activity, inhibiting the growth of *K. pneumoniae* and *S. aureus* with zone of inhibition values of 18.5 and 23.2 mm, respectively, using 25 mg/well. However, *E. coli* and *P. aeruginosa* exhibited resistance to the extract. The findings of this study highlight the antibacterial and antioxidant properties of the ethanolic extract from *H. helix* leaves. The extract exhibited significant phenolic and flavonoid contents, as well as antioxidant activity. It also demonstrated antibacterial activity against selected pathogenic bacteria, suggesting its potential for controlling certain infections. Further research is warranted to identify the active compounds responsible for these activities and to explore their mechanisms of action.

Key words: *Hedera helix*, antioxidant activity, antibacterial activity, phenolic compounds, active compounds, mechanisms of action

1. Introduction

Hedera helix L., commonly known as English ivy, is an evergreen liana belonging to the Araliaceae family. It is native to Europe and Western Asia and is characterized by its variable leaf morphology (Baysal and Zeller 2004; Cwientzek et al. 2011; Orhan et al. 2012). From summer to late autumn, *H. helix* produces small greenish-yellow flowers in umbels that measure 3–5 cm in diameter. In winter, it develops small black berries that ripen. The attractive appearance of *H. helix* has made it a popular choice as an ornamental plant in various countries (Flemming 1998; Demirci et al. 2004; Dumitriu et al. 2013).

In addition to its ornamental value, *H. helix* has a history of medicinal use. The leaves of this plant have been traditionally employed for their therapeutic benefits, including their antibacterial and antioxidant activities (Pop et al. 2017). Recent studies have further investigated the antibacterial properties and mechanism of action of *H. helix* extracts against common bacterial strains. Furthermore, the antioxidant activity of various extracts derived from *H. helix* leaves has been evaluated, suggesting its potential in addressing oxidative stress-related disorders (Demirci et al. 2004).

Antibacterial activity refers to the ability of a substance to inhibit the growth or kill bacteria, thereby providing a potential solution for combating bacterial infections. On the other hand, antioxidant activity involves the ability to scavenge harmful free radicals, which are implicated in various diseases and aging processes (Pop et al. 2017). Other studies have focused on investigating the antibacterial and antioxidant properties of *H. helix* leaves extract, aiming to validate its traditional use and explore its potential applications in modern medicine. These studies have employed different extrac-

tion techniques and experimental methods to evaluate the bioactive constituents and their efficacy, supporting its traditional use in folk medicine. Further research is warranted to elucidate the active constituents responsible for these activities and explore their potential application in the development of novel therapeutic agents for bacterial infections and oxidative stress-related disorders.

Hedera helix (Ivy) leaves extract is not only used as an anti-inflammatory remedy for treating respiratory illnesses such as bronchial asthma and acute bronchitis (Schapowal and Group 2004; Kruttschnitt et al. 2020), but it has also shown effectiveness against upper respiratory infections, providing relief for symptoms such as dyspnea, expectoration, shortness of breath, and cough (Schapowal and Group 2004). Active components present in *H. helix*, including hederagenin, hederasaponin-C, alpha-hederin, and hederacoside C, have been utilized in the treatment of viral-origin acute bronchitis (Shokry et al. 2022). Folkloric medicine practices worldwide make use of *H. helix* leaves extract for pain relief and the treatment of various illnesses. This extract is widely used in regions such as Australia, the East Mediterranean, Europe, and South America, as it can expand the bronchial tubes and promote the production of surfactant to maintain alveolar function (Fazio et al. 2009; Sierocinski et al. 2021). Polyphenol-rich plants have been scientifically proven to be effective in preventing diseases associated with oxidative stress, including cancer, neurodegenerative disorders, and cardiovascular diseases (Manach et al. 2004; Mishra and Verma 2009; Salihoğlu et al. 2013). Additionally, there is a correlation between the content of polyphenolic compounds and their antibacterial and antioxidant activities (Mishra and Verma 2009; Pandey and Rizvi 2009).

Despite the widespread use of *H. helix* extract, there is a knowledge gap regarding its antibacterial effects on specific strains of bacteria, including *Staphylococcus aureus* (ATCC 25923), *Escherichia coli* (ATCC 25922), *Klebsiella pneumoniae* (ATCC 13883), and *Pseudomonas aeruginosa* (ATCC 27553). Therefore, the aim of the current research is to investigate the antibacterial effect of *H. helix* extract against these bacterial strains, as well as evaluation of its antioxidant activity, total phenolic content, and flavonoid content in its ethanolic extract.

