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RESEARCH ARTICLE

Antimicrobial efficacy of different solvent extracts of *Quercus infectoria* G.Olivier against UTI pathogens and evaluation of antioxidant, anti-inflammatory and cytotoxic effects

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Abstract

Multidrug-resistant urinary tract infections (UTIs) are a major global concern for the health sector. *Quercus infectoria* G.Olivier has been used as a medicine for ages in Asia and it is well established of its antibacterial, antifungal, antioxidant, antiparasitic, anti-inflammatory and wound healing properties. The aqueous, ethanolic and methanolic extract and dry powder forms of *Q. infectoria* were investigated for their antibacterial, anti-inflammatory and antioxidant qualities. Various assays were done to determine anti-microbial, cytotoxic effects, antioxidant and anti-inflammatory effects. The aqueous extract of *Q. infectoria* expressed antibacterial properties against *Enterococcus faecalis*, *Escherichia coli* and *Candida albicans*, while the methanolic extract is effective against *Klebsiella* species by the well diffusion assay. The cytotoxic effects of *Q. infectorias'* aqueous extract revealed a 90 % survival rate with 5 and 10 µg/mL in the brine shrimp lethality assay (BSLA) method. The methanolic extract demonstrated an 89.13 % suppression of antioxidant activity at a concentration of 50 µg/mL. The methanolic extract demonstrated 85.13 % suppression of antioxidant activity in the H_2O_2 experiment at a concentration of 50 µg/mL. Similarly, aqueous extract showed dose-dependent anti-inflammatory actions in egg albumin denaturation inhibition assay (EA assay), bovine serum albumin denaturation assay (BSA assay) and membrane stabilisation assay (MSA assay). The current investigation showed that *Q. infectoria* has anti-inflammatory and antioxidant properties. Moreover, it has antimicrobial actions against selected UTI pathogens.

Keywords: anti-inflammatory activity; antimicrobial activity; antioxidant activity; cytotoxic effect; *Quercus infectoria*

Introduction

Quercus infectoria G.Olivier, also known as the Aleppo oak, is a deciduous tree taxonomically classified under the family Fagaceae. The tree is widely distributed in Mediterranean regions, including Greece, Turkey, Iran, Syria and parts of Asia Minor. It is also found in the Himalayan belt in India and its galls, known as "Mazo", possess therapeutic properties and are rich in tannins and other phytochemicals. Traditional Chinese medicine has historically used *Q. infectoria* galls (QIG) to treat a variety of human conditions, including skin diseases, diarrhoea and bleeding. QIGs' medical uses have grown in popularity in the Middle East and Asia Minor (1). Pharmacological evidence and documentation support the antidiabetic, local anaesthetic, antibacterial, antifungal and anti-inflammatory properties of *Q. infectoria* galls. *Quercus infectoria* has many other bioactive compounds like free gallic acid, ellagic acid and tannic acid (2). Other bioactive compound includes glycosides, flavonoids, sterols, phenolic compounds, quercetin, hesperidin, rutin, narigenin, acids and fatty acids (3, 4).

The most frequent causes of genitourinary tract infections are *Escherichia coli*, *Staphylococci* and *Streptococci*. Gram-negative bacteria are also the cause of genitourinary tract infections. Although uncommon, anaerobic bacterial infections can also result in genitourinary tract infections and may be linked to pyometra. Geographically, genitourinary infections are found all over the world (4). Recurrent and complicated UTIs can lead to significant morbidity, renal damage and healthcare burden.

Since reactive oxygen species (ROS) target cell macromolecules, oxidative damage is a primary cause of many diseases (4, 5). *Quercus infectoria* neutralise these free radicals and antioxidants are essential in preventing ROS from damaging cells (6). Plant-based antioxidants protect cells by scavenging free

oxygen radicals and counteracting ROS (6, 7). This has been made feasible by the presence of specific phytochemicals that give plants antioxidant action, such as flavonoids. Many diseases have been linked to nitric oxide (NO) and bodys' overproduction of NO can lead to cell damage, neuronal cell death and DNA fragmentation (8). Because of their effective NO scavenging activity, plants can significantly contribute to the reduction of NO levels (9, 10). Although some infections are asymptomatic, lower UTI can cause dysuria, haematuria, or pollakiuria. Fever or stomach-ache may accompany acute pyelonephritis and pyometra (11). If sepsis develops, symptoms could include diarrhoea, vomiting and lethargy. Abdominal distention and/or vaginal discharge may accompany pyometra (12).

A well–known substance on *Q. infectoria*, mazo is used to treat infectious disorders, particularly UTIs and to help women in the postpartum phase (13). Although other microbes may also be responsible, the three most significant ones that cause vaginal infections in women are *Candida albicans*, *Trichomonas vaginalis* and *Gardnerella vaginalis* (14). When the extract was prepared using ethanol, it was demonstrated that there was inhibition on the β–lactamase and autolysin enzymes of *Staphylococcus aureus*. It has antimicrobial actions against methicillin-resistant *Staphylococcus aureus* (MRSA) (15). Moreover, it was established that Mazo aqueous and methanol extracts had antibacterial properties against *Klebsiella pneumonia*, *Escherichia coli*, *Staphylococcus*, *Enterococcus faecalis*, *Proteus mirabilis*, *Streptococcus agalactiae* and *Proteus vulgaris* (16).

Pathogens, causing urinary tract infections have been thoroughly investigated in this study. The studys' originality lies in the fact that finding alternative or herbal therapies for the treatment of UTIs is vital, given the rise in antimicrobial resistance (AMR) (17). The purpose of the study helps us to understand the therapeutic potential and safety profile of *Q. infectoria*, which also has antibacterial, antioxidant and anti-inflammatory activity and cytotoxic effect. This research aids in the hunt for potent all-natural antimicrobials. The reliability and application of the results are increased by using a variety of assays to assess the bioactivities, which yield solid and thorough data.

Materials and Methods

Preparation of *Q. infectoria* extract

The powdered *Q. infectoria* was purchased from Annai Herbals in Poonamallee, Chennai and 2.5 g of *Q. infectoria* was weighed. Annai Aravindh Herbals, founded by Dr. P. Rajalingam and Dr. R. Vijayalakshmi, focuses on developing scientifically prepared herbal medicines. With GMP and ISO 9001 and 22000 certifications, the company ensures strict quality control from sourcing to processing. Their products are made using a purification method to guarantee safety and effectiveness. Aqueous extract was made ready by following the method: 2.5 g of *Q. infectoria was* mixed in 50 mL of distilled water and heated with the heating mantle. The resultant mixture was kept for 15 to 20 min at 50 to 60 °C. To get rid of insoluble particles, it was filtered and the resulting translucent filtrate, which comprised the aqueous extract of *Q. infectoria*, was immediately refrigerated and kept in the refrigerator for further experimental investigation.

Methanol extract was prepared by taking 2.5 g of *Q. infectoria*. It was mixed with 25 mL of methanol and shaken with an orbital shaker for a full day. For 5 min, the methanol-mediated *Q. infectoria* extract was heated to 50 °C in a heating mantle. Methanolic extract was then employed for other biological uses (18). Similarly, ethanol extract was prepared by taking 2.5 g of *Q. infectoria*. 2.5 g of *Q. infectoria* was added to 25 mL of ethanol. This mixture was taken in an orbital shaker and thorough mixing was done for 24 hr in the shaker. The ethanol-mediated *Q. infectoria* extract was placed in the heating mantle for 5 min at 50 °C. Further experiments were conducted using the ethanolic extract (19).

Antimicrobial activity

The well diffusion method was used on agar plates and the method used was the Mueller-Hinton method. The plates were autoclaved at 121 °C for 15–20 min. Sterile Petri plates were covered with the medium. After taking out the agar plates from the autoclave, they were placed on a stand so that the plates could cool down to room temperature. Utilising a sterilised cotton swab, *E. coli*, *Klebsiella* species, *E. faecalis* and *C. albicans* microbial suspension were applied evenly over the agar plates. In these agar plates, well–shaped depressions were cut, measuring 9 mm. To these wells, 50 µg/mL of the different extracts–aqueous, ethanolic and methanolic of *Q. infectoria* were added to the wells. The well for dry powder of *Q. infectoria* was also done. Bacterial culture was incubated for 24 hr and fungal culture was incubated for 48 hr. The temperature was maintained at 36 °C. A clear area around each well is due to the antimicrobial activity. This is measured with the scale in mm (20).

Cytotoxic effect

Brine shrimp lethality assay

10 mL of saline water was poured into Enzyme-Linked Immunosorbent Assay (ELISA) plates with 6 wells. Now, ten nauplii were gradually introduced to each of these wells to which *Q. infectoria* solvent extract was also added. The percentage of live nauplii was estimated using the formula that was used in previous studies (21).

Antioxidant property

2,2-Diphenyl-1-picrylhydrazyl scavenging assay (DPPH scavenging assay)

After preparing the stock solution, it was diluted in methanol. 200 µL of working solution for DPPH was prepared fresh for each assay and poured into the 96-well ELISA plate. To these wells, *Q. infectoria* extracts (10–50 µg/mL) were loaded and for 10 min, the plate was left at room temperature in a dim environment. Using menthol as a blank and a spectrophotometer at 517 nm, the absorbance was measured (19).

% DPPH Scavenging activity = (Absorbance of control - Absorbance of sample × 100) / Absorbance of control (Eqn. 1)

In the above equation, the DPPH solution without *Q. infectoria* serves as the control and the DPPH solution containing *Q. infectoria* serves as the sample. As a standard, ascorbic acid was employed.

Hydrogen peroxide (H_2O_2) radical scavenging assay

Q. infectoria has radical-removing property and this was tested with an H_2O_2 radical scavenging assay. 0.6 mL of the 40 mM H_2O_2 solution was taken after it had been prepared in phosphate buffer (pH 7.4). To this was added *Q. infectoria* extract and a standard solution of ascorbic acid in concentrations (10–50 µg/mL). The absorbance was detected at 532 nm using spectrophotometry following 10 min of

incubation in a dark environment. The standard used was vitamin C. The above formula, used for the DPPH assay, was used to express the percentage of scavenging property of H_2O_2 (22).