2. Materials and methods

2.1. Plant material

Hedera helix plants were collected from the wild in the Jenin district of the West Bank in May 2021. The leaves were thoroughly rinsed with distilled water, dried in the shade, and then finely ground using a mortar and pestle.

2.2. Extraction

Approximately 10 g of dried *H. helix* leaves were soaked in 100 mL of 99% ethanol and subjected to ultra-sonication for 90 min. The resulting mixture was then filtered, and the solvent was evaporated using a rotary evaporator under reduced pressure. The obtained viscous crude extract was carefully collected and transferred into small glass screw-capped bottles, which were then stored in a refrigerator for further analysis. Prior to analysis, the extract was dissolved in 99% ethanol to achieve a concentration of 1.0 mg/mL. The extraction process was performed in triplicate for each of the three samples.

2.3. Determination of total phenolic content

To determine the total phenolic content of the extract, 1.8 mL of Folin–Ciocalteu reagent (diluted by adding 5 mL of the reagent to 50 mL of distilled water) was added to 50 μL of the sample extract. After 5 min, 1.2 mL of 7.5% sodium carbonate solution was added to the mixture. The mixture was then left for 120 min, allowing the reaction to occur. Subsequently, the absorbance at 760 nm was measured using a spectrophotometer. The total phenolic content of the extract was calculated using a calibration curve prepared with gallic acid standards. The results were expressed as milligrams of gallic acid per gram of crude extract.

2.4. Determination of total flavonoid content

To determine the total flavonoid content (TFC) of the extract, 100 μL of the crude extract was mixed with 0.3 mL of 10% $AlCl_3$, 4 mL of distilled water, and 0.3 mL of 5% $NaNO_2$ solution. After a 6 min incubation period, 2.5 mL of distilled water and 2 mL of 1N NaOH were added to the mixture. The absorbance of the resulting solution was then measured at 510 nm using a spectrophotometer.

A calibration curve prepared with catechin standards was utilized to determine the TFC of the extracts. The results were expressed as milligrams of catechin per gram of crude extract.

2.5. Free-radical scavenging activity using DPPH (antioxidant activity)

The 1, 1-diphenyl-2-picryl hydrazyl (DPPH) radical method is employed to assess the antioxidant capacity of substances by measuring their ability to scavenge the stable DPPH radical. In this study, aliquots of a 0.0634 mmol/L DPPH solution in methanol (4 mL) were mixed with different concentrations (0.1–2 mg/mL) of the extract (50 μL). The mixtures were then incubated in the dark at 25 °C for 30 min. A positive standard containing vitamin C was prepared using the same method.

The absorbance of the sample and standard solutions was measured at 515 nm. The scavenging activity of the DPPH radicals was calculated using the equation: scavenging activity (%) = (Abs (Control) Abs (Sample))/ Abs (Control) ×100, where Abs (Sample) represents the absorbance of the tested plant extract and Abs (Control) is the absorbance of the control (containing all reagents except the plant extract.)

By plotting the inhibition (%) against the extract concentration, the 50% inhibitory concentration (IC50) of the extract was determined. The IC50 of vitamin C was also determined and used as a positive control for comparison.

2.6. FRAP assay

The antioxidant activity of the extracts was determined using the ferric reducing/antioxidant power (FRAP) assay, adapted from the method described by Benzie and Strain

(1999). In this assay, 40 μL of the extract was mixed with 3.0 mL of freshly prepared FRAP reagent, which was pre-warmed to 37 °C. The reaction mixtures were then incubated at 37 °C. Instead of the original 4 min incubation period used in the FRAP experiment, the absorbance at 593 nm was measured after 1 h of incubation, comparing it to a reagent blank containing distilled water that was also incubated at 37 °C.

For calibration, aqueous solutions with known concentrations of Fe^{+2} ranging from 2 to 5 mmol/L were used. The results were expressed as mill moles of Fe^{+2} per gram of the extract.

2.7. Antimicrobial activity using well diffusion method

The antibacterial activities of the ethanolic extract of *H. helix* leaves were evaluated using the well diffusion method on Mueller–Hinton agar (MHA). Four different concentrations of the extract (25, 12.5, 6, and 3 mg/well) were tested in triplicate against the selected bacteria.

The bacterial strains used for the antibacterial assay were *S. aureus* (ATCC 25923), *E. coli* (ATCC 25922), *K. pneumoniae* (ATCC 13883), and *P. aeruginosa* (ATCC 27553). Bacterial colonies were obtained from agar plates after 18–24 h of incubation to prepare a bacterial suspension with a turbidity equivalent to 0.5 McFarland standard (corresponding to 1.5×10^8 colony-forming units/mL). The turbidity of the bacterial suspension was measured at 625 nm. Under aseptic conditions, the MHA agar plates were inoculated with each bacterial strain. Wells with a diameter of 6 mm were created, and 50 μL of the test samples were added to the wells. The plates were then incubated at 37 °C for 24 h.

Following the incubation period, the diameter of the growth inhibition zones around the wells was measured in millimeters. All tests were performed in triplicate to ensure accuracy and reproducibility.

2.8. Statistical analysis

Statistical analyses were conducted using the SAS software (SAS Institute Inc., Cary, USA, Release 8.02, 2001). Mean comparisons were performed using the GLM procedure, treating the main factors separately through one-way Analysis of variance. To maintain an experiment-wise significance level of 5%, the Bonferroni procedure was applied with multiple t tests. Statistical significance was considered at a p value less than 0.05.

The results are presented as mean ± standard deviation and were obtained from triplicate measurements.

3. Results

3.1. Total phenolic contents (TPC)

The total phenolic content of the *H. helix* leaf extract was determined using the Folin method and a calibration curve prepared with various concentrations of gallic acid. The results were expressed as milligrams of gallic acid per gram of crude extract. The total phenolic content of the plant ethanolic extract was found to be 134.3 ± 4.9 mg/g (Table 1).

Table 1. Antioxidant activity (ferric reducing/antioxidant power (FRAP), mmol Fe^{+2}/g DW) and total phenolic (mg gallic acid/g) and flavonoids contents (mg catechin/g) of ethanolic extracts of *Hedera helix*.

FRAP*	Total phenolic content	Total flavonoids content
9.5 ± 0.9	134.3 ± 4.9	42.4 ± 3.6

*Results are expressed as average ± standard deviation of three samples.

3.2. Total flavonoid content (TFC)

The TFC in the extracts was determined using the aluminum chloride method with a calibration curve prepared using various concentrations of catechin. The results are expressed as milligrams of catechin per gram of crude extract. The TFC in the ethanolic extracts of the investigated plant materials was found to be 42.4 ± 3.6 mg catechin/g, indicating a rich presence of flavonoids in the plant (Table 1).

3.3. Antioxidant activity

Antioxidant activity is indicative of the presence of potent oxygen radical scavengers such as phenolic compounds. There are two main types of antioxidant assays used to evaluate the antioxidant activity of plant extracts. The first category measures the reducing potential of plant extracts such as the ferric reducing/antioxidant power assay, which assesses the ability of plant extracts to reduce ferric ions to ferrous ions. The second category evaluates the ability of plant extracts to scavenge free radicals with examples such as the ABTS and DPPH assays.

3.3.1. Reducing potential of plant extracts (FRAP assay)

The FRAP assay measures the reducing capability of antioxidants by reacting with a ferric tripyridyltriazine (Fe^{3+}-TPTZ) complex, producing a colored ferrous tripyridyltriazine (Fe^{2+}-TPTZ) compound. The presence of compounds that can donate hydrogen atoms to break free radical chains determines their reducing capabilities. Under low pH conditions, the iron-TPTZ complex is reduced to a blue-colored iron-TPTZ compound. The antioxidant activity of the plant extracts based on the FRAP assay is presented in Table 1, with a calculated value of 9.5 ± 0.9 mmol Fe^{2+}/g of dry plant material.

3.3.2. DPPH assay

The DPPH assay is based on the ability of the stable free radical 2,2-diphenyl-1-picrylhydrazyl (DPPH) to react with hydrogen donors such as phenolics. The bleaching of the DPPH solution increases linearly with increasing extract concentration. Figure 1 displays the percentage inhibition of DPPH at various concentrations of the crude extract (ranging from 10 to 150 μg/mL). The results demonstrate a dose-dependent scavenging action of the extracts, showing a linear correlation between the concentration and the percentage of DPPH inhibition. This suggests that higher concentrations of the extract exhibit a stronger ability to neutralize free radicals.