Ferric reducing antioxidant power assay (FRAP Assay)

Reagents for FRAP assay: To the mixture of sodium acetate trihydrate and glacial acetic acid, distilled water is added to bring the level up to 1 L. Ferric chloride hexahydrate and tripyridyl–s–triazine are the additional components. The above 3 reagents are mixed in the ratio of 10:1:1 and are known as the FRAP reagent. A solution was prepared with the FRAP reagent (2.7 mL) and *Q. infectoria* extracts (0.7 mL). After taking each of the three extracts in varying concentrations, they were kept at room temperature in a dark environment for some time. A spectrophotometer analysis of the reduction was tested using ascorbic acid. A spectrophotometric measurement was taken at 593 nm (19).

2,2'–azino–bis(3–ethylbenzothiazoline–6–sulphonic acid (ABTS assay)

ABTS was mixed with potassium persulfate; this reagent was refrigerated for 24 hr and just before the assay, 50 % ethanol was used to dilute the reagent. 96-well microplates were taken and 250 µL of ABTS+ and 20 µL of *Q. infectoria* samples of different amounts were loaded into the wells. The standard used was ascorbic acid and the blank was 20 µL of ethanol. The microplate was kept in the dark for 10 min and a spectrophotometric measurement was taken at 734 nm. The same formula used for scavenging activity was used here.

Nitric oxide (NO) radical inhibition assay

In the present study, a modified Griess–Illosvoy reagent was used. 3 mL of the reagent, consisting of sodium nitroprusside (2 mL), phosphate-buffered saline (0.5 mL) and Q. *infectoria* different solvent extracts (10–50 µg/mL) and standard solution (ascorbic acid, 0.5 mL), was taken. This mixture of reagents was stored at 25 °C for 2.5 hr. After the procedure was finished, 1 mL of sulfanilic acid reagent was mixed with 0.5 mL of the reaction mixture and the combination was left for 5 min to achieve full diazotisation. A pink chromophore appeared in diffused light when this combination was mixed with modified Griess Illosvoy reagent and allowed to stand for 30 min at 25 °C. A spectrophotometer was used to test these solutions' absorbance at 540 nm in relation to the corresponding blank solutions (23).

Anti–inflammatory activity

Bovine serum albumin denaturation assay

0.45 mL of bovine serum albumin was mixed with 0.05 mL of *Q. infectoria* at various concentrations (10–50 µg/mL). The pH is kept at 6.3. The mixture is allowed to stand at room temperature for 10 min before being incubated in a water bath at 55 °C for 30 min. Dimethyl sulfoxide served as the control and diclofenac sodium as the benchmark. At 660 nm, spectrophotometric measurements were made. The percentage for estimating protein denaturation was computed using the established formula (24).

Egg albumin denaturation assay

0.2 mL of fresh egg albumin was mixed with 2.8 mL of 1X phosphate buffer. *Quercus infectoria* was added to this combination in varying doses (10–50 µg/mL), the pH was kept at 6.3 and it was left to remain at room temperature for 10 min. For 30 min, this mixture was incubated at 55 °C in a water bath. Dimethyl sulfoxide served as the control and diclofenac sodium served as the benchmark. The samples were then analyzed at 660 nm using spectrophotometry (25).

The percentage of protein denaturation was determined utilising the following formula :

% inhibition = Absorbance of control-Absorbance of sample × 100/ Absorbance of control (Eqn. 2)

Membrane stabilisation assay (MSA)

For membrane stabilisation activities, we preferred the *in vitro* MSA in the current study. Here, cell membrane stabilisation is assessed while treating with the bioactive compound and observing its ability to prevent cell membrane disruption. The following items were required for the procedure: human red blood cells, Tris–HCl buffer (50 mM, pH 7.4), phosphate-buffered saline, various doses and various solvents of *Q. infectoria* (10–50 µg/mL), centrifuge tubes and UV–Vis spectrophotometers (26).

Assay procedure

Each centrifuge tube received 1 mL of RBC suspension before different concentrations of *Q. infectoria* (different solvent extracts) (10–50 µg/mL) were applied. After gradually mixing the aforesaid RBC mixture with different amounts of *Q. infectoria*, the tubes were incubated for 30 min at 37 °C. To pellet the RBCs, centrifugation was performed at 2500 rpm for 5 min at room temperature. A UV–Vis spectrophotometer was used to measure the supernatants' absorbance at 560 nm.

Using the following formula, the % inhibition of hemolysis was calculated:

% inhibition= [(OD control – OD sample) / OD control] × 100 (Eqn. 3)

where the absorbance of the RBC suspension with the test compound is known as the OD sample and the absorbance of the RBC suspension without the test compound is known as the OD control.

Results

Preparation of extracts

An aqueous extract of *Q. infectoria* was prepared by weighing 2.5 g of the plant material and dissolving it in 50 mL of distilled water. The mixture was boiled using a heating mantle at 50–60 °C for 15–20 min, followed by filtration through a muslin cloth to obtain the aqueous extract (Fig. 1a–1e).

For the ethanolic extract, 2.5 g of *Q. infectoria* was mixed with 25 mL of ethanol and shaken on an orbital shaker for 24 hr to obtain the ethanol extract, which was used for biomedical applications (Fig. 2).

Similarly, the methanolic extract was prepared by mixing 2.5 g of *Q. infectoria* with 25 mL of methanol and shaking on an orbital shaker for 24 hr, and the resulting extract was used for biomedical applications (Fig. 3).

Antimicrobial activity

The agar well diffusion technique was used to demonstrate the antimicrobial activity of *Q. infectoria*. Thereby, it was demonstrated that *Q. infectoria* was effective for UTI pathogens, like *E. coli*, *Klebsiella* sp., *E. faecalis* and *C. albicans*. Three different solvents of *Q. infectoria*—aqueous extract, ethanolic extract and methanolic extract—and dry powder of *Q. infectoria* (10 µg/mL) were tested.

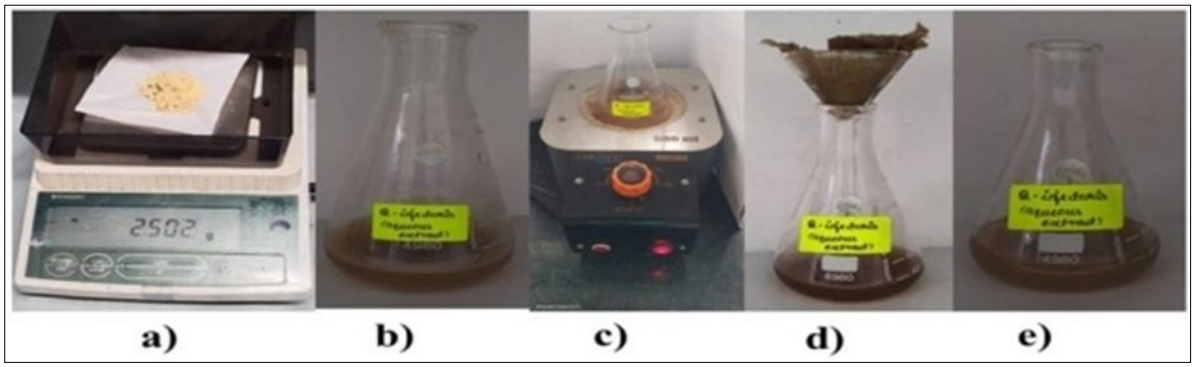


Fig. 1. a–e. The preparation of plant extract and aqueous extract of *Q. infectoria*.

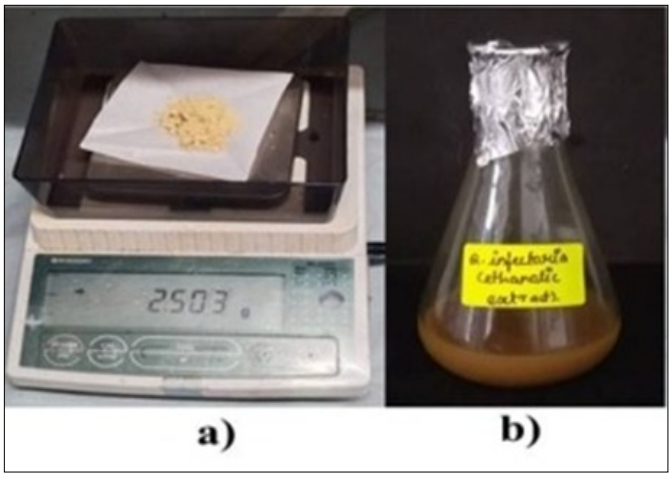


Fig. 2. a, b. Ethanol extract of *Q. infectoria*.

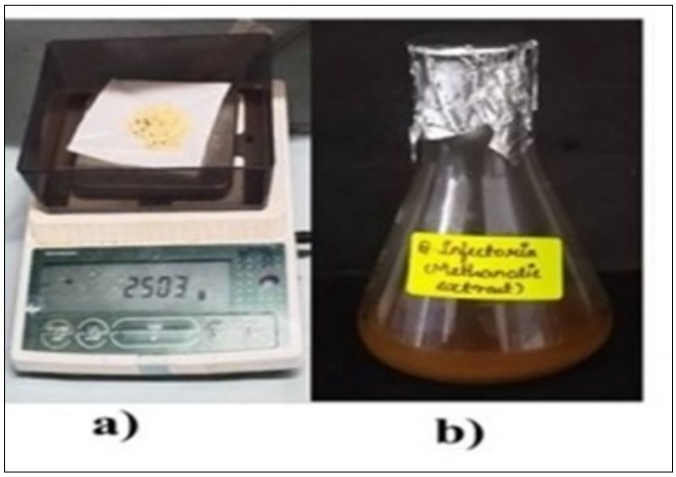


Fig. 3. a, b. Methanol extract of *Q. infectoria*.

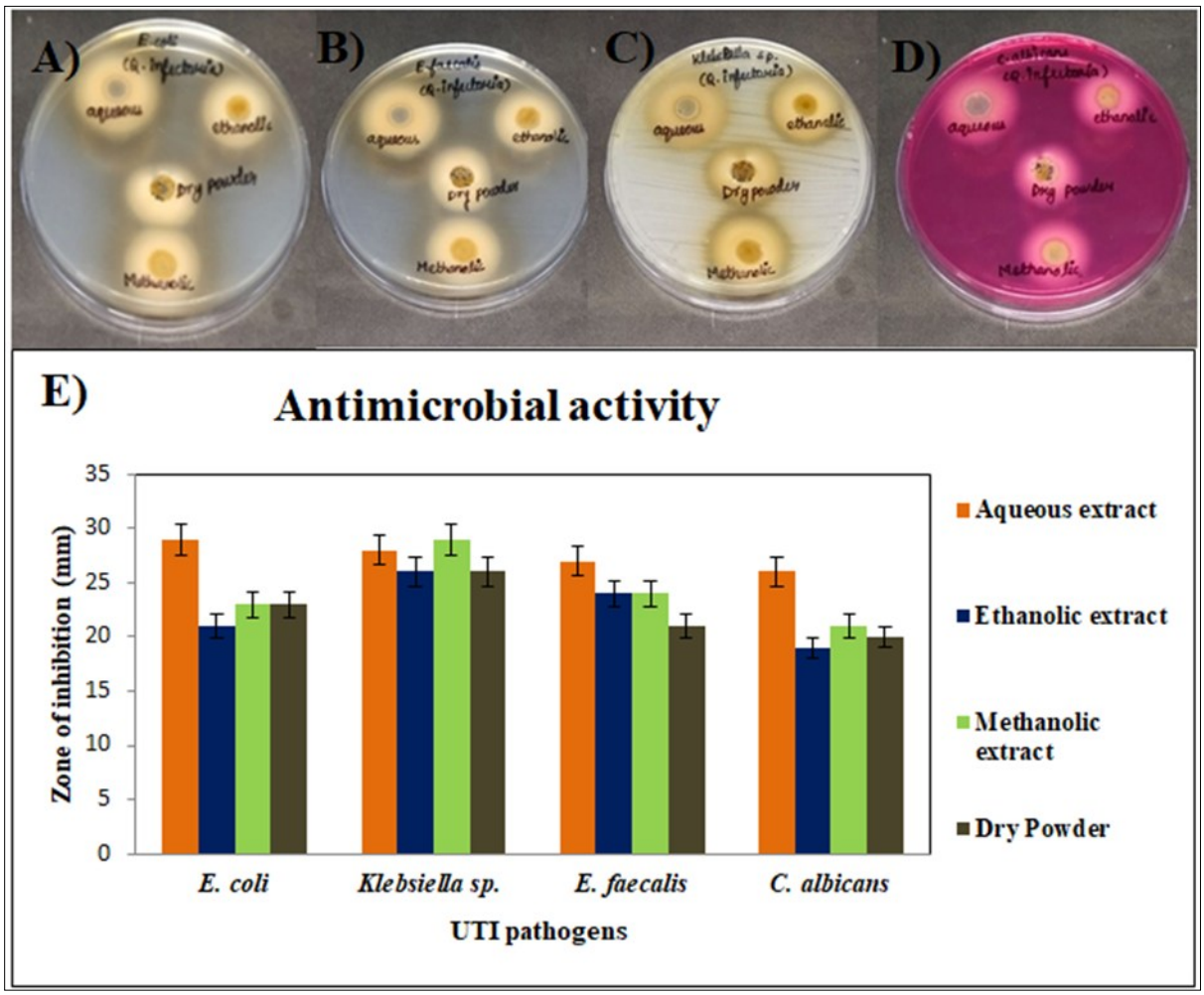


Fig. 4. The microbial plates represented the antimicrobial activity of different solvents and dry powder of *Q. infectoria* against UTI pathogens: (A) *E. faecalis*; (B) *Klebsiella* sp.; (C) *C. albicans*; (D) *E. coli*; (E) The bar diagram shows the antimicrobial effect of *Q. infectoria* aqueous extract, ethanolic extract, methanolic extract and dry powder against UTI pathogens.

Fig. 4 demonstrated that *Q. infectoria* was effective against all UTI pathogens. The zone of inhibition was greatest for the aqueous extract. At 50 µg concentration, *Q. infectoria*-aqueous extract showed inhibition zone of 29 mm (*E. coli*), 28 mm (*Klebsiella* sp.), 27 mm (*E. faecalis*) and 26 mm (*C. albicans*), while the ethanolic extract exhibited 21 mm (*E. coli*), 26 mm (*Klebsiella sp.*), 24 mm (E. *faecalis*) and 19 mm (*C. albicans*), while the methanolic extract exhibited an inhibition zone of 23 mm (*E. coli*), 29 mm (*Klebsiella* sp.), 24 mm (*E. faecalis*) and 21 mm (*C. albicans*) and *Q. infectoria* dry powder exhibited an inhibition zone of 23 mm (*E. coli*), 26 mm (*Klebsiella sp.*), 21 mm (E. *faecalis*) and 20 mm (*C. albicans*). The present study demonstrates that *Q. infectoria* aqueous extract has high antimicrobial activity, thus being effective against microbial growth.

Cytotoxic effect

The cytotoxic activity of the methanolic extract of *Q. infectoria* against *Artemia* nauplii is presented in Fig. 5A. *Quercus infectoria* methanolic extract of different concentrations ranging from 5 µg/mL to 80 µg/mL was compared to the control. The findings demonstrated that 100 % live nauplii were present on day 1, indicating lower toxicity. On the second day, 5 µg/mL, 10 µg/mL and 20 µg/mL concentrations demonstrated 80 % of live nauplii, whereas 40 µg/mL showed 60 % and 80 µg/mL concentrations and 50 % of live nauplii. Using concentrations ranging from 5 µg/mL to 80 µg/mL, the cytotoxic effect of *Q. infectoria* aqueous extract was compared with that of the control (Fig. 5B). The trials' findings showed that, in the first 24 hr, it did not result in the death of *Artemia* nauplii. On Day 2, at concentrations up to 40 µg/mL, *Q. infectoria* aqueous extract demonstrated 80 % live nauplii and only 40 % live nauplii at higher concentrations of 80 µg/mL. Fig. 5C shows the Cytotoxic effect of *Q. infectoria-ethanolic* extract. The results showed that on Day 1, there was 100 % live nauplii at all concentrations, indicating lower toxicity. On day two, a 5 µg/mL concentration showed 80 % live nauplii and an 80 µg/mL concentration showed 50 % live nauplii.

Antioxidant activity

The antioxidant activity of the methanolic extract of *Q. infectoria* was evaluated using multiple *in vitro* assays. The DPPH radical scavenging assay showed a concentration-dependent increase in antioxidant activity, with percentage inhibition values of 61.27, 72.61, 80.56, 84.69 and 89.13 at concentrations ranging from 10 to 50 µg/mL (Fig. 6A). The hydrogen peroxide (H_2O_2) scavenging assay exhibited inhibition percentages of 49.2, 51.6, 63.2, 72.9 and 85.4 across the same concentration range (Fig. 6B). The ferric reducing antioxidant power (FRAP) assay, compared with standard antioxidants, demonstrated percentage inhibition values of 68.53, 73.17, 77.49, 80.38 and 87.55 at concentrations of 10–50 µg/mL (Fig. 6C). The nitric oxide (NO) scavenging assay revealed inhibition percentages of 66.32, 72.65, 75.84, 80.22 and 84.61 at concentrations of 10–50 µg/mL (Fig. 6D). The ABTS radical scavenging assay further confirmed the antioxidant potential of the methanolic extract, with inhibition values of 65.38, 71.46, 78.25, 82.54 and 87.63 at concentrations of 10–50 µg/mL (Fig. 6E).The antioxidant activity of the ethanolic extract of *Q. infectoria* was assessed using multiple *in vitro* assays. The DPPH radical scavenging assay demonstrated concentration-dependent activity, with percentage inhibition values of 63.15, 74.58, 82.36, 85.44 and 89.69 at concentrations ranging from 10 to 50 µg/mL (Fig. 7A). The hydrogen peroxide (H_2O_2) scavenging assay showed inhibition percentages of 48.6, 53.7, 64.1, 74.6 and 86.2 at the same concentration range (Fig. 7B). The ferric reducing antioxidant power (FRAP) assay, compared with standard antioxidants, revealed inhibition values of 68.95, 73.49, 78.12, 82.57 and 87.63 at concentrations of 10–50 µg/mL (Fig. 7C). The nitric oxide (NO) scavenging assay indicated inhibition percentages of 69.28, 75.47, 76.51, 81.36 and 86.93 across the tested concentrations (Fig. 7D). The ABTS radical scavenging assay further confirmed antioxidant activity, with inhibition percentages of 68.53, 72.41, 79.84, 83.22 and 87.65 at concentrations of 10–50 µg/mL (Fig. 7E).

Similarly, the aqueous extract of *Q. infectoria* exhibited notable antioxidant activity across all assays. In the DPPH assay, the extract showed percentage inhibition values of 63.57, 74.68, 82.17, 85.43 and 89.65 at concentrations of 10–50 µg/mL (Fig. 8A). The H_2O_2 scavenging assay demonstrated inhibition percentages of 48.9, 54.6, 63.1, 74.8 and 86.7 at the same concentration range (Fig. 8B). The FRAP assay revealed antioxidant activity with inhibition values of 69.84, 73.56, 77.43, 82.55 and 87.41 at concentrations of 10 –50 µg/mL (Fig. 8C). The NO scavenging assay showed percentage inhibition values of 67.81, 74.62, 77.49, 81.53, and 86.46 across the tested concentrations (Fig. 8D). The ABTS assay also demonstrated strong antioxidant potential, with inhibition percentages of 69.84, 73.56, 77.43, 82.55 and 87.41 at concentrations of 10–50 µg/mL (Fig. 8E).

Anti–inflammatory activity

The anti-inflammatory activity of *Q. infectoria* was evaluated using bovine serum albumin (BSA), egg albumin (EA) and membrane stabilization assays (MSA), with comparisons made against standard drugs (Fig. 9A–C). The aqueous extract of *Q. infectoria* exhibited concentration-dependent anti-inflammatory activity, showing percentage inhibition values of 44, 56, 68, 74 and 79 at concentrations of 10–50 µg/mL in the BSA assay (Fig. 9A). In the EA assay, the aqueous extract demonstrated inhibition percentages of 48, 59, 64, 67 and 77 across the same concentration range (Fig. 9B). Similarly, the MSA assay revealed inhibition values of 43, 57, 66, 74 and 80 at concentrations of 10–50 µg/mL for the aqueous extract (Fig. 9C).