Fig. 1. Percentage inhibition of 1, 1-diphenyl-2-picryl hydrazyl (DPPH) free radicals by different concentrations of *Hedera helix* plant extract. (Results are expressed as average ± standard deviation of three samples.)

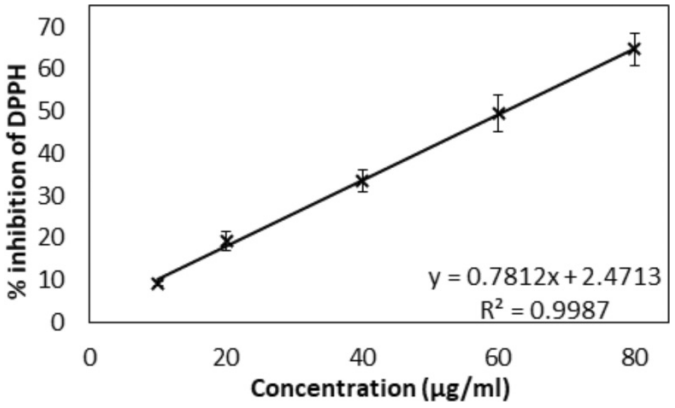

Table 2. Inhibition zones (mm) for *Hedera helix* extract at different concentrations (mg/well) against different microbes.

Microbes	25	12.5	6	3
S. aureus	23.2 ± 1.5	14.4 ± 2.0	8.0 ± 2.2	R
E. coli	R	R	R	R
P. aeruginosa	R	R	R	R
K. pneumonia	18.5 ± 2.0	12.5 ± 1.5	7.5 ± 2.0	R

Note: Data shown as means ± SD ($n = 3$), R = no sensitivity (zone of inhibition < 7.0 mm).

3.4. Antimicrobial activity

The antibacterial properties of the extract were evaluated using the Agar well diffusion method given by NCCLS (1997, 1999). The inhibitory zones (mm) were measured, and the antibacterial activity was categorized based on the criteria established by NCCLS (1997): inactive: inhibitory zone < 9 mm, less active inhibitory zone between 9 and 12 mm, active: inhibitory zone between 13 and 18 mm, and very active: inhibitory zone > 18 mm.

Table 2 presents the antibacterial activity of the *Hedera helix* extract against the targeted bacteria in terms of inhibition zones (mm). The results indicate that the *H. helix* extract exhibited inhibitory effects against *K. pneumoniae* and *S. aureus*, while *E. coli* and *P. aeruginosa* were not affected. These findings suggest that the *H. helix* extract may have potential therapeutic applications in the treatment of respiratory illnesses caused by *S. aureus* and *K. pneumoniae*.

4. Discussion

This study deals with investigation of antibacterial and antioxidant activities of *H. helix* leaves from Palestine. The extract was prepared by soaking 10 g of H. helix leaves in 100mL of 99% ethanol and subjected to ultra-sonication for 90 min, and this may yield different constituents with novel antibacterial and antioxidant properties.We used comparative analysis in our investigation and compared our findings with the previous ones whether in correlations or contradictions.

4.1. Total phenolic and flavonoids contents

The results of the study revealed a significant total phenolic content in the ethanolic extract of *H. helix* leaves. This indicates that the extract is rich in phenolic compounds, which are known for their potential health benefits and biological activities. The presence of high phenolic content in *H. helix* has been reported in previous studies. For example, a study by Zaiter et al. (2018) investigated the phenolic composition of various *H. helix* extracts and found significant levels of phenolic compounds, including gallic acid and its derivatives. Antibacterial activity is closely associated with the phenolic content of plant extracts. Phenolic compounds act as potent antioxidants by scavenging free radicals and protecting cells from oxidative damage. In line with the high phenolic content, the ethanolic extract of *H. helix* leaves in this study demonstrated antioxidant activity.

Moreover, the presence of phenolic compounds in *H. helix* has been linked to its potential antimicrobial properties. A study by Pop et al. (2017) investigated the antimicrobial activity of *H. helix* leaf extracts and found that they exhibited inhibitory effects against various bacterial strains, including *S. aureus* and *E. coli*.