The ethanolic extract of *Q. infectoria* also showed notable anti-inflammatory activity in all three assays. In the BSA assay, the ethanolic extract exhibited percentage inhibition values of 42, 57, 67, 72 and 78 at concentrations of 10–50 µg/mL (Fig. 9A). The EA assay showed inhibition percentages of 48, 57, 60, 64 and 76 for the ethanolic extract across the same concentration range (Fig. 9B). In the MSA assay, inhibition values of 43, 55, 65, 73 and 81 were observed at concentrations of 10–50 µg/mL (Fig. 9C).

Discussion

In the current study, it is hypothesised that aqueous, ethanolic and methanolic extracts of *Q. infectoria* demonstrate antimicrobial properties for UTI pathogens. The assay of *Q. infectoria* demonstrated the antibacterial and cytotoxic effects, antioxidant and anti–inflammatory properties. A 50 µg concentration of aqueous extract of *Q. infectoria* showed a maximum zone of inhibition (ZOI) of 29 mm for *E. coli*. Similarly, ethanolic extract, methanolic extract and dry powder showed ZOIs of 26 mm, 29 mm and 26 mm, respectively, for *Klebsiella*. The antimicrobial action of *Q. infectoria* showed that the aqueous extract of *Q. infectoria* is highest against *E. coli*, *E. faecalis* and *C. albicans*, whereas the

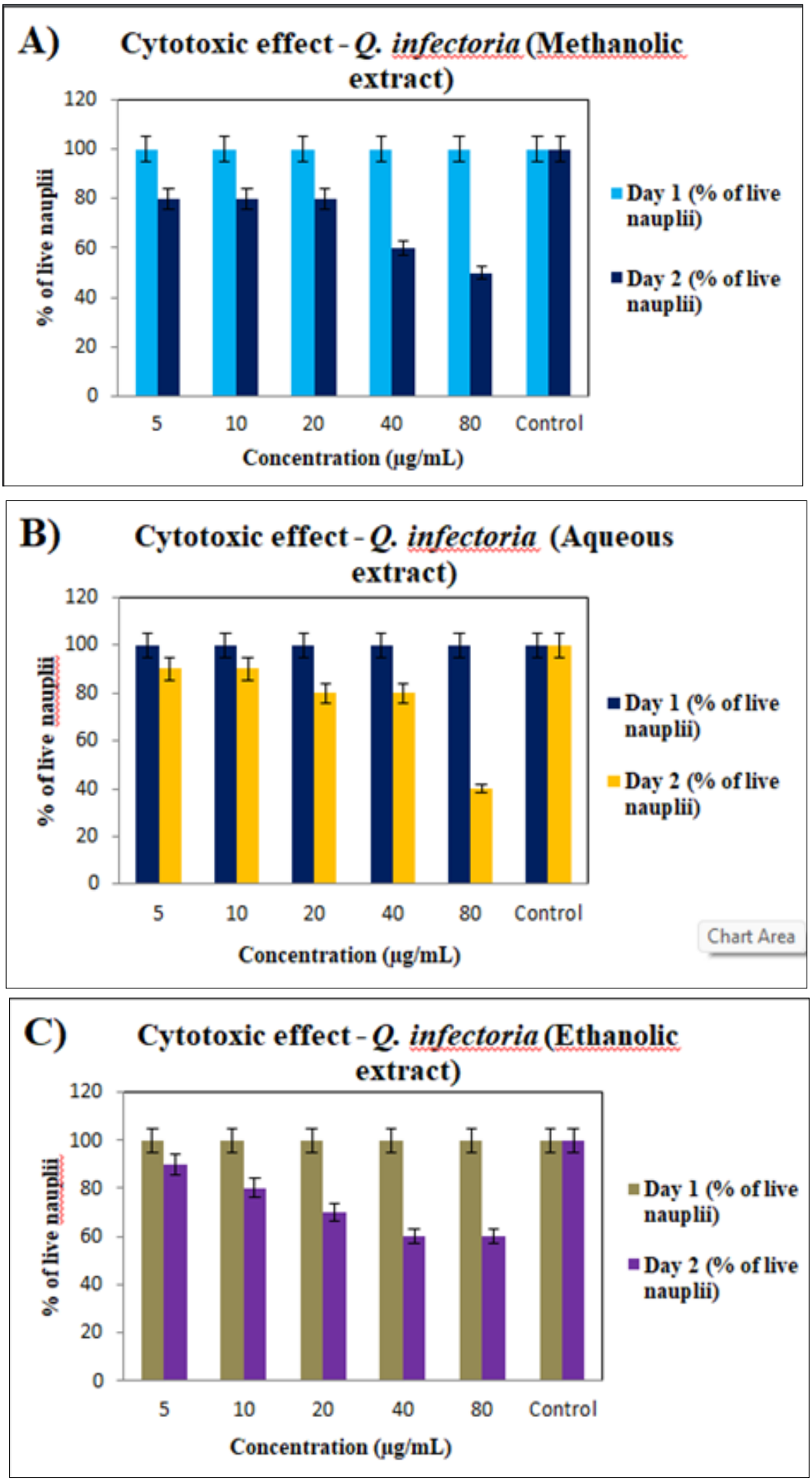
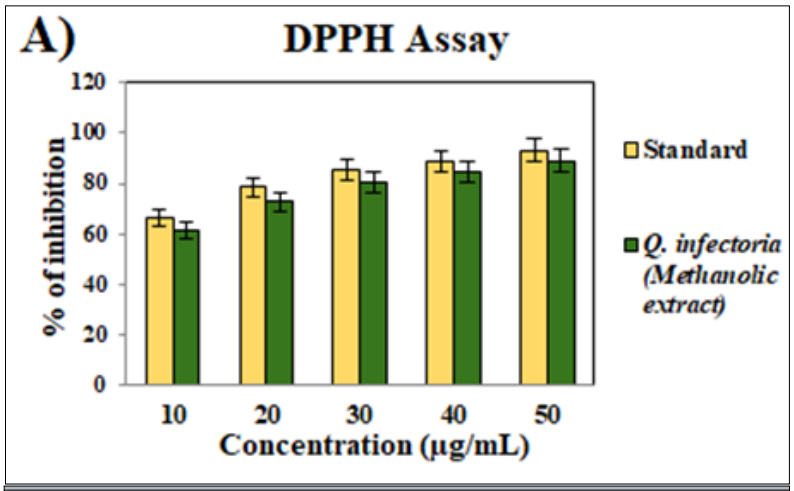
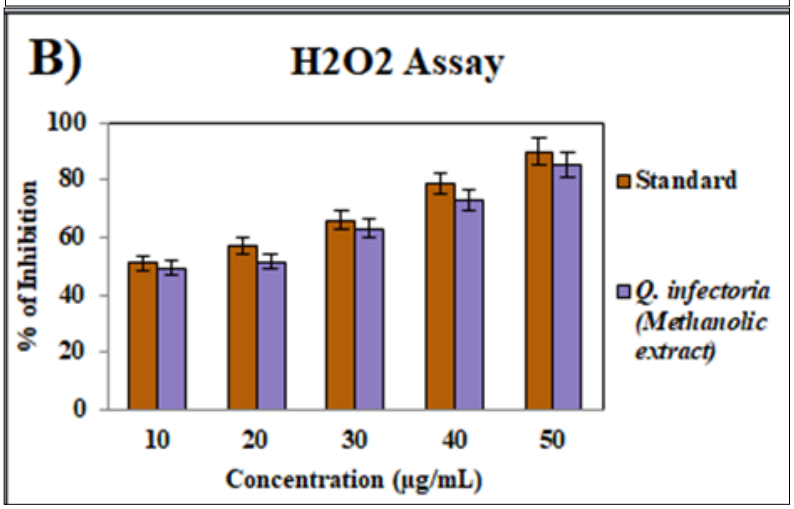
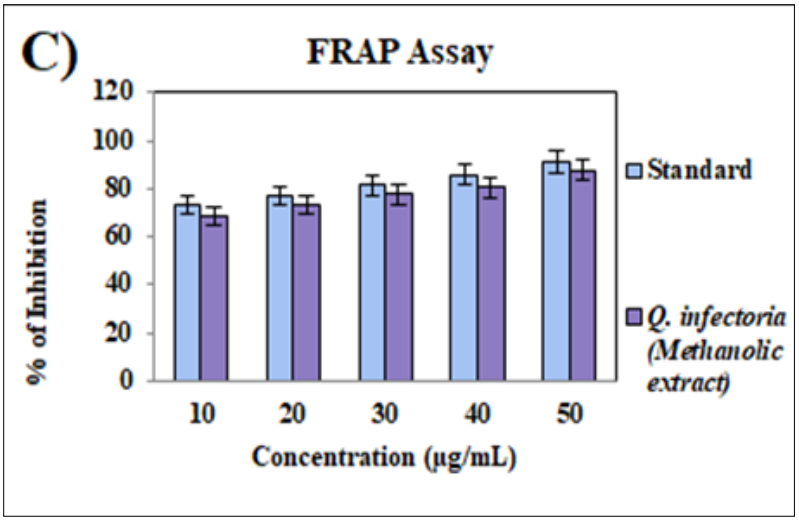
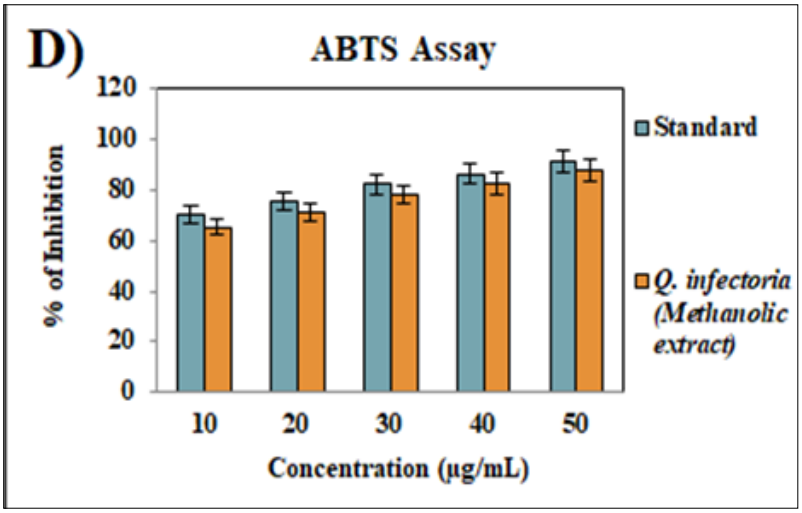


Fig. 5. (A–C) The cytotoxic effect of *Q. infectoria*. On day 1, all extracts had a safety profile compared to the control in all concentrations. On day 2, there was concentration dependent reduction in the percentage of live nauplii in all extracts.


A)
DPPH Assay
% of inhibition
120
100
80
60
40
20
0
10
20
30
40
50
Concentration (µg/mL)
Standard
Q. infectoria (Methanolic extract)


B)
H2O2 Assay
% of Inhibition
100
80
60
40
20
0
10
20
30
40
50
Concentration (µg/mL)
Standard
Q. infectoria (Methanolic extract)


C)
FRAP Assay
% of Inhibition
120
100
80
60
40
20
0
10
20
30
40
50
Concentration (µg/mL)
Standard
Q. infectoria (Methanolic extract)


D)
ABTS Assay
% of Inhibition
120
100
80
60
40
20
0
10
20
30
40
50
Concentration (µg/mL)
Standard
Q. infectoria (Methanolic extract)

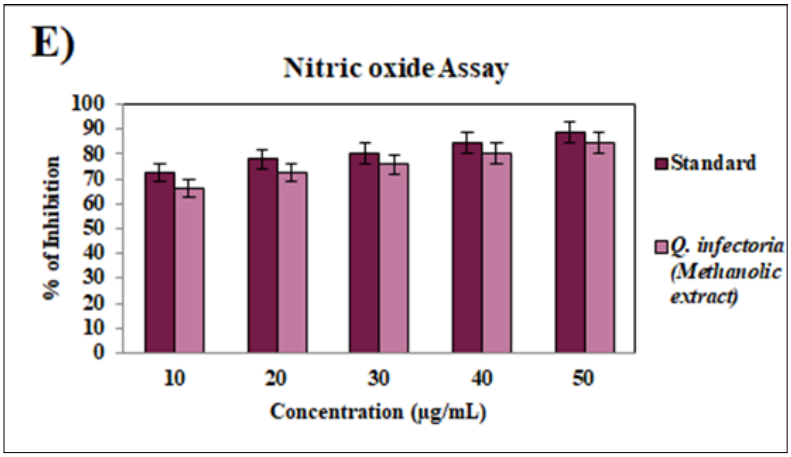
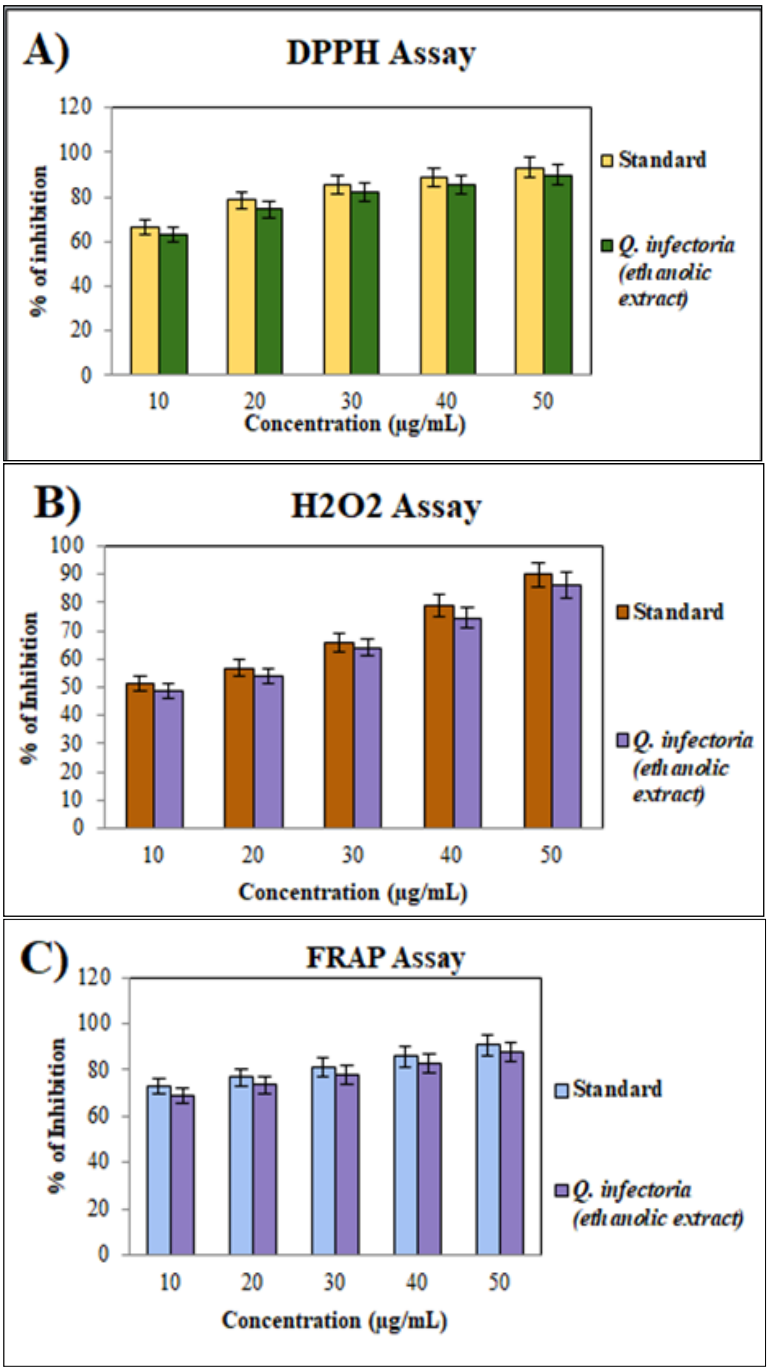


Fig. 6. Different antioxidant assays of *Q. infectoria* methanolic extract. (A) DPPH assay; (B) H_2O_2 assay; (C) FRAP assay; (D) ABTS assay; (E) NO assay showing the percentage of inhibition at different concentrations with comparable antioxidant activity when comparing the extract and standards. This shows the antioxidant property of *Q. infectoria* methanolic extract.

DPPH assay: 1,1–diphenyl–2–picrylhydrazyl radical scavenging assay; H_2O_2 assay: hydrogen peroxide scavenging assay; FRAP assay: ferric reducing antioxidant power assay; NO assay: nitric oxide radical scavenging assay; ABTS assay: 2,2'–azino–bis(3–ethylbenzothiazoline–6–sulfonic acid) radical scavenging assay.



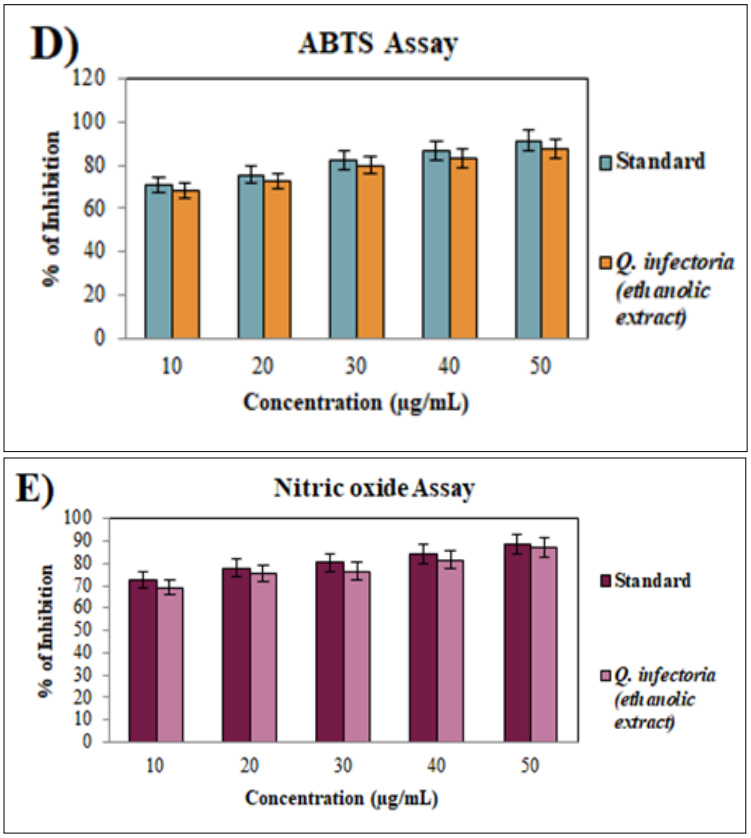
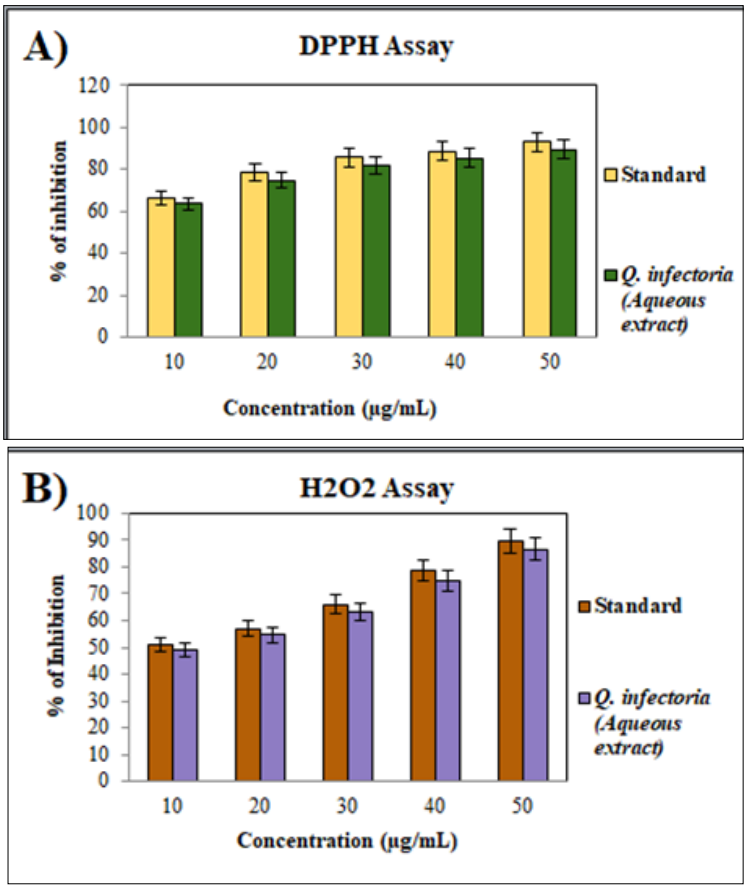


Fig. 7. Antioxidant activity of *Q. infectoria* ethanolic extract. (A) DPPH assay; (B) H_2O_2 assay; (C) FRAP assay; (D) ABTS assay; (E) NO assay. In the various assays, the percentage of inhibition at different concentrations was compared with the standard. This shows the antioxidant property of *Q. infectoria* ethanolic extract.