The results of the present study are consistent with these previous findings, as the ethanolic extract of *H. helix* leaves showed significant total phenolic content and exhibited antibacterial activity against selected pathogenic bacteria, such as *S. aureus* and *K. pneumoniae* (Shokry et al. 2022). In conclusion, the findings of this study highlight the high total phenolic content of the ethanolic extract from *H. helix* leaves. This supports the plant's potential for various biological activities, including antioxidant and antibacterial properties. The presence of phenolic compounds in the extract suggests its potential use in developing natural remedies for oxidative stress-related diseases and as a potential antimicrobial agent. Further studies can explore the specific phenolic compounds present in the extract and their individual contributions to the observed activities.

Results of the study also indicate a significant TFC in the ethanolic extracts of the investigated plant materials. This finding suggests that the plant is rich in flavonoids, which are a diverse class of secondary metabolites known for their various biological activities and potential health benefits. Several studies have reported the presence of flavonoids in *H. helix* and highlighted their potential bioactivities. For example, a study by Roşca-Casian et al. (2017) analyzed the flavonoid composition of different *H. helix* extracts and identified several flavonoids, including catechin and its derivatives. These flavonoids have been associated with antioxidant, anti-inflammatory, and antimicrobial properties.

4.2. Bioactivities of phenolic and flavonoid contents

The antioxidant activity of flavonoids is well documented in the literature. Flavonoids act as potent antioxidants by scavenging free radicals and reducing oxidative stress. The presence of a high flavonoid content in the ethanolic extracts

of the investigated plant materials suggests their potential antioxidant activity. Moreover, flavonoids have been reported to possess antimicrobial properties. In a study by Pop et al. (2017), the antimicrobial activity of *H. helix* leaf extracts was evaluated, and it was found that the extracts exhibited inhibitory effects against various bacterial strains. Flavonoids present in the extracts could contribute to their antimicrobial activity.

The results of this study align with previous research, as the ethanolic extracts of the investigated plant materials demonstrated a significant TFC. This supports the potential bioactivities associated with flavonoids, including their antioxidant and antimicrobial properties.

In conclusion, the findings of this study indicate a high TFC in the ethanolic extracts of the investigated plant materials. This suggests the presence of bioactive flavonoids, which may contribute to the observed antioxidant and antimicrobial activities. Further research can focus on identifying and characterizing specific flavonoid compounds present in the extracts and exploring their individual contributions to the biological activities exhibited by the plant materials.

4.3. Antioxidant activity

The results of the FRAP assay in this study indicate the antibacterial activity of the plant extracts. The FRAP assay measures the reducing power of antioxidants by assessing their ability to donate hydrogen atoms and break free radical chains. The reaction between the antioxidants and the Fe^{3+}-TPTZ complex results in the formation of a colored Fe^{2+}-TPTZ compound. The calculated FRAP value of 9.5 ± 0.9 mmol Fe^{2+}/g of dry plant material suggests that the investigated plant extracts possess significant antioxidant activity. This value represents the reducing potential of the extracts, indicating their capacity to neutralize oxidants and reduce ferric ions. Several studies have employed the FRAP assay to evaluate the antibacterial activity of plant extracts and identify potential sources of natural antioxidants. The FRAP assay is widely recognized and utilized due to its simplicity, reproducibility, and ability to provide quantitative results. In support of the findings in this study, previous research has demonstrated the antibacterial activity of *H. helix* extracts using the FRAP assay. For instance, a study by Šuran et al. (2021) evaluated the antioxidant properties of various plant extracts, including *H. helix*, using the FRAP assay. The authors reported significant FRAP values, indicating the antioxidant potential of *H. helix* extracts. Moreover, the antioxidant activity of plant extracts measured by the FRAP assay has been linked to the presence of phenolic compounds, such as flavonoids and phenolic acids. These compounds possess strong reducing capabilities and contribute to the overall antioxidant capacity of the extracts. In conclusion, the results of the FRAP assay in this study demonstrate the antioxidant activity of the investigated plant extracts. The calculated FRAP value of 9.5 ± 0.9 mmol Fe^{2+}/g of dry plant material indicates the reducing potential of the extracts. These findings are consistent with previous studies that have evaluated the antioxidant properties of *H. helix* extracts using the FRAP assay. The presence of phenolic compounds in the extracts may contribute to their antioxidant activity.