DPPH assay: 1,1–diphenyl–2–picrylhydrazyl radical scavenging assay; H_2O_2 assay: hydrogen peroxide scavenging assay; FRAP assay: ferric reducing antioxidant power assay; NO assay: nitric oxide radical scavenging assay; ABTS assay: 2,2'–azino–bis(3–ethylbenzothiazoline–6–sulfonic acid) radical scavenging assay.



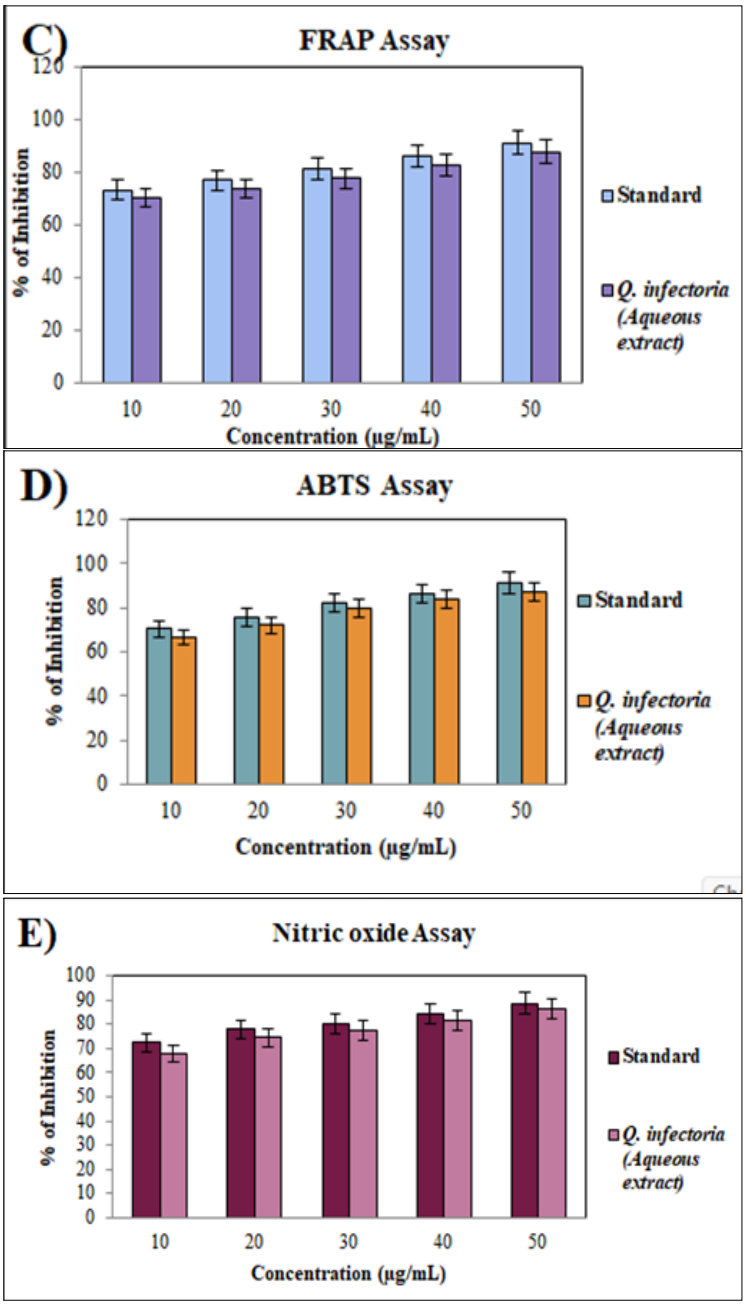
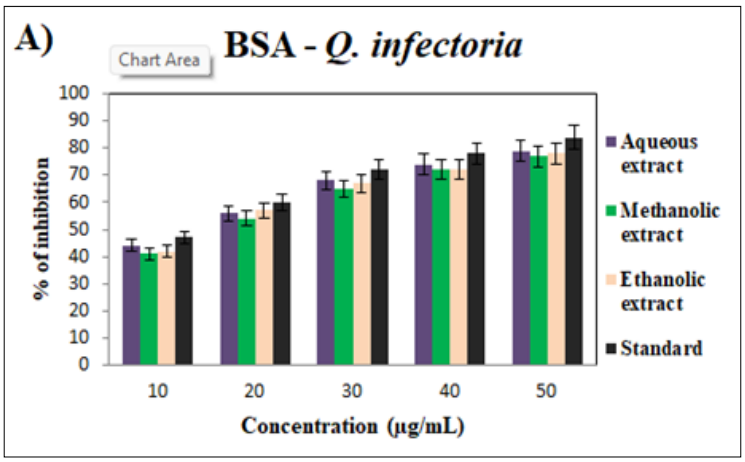


Fig. 8. Antioxidant activity of *Q. infectoria* aqueous extract using different assays. (A) DPPH assay; (B) H_2O_2 assay; (C) FRAP assay; (D) ABTS assay; (E) NO assay. The percentage of inhibition of *Q. infectoria* aqueous extract was compared with the standard, thus demonstrating the antioxidant activity.

DPPH assay: 1,1–diphenyl–2–picrylhydrazyl radical scavenging assay; H_2O_2 assay: hydrogen peroxide scavenging assay; FRAP assay: ferric reducing antioxidant power assay; NO assay: nitric oxide radical scavenging assay; ABTS assay: 2,2′–azino–bis(3–ethylbenzothiazoline–6–sulfonic acid) radical scavenging assay.



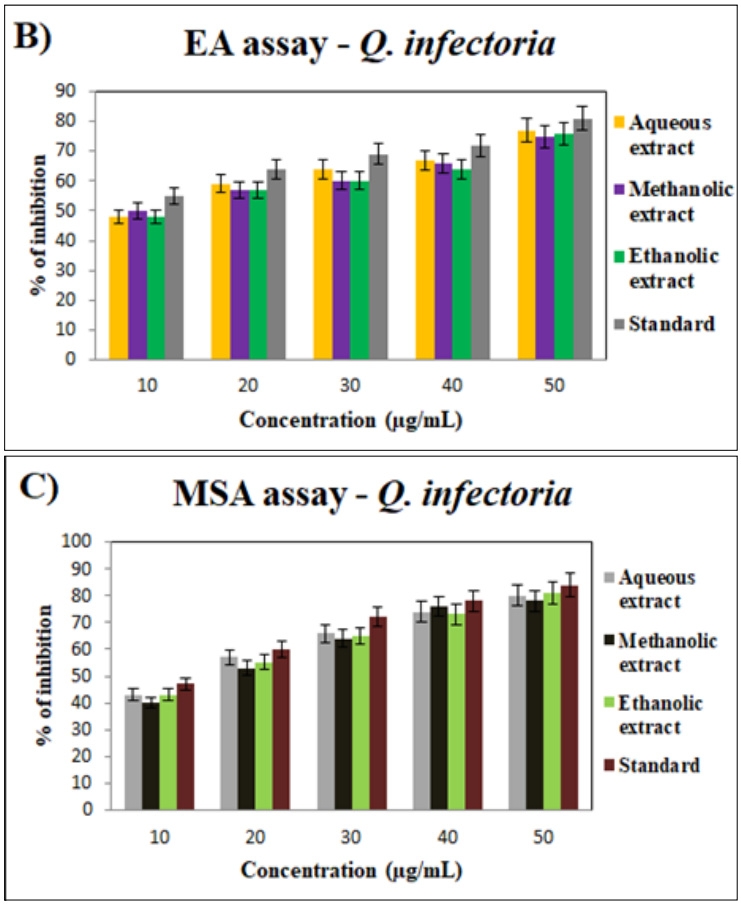


Fig. 9. Anti-inflammatory activity. (A) BSA assay; (B) EA assay; (C)MSA assay with (A1, B1, C1) aqueous extract, (A2, B2, C2) ethanolic extract and (A3, B3, C3) methanolic extract, demonstrating the percentage of inhibition at various concentrations with similar anti–inflammatory activity when the *Q. infectoria* extract and standards are compared.

methanolic extract is effective against *Klebsiella* species in comparison to other extracts. This shows that *Q. infectoria* exhibits antimicrobial properties against pathogens that cause UTIs.

Many herbal extracts do have antimicrobial actions against UTI pathogens. The fruit of *Vaccinium macrocarpon* (cranberry), *Juniperus* spp. (juniper) and *Arctostaphylos uva–ursi* (uva ursi) all release antimicrobial chemicals that can either directly kill microorganisms or inhibit their ability to adhere to epithelial cells, hence avoiding acute and persistent UTIs (27). Multiple research studies have proved that many of the herbal extracts can be used against UTI pathogens in this era of antibiotic resistance (28). One study that was conducted on *Q. infectoria* extract and decontamination of eggs shows that it has antimicrobial actions against *E. coli* and *C. albicans* (29, 30).

The cytotoxic effects of aqueous extract of *Q. infectoria* demonstrated a 90 % survival rate with 5 µL and 10 µL in comparison to ethanolic and methanolic extracts, which showed lower survival rates in this study, as demonstrated by the brine shrimp lethality assay (BSLA) method, which points to the safety of these extracts. In a similar study, cytotoxic effect using BSLA for their compounds, such as *Clitoria ternatea* L. flower and *Camellia sinensis* (L.) Kuntze leaf, was 95 % naupli survival when they used 5 µL and 10 µL of ethanolic extract (31). Use of the brine shrimp lethality assay is one of the methods to assess the toxicological effects of different herbs (32–34).

The antioxidant capacity of *Q. infectoria* (aqueous, ethanolic and methanolic extracts) was assessed using DPPH, H_2O_2, FRAP, NO and ABTS assays and was comparable with the standard compound. The concentration-dependent antioxidant property using DPPH expressed by *Q. infectoria* was comparable with the standard compound. In the present study, at the concentration of 50 µg/L, the methanol extract showed 89.13 % inhibition of antioxidant activity. In the H_2O_2 assay, at a concentration of 50 µg/L, the methanol extract showed 85.13 % inhibition of antioxidant activity. This result is similar to a previous study in which the researchers found the antioxidant activity of *Q. infectoria* using DPPH, FRAP and ABTS assays and concluded that *Q. infectoria* gall ethanolic extract reduces inflammation and oxidative damage in dermal fibroblasts to hasten wound healing (35). The DPPH method is used for the determination of antioxidant properties in the context of herbal preparations (19). Antioxidant property is an important characteristic of therapeutic agents that act against UTI pathogens (36). In another study, the researchers have shown that *Citrus macroptera* Montrouz. has a reservoir of natural antioxidants, thus enhancing the therapeutic benefits of the herbal formulation (37).

Another study found that the DMSO extract of *Acorus calamus* Linn. contains strong antioxidants, possibly as a result of the presence of additional antioxidants and phenolic components. As a medicinal agent, its antioxidant action is crucial (38). Galls of *Q. infectoria* have antioxidant properties and prevent functional changes in murine macrophages brought on by oxidative stress (39). Tannic acid present in the *Q. infectoria* is the main component contributing towards the antioxidant property of the plant (40). Previous studies show that the antioxidant activities of bioactive compounds can be related to their apoptotic effects (41). Similarly, certain other herbals also have antioxidant properties (42–44).

In the current study, while assessing the anti-inflammatory

activity of *Q. infectoria* using the BSA assay, the percentage of inhibition was 44, 56, 68, 74 and 79 at concentrations of 10–50 µg/mL for aqueous extract. On the other hand, these values were lowest for the methanolic extract. Similarly, the EA assay showed 48, 59, 64, 67 and 77 % inhibition for the same concentrations and the methanolic extract showed the lowest inhibition. In the MSA study, the percentage of inhibition was 43, 57, 66, 74 and 80 at concentrations of 10–50 µg/mL and again, it was the methanolic extract that had the lowest values of inhibition.

It is well known that the alcoholic extract of *Q. infectoria* has anti-inflammatory actions due to the presence of increased amounts of tannic acid that reduce the release of inflammatory mediators such as NO and prostaglandin (PGE2) (45). A previous anti-inflammatory study conducted on different herbs, including *Q. infectoria* and concluded that the anti-inflammatory effects of these herbs were significantly higher than those of standard steroids used and the COX–2 inhibitor (46, 47).

Because plant extracts contain phytochemicals, including alkaloids, terpenoids and flavonoids, they have anti-inflammatory qualities (48). Anti-inflammatory medications can be made with these substances and can be effective against UTI pathogens (49). In a previous study, the researchers conducted their experiment on a few bacteria and concluded that *Aloe vera* (L.) Burm.f. acts against them (50). In another study, researchers demonstrated that mouth paint made with titanium dioxide nanoparticles created with formulations of dried ginger and lemongrass had strong antibacterial properties as well as possible anti-inflammatory properties (51). Likewise, in another study, researchers concluded copper nanoparticles mediated by *Musa sapientum* L. had a moderate level of antimicrobial properties against certain important bacteria (52, 53). The above findings show the explicit role of herbals in antimicrobial treatment, especially against UTI pathogens.

Limitations

In this study, the biochemical compound responsible for the properties of *Q. infectoria* is not separated. On the other hand, the experiments were conducted *in vitro* only and hence, animal studies should be done in the future to prove the efficacy and safety of the extract. This study offers basic insights, but future research must overcome these limitations so that the medical application of *Q. infectoria* for UTI can be improved and thus have a therapeutic benefit.

Conclusion

This work is the first to report on the antimicrobial properties of the aqueous extract of *Q. infectoria* against UTI pathogens. *Escherichia coli* was found to have the highest ZOI for the aqueous extract. The findings of the experiments collectively highlight the pharmacological activity of *Q. infectoria* (aqueous, ethanolic and methanolic extracts) against *Candida albicans*, *Klebsiella species*, *E. coli* and *E. fecalis*, which are UTI pathogens. The cytotoxic studies using BSLA showed that the aqueous extract of *Q. infectoria* is much safer than the ethanolic and methanolic extracts of *Q. infectoria*. This knowledge can be used for future *in vivo* and human trials for the safety of drug development. Thus, strong antibacterial, anti-inflammatory and antioxidant properties were displayed by the extract, proving it to be an effective herbal drug against UTI. Additionally, the study shows dose-dependent effects, highlighting the significance of dosage optimisation for maximal performance. Higher concentrations of *Q. infectoria* offer more substantial therapeutic advantages. These results lay the scientific foundation for *Q. infectorias*' medicinal potential and support its traditional uses in complementary and alternative medicine. Further research should be conducted in this direction to isolate and identify the specific bioactive compound responsible for these effects. Future studies should be conducted *in vivo* and in clinical trials to develop antimicrobial drugs for UTIs using *Q. infectoria*.

To the best of our knowledge, this study is among the first to report the antimicrobial activity of the aqueous extract of *Q. infectoria* against UTI pathogens. *E. coli* exhibited the largest inhibition zone, followed by *Klebsiella* species and *E. faecalis*. Among all extracts, the aqueous form showed the most favourable balance of efficacy and safety in cytotoxic studies. The strong antibacterial, antioxidant and anti-inflammatory activities observed may be attributed to the phenolic and tannin constituents of *Q. infectoria*. These findings support its traditional use and provide a scientific foundation for developing safe, plant-based therapeutics for urinary tract infections. Further *in vivo* and clinical studies are recommended to isolate and characterise the active compounds responsible for these effects. These findings provide a scientific basis for developing standardised, plant-based formulations for UTI management.

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Authors' contributions

NJ, JPK and RS contributed to the study design, data analysis and conception. SG contributed to data collection and data analysis. PV, NP and SD contributed to the preparation and data analysis. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interests to declare.

Ethical issues: None

References

1. Elham A, Arken M, Kalimanjan G, Arkin A, Iminjan M. A review of the phytochemical, pharmacological, pharmacokinetic and toxicological evaluation of *Quercus infectoria* galls. J Ethnopharmacol. 2021;273:113592. https://doi.org/10.1016/j.jep.2020.113592
2. Basri DF, Fan SH. The potential of aqueous and acetone extracts of galls of *Quercus infectoria* as antibacterial agents. Indian J Pharmacol. 2005;37(1):26–9. https://doi.org/10.4103/0253-7613.13851
3. Taib M, Rezzak Y, Bouyazza L, Lyoussi B. Medicinal uses, phytochemistry and pharmacological activities of *Quercus* species. Evid Based Complement Alternat Med. 2020;2020:1920683. https://doi.org/10.1155/2020/1920683
4. Purbowati R, Taufikurohmah T, Syahrani A. Green extraction of *Quercus infectoria* gall with supercritical CO_2 and methanol co–

solvent. Environ Sci Pollut Res. 2023;30(55):116952–9. https://doi.org/10.1007/s11356-023-28047-1

5. Sykes JE, Westropp JL. Bacterial infections of the genitourinary tract. In: Canine and feline infectious diseases. 2013. p. 871. https://doi.org/10.1016/B978-1-4377-0795-3.00089-2

6. Phaniendra A, Jestadi DB, Periyasamy L. Free radicals: properties, sources, targets and their implication in various diseases. Indian J Clin Biochem. 2015;30(1):11–26. https://doi.org/10.1007/s12291-014-0446-0

7. Abdulrahman HA, Dyary HO, Mohammed RN, Hamad DS, Abdul–Star F, Saeed NM. Preventing free radical damage: The significance of including antioxidants in diet to strengthen immunity. Open Vet J. 2024;14(7):1526. https://doi.org/10.5455/OVJ.2024.v14.i7.2

8. Shao HB, Chu LY, Lu ZH, Kang CM. Primary antioxidant free radical scavenging and redox signaling pathways in higher plant cells. Int J Biol Sci. 2007;4(1):8. https://doi.org/10.7150/ijbs.4.8

9. Sharara A, Badran A, Hijazi A, Albahri G, Bechelany M, Mesmar JE, et al. Comprehensive review of *Cyclamen*: development, bioactive properties and therapeutic applications. Pharmaceuticals. 2024;17(7):848. https://doi.org/10.3390/ph17070848

10. Sarwar R, Farooq U, Khan A, Naz S, Khan S, Khan A, et al. Evaluation of antioxidant, free radical scavenging and antimicrobial activity of *Quercus incana* Roxb. Front Pharmacol. 2015;6:277. https://doi.org/10.3389/fphar.2015.00277

11. Iova OM, Marin GE, Lazar I, Stanescu I, Dogaru G, Nicula CA, et al. Nitric oxide/nitric oxide synthase system in the pathogenesis of neurodegenerative disorders—An overview. Antioxidants. 2023;12(3):753. https://doi.org/10.3390/antiox12030753

12. Moncada S, Higgs EA. Endogenous nitric oxide: physiology, pathology and clinical relevance. Eur J Clin Invest. 1991;21(4):361–74. https://doi.org/10.1111/j.1365-2362.1991.tb01383.x

13. Walthall L, Heincelman M. Pyometra: an atypical cause of abdominal pain. J Investig Med High Impact Case Rep. 2021;9:23247096211022481. https://doi.org/10.1177/23247096211022481

14. Mohaddese M. *Quercus infectoria* fruit hulls and galls and female genital disorders. Clin Phytosci. 2020;6(1)44. https://doi.org/10.1186/s40816–020–00194–9

15. Abdul–Aziz M, Mahdy MA, Abdul–Ghani R, Alhilali NA, Al–Mujahed LK, Alabsi SA, et al. Bacterial vaginosis, vulvovaginal candidiasis and trichomonal vaginitis among reproductive–aged women seeking primary healthcare in Sana'a city, Yemen. BMC Infect Dis. 2019;19(1):879. https://doi.org/10.1186/s12879-019-4549-3

16. Chusri S, Voravuthikunchai SP. *Quercus infectoria*: a candidate for the control of methicillin–resistant *Staphylococcus aureus* infections. Phytother Res. 2008;22(4):560–2. https://doi.org/10.1002/ptr.2377

17. Hanon NA, Abd FN. The antimicrobial activity of *Quercus infectoria* extracts against bacteria isolated from wounds infection. Al–Mustansiriyah J Sci. 2021;32(1):1–4. https://doi.org/10.23851/mjs.v32i1.919