The results of the DPPH assay in this study indicate the antioxidant activity of the investigated plant extracts. The DPPH assay is widely used to assess the ability of compounds to scavenge free radicals. The DPPH stable free radical reacts with hydrogen donors, such as phenolic compounds, resulting in the bleaching of the DPPH solution. The extent of bleaching is directly proportional to the antioxidant activity of the tested extracts.

The findings of this study are consistent with previous research highlighting the antioxidant activity of *H. helix* extracts using the DPPH assay. For example, a study conducted by Pop et al. (2017) evaluated the antioxidant potential of different extracts from *H. helix* leaves using the DPPH assay. The authors reported dose-dependent scavenging activity of the extracts, supporting the notion that *H. helix* possesses significant antioxidant properties. Furthermore, numerous studies have employed the DPPH assay to assess the antioxidant activity of various plant extracts and identify potential natural antioxidants. The linear correlation between the extract concentration and the percentage of DPPH inhibition is a commonly observed trend, indicating the effectiveness of the tested extracts in scavenging free radicals. In conclusion, the results of the DPPH assay in this study provide evidence of the antioxidant activity of the investigated plant extracts. The dose-dependent scavenging action demonstrated by a linear correlation between the extract concentration and the percentage of DPPH inhibition confirms the ability of the extracts to neutralize free radicals. These findings align with previous research highlighting the antioxidant potential of *H. helix* extracts using the DPPH assay.

4.4. Antimicrobial activity

The results obtained from the Agar well diffusion method in this study provide insights into the antibacterial properties of the *H. helix* extract. The inhibitory zones, measured in millimeters, were used to assess the antibacterial activity against selected bacteria. Different ranges of inhibitory zone sizes were categorized as inactive, less active, active, and very active.

Hedera helix extracts in this study exhibited inhibitory effects against *K. pneumoniae* and *S. aureus*, as evidenced by the measured inhibition zones. On the other hand, the extract did not show significant activity against *E. coli* and *P. aeruginosa*. These findings are consistent with previous studies that have investigated the antibacterial activity of *H. helix* extracts. For instance, a study conducted by Iqbal (2018) evaluated the antibacterial potential of different extracts from *H. helix* against various pathogenic bacteria. The authors reported inhibitory effects of the extracts against both gram-positive (including *S. aureus*) and gram-negative bacteria (including *K. pneumoniae*), supporting the antimicrobial properties of *H. helix*. The observed inhibitory effects against *S. aureus* and *K. pneumoniae* are particularly significant, as these bacteria are known to cause respiratory illnesses. In regard to the activity against *E. coli* and *P. aeruginosa*, our findings were contradictory to previous study, which could be because this study used

different bacterial strains with different resistance (Pop et al. 2017; Shokry et al. 2022). The data obtained in this study were confirmed and validated in different trails and by statistical analysis. The potential therapeutic applications of the *H. helix* extract in the treatment of respiratory infections caused by these bacteria are supported by the results of this study. In conclusion, the results obtained from the Agar well diffusion method indicate that the *H. helix* extract possesses antibacterial activity against *K. pneumoniae* and *S. aureus*. These findings align with previous research highlighting the antimicrobial properties of *H. helix* extracts. The potential therapeutic applications of the extract in the treatment of respiratory illnesses caused by *S. aureus* and *K. pneumoniae* are supported by the observed inhibitory effects. Although previous studies had similar and different findings, our investigation's uniqueness was in the way we extracted the leaves, and some of our findings were different than others. Overall, our study has strengthened the need for the use of *H. helix* extract in herbal medicine.

5. Conclusions

This study aimed to investigate and summarize the antibacterial, antioxidant, total phenolic content, and total flavonoid contents of *H. helix*, a plant abundantly grown in Palestine. The results revealed that the plant extract exhibited antibacterial activity against *S. aureus* and *K. pneumoniae*, indicating its potential as a natural antibacterial agent. Furthermore, the plant extract showed high levels of flavonoids, phenolics, and antioxidants, suggesting its significant antioxidant potential. These findings highlight the potential of *H. helix* as a valuable natural source of potent antioxidants, which could have preventive effects against various diseases. The plant extract's potential applications extend to the food, cosmetics, and pharmaceutical industries. Further research is warranted to explore the specific compounds responsible for the antioxidant activity, such as phenolic acids and flavonoids, and to investigate the mechanisms of their action in both in vitro and in vivo settings.

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