18. Shanmugam R, Tharani M, Abullais SS, Patil SR, Karobari MI. Black seed assisted synthesis, characterization, free radical scavenging, antimicrobial and anti–inflammatory activity of iron oxide nanoparticles. BMC Complement Med Ther. 2024;24(1):241. https://doi.org/10.1186/s12906-024-04552-9

19. Wunnoo S, Sermwittayawong D, Praparatana R, Voravuthikunchai SP, Jakkawanpitak C. *Quercus infectoria* gall ethanolic extract accelerates wound healing through attenuating inflammation and oxidative injuries in skin fibroblasts. Antioxidants. 2024;13(9):1094. https://doi.org/10.3390/antiox13091094

20. Aziz ZA, Al Kaabi SJ. *Quercus infectoria* bark extract shows antibacterial activity against *Staphylococcus* species. Integr Biomed Res. 2024;8(4):1–6. https://doi.org/10.25163/angiotherapy.849542

21. Waghulde S, Kale MK, Patil V. Brine shrimp lethality assay of the aqueous and ethanolic extracts of selected medicinal plants. Proc. 2019;41(1):47. https://doi.org/10.3390/ecsoc-23-06703

22. Ali BM, Boothapandi M, Nasar AS. Nitric oxide, DPPH and hydrogen peroxide radical scavenging activity of TEMPO terminated polyurethane dendrimers. Data Brief. 2020;28:104972. https://doi.org/10.1016/j.dib.2019.104972

23. Pennisi R, Ben Amor I, Gargouri B, Attia H, Zaabi R, Chira AB, et al. Analysis of antioxidant and antiviral effects of olive (*Olea europaea* L.) leaf extracts using cancer cell model. Biomolecules. 2023;13(2):238. https://doi.org/10.3390/biom13020238

24. Nandita R, Jeevitha M, Gurumoorthy K, Shanmugham R, Ravichandran S, Raj K. Anti–inflammatory and free radical scavenging activity of *Tridax procumbens* leaves–based chitosan gel. J Pharm Bioallied Sci. 2024;16(Suppl 4):S4081–4. https://doi.org/10.4103/jpbs.jpbs_1439_24

25. Mahalingam M, Pillai DS, Shanmugam RK. Assessment of anti–inflammatory and cytotoxic property of herbal gel containing saffron, *Withania somnifera*, dry ginger and tulsi. J Indian Acad Oral Med Radiol. 2024;36(3):278–82. https://doi.org/10.4103/jiaomr.jiaomr_43_24

26. Yesmin S, Paul A, Naz T, Rahman AA, Akhter SF, Wahed MI, et al. Membrane stabilization as a mechanism of anti–inflammatory activity of ethanolic root extract of *Piper chaba*. Clin Phytosci. 2020;6(1):59. https://doi.org/10.1186/s40816-020-00207-7

27. Thiraviarajan A, Shanmugam R, Munusamy T. Preparation of *Andrographis paniculata* and *Rosa* formulation and its antibacterial activity against urinary tract infection pathogens. J Pharm Bioallied Sci. 2024;16(Suppl 2):S1360–4. https://doi.org/10.4103/jpbs.jpbs_574_23

28. Yarnell E. Botanical medicines for the urinary tract. World J Urol. 2002;20(5):285–93. https://doi.org/10.1007/s00345-002-0293-0

29. Sharma A, Verma R, Ramteke P. Antibacterial activity of medicinal plants used by tribals against UTI pathogens. World Appl Sci J. 2009;7(3):332–9.

30. Subash HA, Santhosh K, Kannan K, Pitchiah S. Extraction of keratin–degrading enzyme from marine actinobacteria of *Nocardia* sp. and antibacterial potential. Oral Oncol Rep. 2024;9:100184. https://doi.org/10.1016/j.oor.2024.100184

31. Tayel AA, El–Sedfy MA, Ibrahim AI, Moussa SH. Application of *Quercus infectoria* extract as a natural antimicrobial agent for chicken egg decontamination. Rev Argent Microbiol. 2018;50(4):391–7. https://doi.org/10.1016/j.ram.2017.12.003

32. Surya KG, Shanmugam R, Behera A, Jothinathan MK. Antibacterial properties and cytotoxic effect of *Clitoria ternatea* and *Camellia sinensis* formulation. Cureus. 2024;16(4):e58287. https://doi.org/10.7759/cureus.58287

33. Subramanian AK, Prabhakar R, Vikram NR, Dinesh SS, Rajeshkumar S. In vitro anti–inflammatory activity of silymarin/hydroxyapatite/chitosan nanocomposites. J Popul Ther Clin Pharmacol. 2022;28(2):e71–7. https://doi.org/10.47750/jptcp.2022.874

34. Abitha ST, Rajeshkumar S, Lakshmi T, Roy A. Cytotoxic effects of silver nanoparticles synthesized using *Phyllanthus emblica* seed. Drug Invent Today. 2019;11(9):229–35. https://doi.org/10.22377/ijgp.v13i3.2593

35. Anushya P, Geetha RV, Rajeshkumar S. Evaluation of anti–inflammatory and cytotoxic effect of copper nanoparticles synthesized using *Mucuna pruriens* seed extract. J Pharma Res Intl. 2021;33(47B):816–24. https://doi.org/10.9734/jpri/2021/v33i47B33188

36. Baliyan S, Mukherjee R, Priyadarshini A, Vibhuti A, Gupta A, Pandey RP, et al. Determination of antioxidants and phytochemical analysis of *Ficus religiosa*. Molecules. 2022;27(4):1326. https://doi.org/10.3390/molecules27041326

37. Amani T, Surenthar M, Shanmugam R. Anti–inflammatory and antioxidant activity of *Cucumis sativus* and *Citrus macroptera* formulation. Cureus. 2024;16(1). https://doi.org/10.7759/cureus.51818

38. Haran P, Shanmugam R, Deenadayalan P. Free radical scavenging, anti–inflammatory and antibacterial activity of *Acorus calamus* against *Pseudomonas aeruginosa* and *Staphylococcus aureus*. Cureus. 2024;16(3):55987. https://doi.org/10.7759/cureus.55987

39. Kaur G, Athar M, Alam MS. *Quercus infectoria* galls possess antioxidant activity and abrogate oxidative stress–induced alterations in murine macrophages. Chem Biol Interact. 2008;171(3):272–82. https://doi.org/10.1016/j.cbi.2007.10.002

40. Arina MI, Harisun Y. Effect of extraction temperature on tannin content and antioxidant activity of *Quercus infectoria*. Biocatal Agric Biotechnol. 2019;19:101104. https://doi.org/10.1016/j.bcab.2019.101104

41. Rajeshkumar S, Parameswari RP, Jayapriya J, Tharani M, Ali H, Aljarba NH, et al. Apoptotic and antioxidant activity of gold nanoparticles synthesized using marine brown seaweed. Biomed Res Int. 2022;2022:5746761. https://doi.org/10.1155/2022/5746761

42. Keerthiga N, Anitha R, Rajeshkumar S, Lakshmi T. Antioxidant activity of cumin oil mediated silver nanoparticles. Pharmacogn J. 2019;11(4). https://doi.org/10.5530/pj.2019.11.125

43. Mohapatra S, Leelavathi L, Rajeshkumar S, Sakthi DS, Jayashri P. Cytotoxicity, anti–inflammatory and antioxidant activity of zinc oxide nanoparticles synthesized using clove and cinnamon. J Evol Med Dent Sci. 2020;9(25):1859–65. https://doi.org/10.14260/jemds/2020/405

44. Shahidi F, Zhong Y. Measurement of antioxidant activity. J Funct Foods. 2015;18:757–81. https://doi.org/10.1016/j.jff.2015.01.047

45. Kaur G, Hamid H, Ali A, Alam MS, Athar M. Anti–inflammatory evaluation of alcoholic extract of *Quercus infectoria* galls. J Ethnopharmacol. 2004;90(2–3):285–92. https://doi.org/10.1016/j.jep.2003.10.009

46. Aroonrerk N, Kamkaen N. Anti–inflammatory activity of *Quercus infectoria*, *Glycyrrhiza uralensis*, *Kaempferia galanga* and *Coptis chinensis*. J Health Res. 2009;23(1):17–22.

47. Rajkumar M, Presley SD, Govindaraj P, Girigoswami K, Meenambigai K, Deepak P, et al. Biomedical prowess of *Clitoria mariana* (L.) flower methanolic extract. Chem Biodivers. 2025; e01051.

48. Chirumamilla P, Vankudoth S, Dharavath SB, Dasari R, Taduri S. In vitro anti–inflammatory activity of silver nanoparticles and *Solanum khasianum* leaf extract. Proc Natl Acad Sci India B Biol Sci. 2022;92(2):301–7. https://doi.org/10.1007/s40011-021-01337-9

49. Gonfa YH, Tessema FB, Bachheti A, Rai N, Tadesse MG, Singab AN, et al. Anti–inflammatory activity of phytochemicals and their nanoparticles. Curr Res Biotechnol. 2023;6:100152. https://doi.org/10.1016/j.crbiot.2023.100152

50. Rajeshkumar S, Tharani M, Jeevitha M, Santhoshkumar J. Anticariogenic activity of *Aloe vera* gel mediated copper oxide nanoparticles. Indian J Public Health Res Dev. 2019;10(11)3664. https://doi.org/10.5958/0976-5506.2019.04158.5

51. Rifaath M, Rajeshkumar S, Anandan J, Munuswamy T, Govindharaj S, Shanmugam R, et al. Preparation of herbal nano–formulation–assisted mouth paint using titanium dioxide nanoparticles and its biomedical applications. Cureus 15(11):e48332. https://doi.org/10.7759/cureus.48332

52. Yazhlini P, Ganesh SB, Rajeshkumar S. Antimicrobial activity of *Musa sapientum* mediated copper nanoparticles. J Pharm Res Int. 2021;33(61B):426–35. https://doi.org/10.9734/jpri/2021/v33i61B35702

53. Krishnaswamy J, Christupaul Roseline P, Kannan K, Dhanraj G, Sivaperumal P. Biosynthesis and anticoagulant properties of copper nanoparticles from *Acanthophora* sp. Phytochem Anal. 2025;36(8):2228–35. https://doi.org/10.1002/pca.3384

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