

In Vitro Antioxidant and Antibacterial Evaluation of Optimized Dragon Fruit and Kiwi Fruit Herbal Syrup

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Abstract:

Background: Fruit peels are sources of phytoconstituents including polyphenols and flavonoids. Dragon fruit and kiwi fruit peel extracts were incorporated into an optimized herbal syrup to utilize their biological potential. Objective: The study evaluated the in vitro antioxidant and antibacterial activities of an optimized syrup containing ethanolic and aqueous peel extracts of dragon fruit and kiwi fruit. Methods: The optimized syrup contained a total peel-extract load of 5.0% w/v. Antioxidant activity was evaluated using the DPPH radical-scavenging assay at syrup concentrations of 10-50 mg/mL, with ascorbic acid at 20-100 µg/mL as standard. Antibacterial activity was evaluated by agar well diffusion against *Staphylococcus aureus* and *Escherichia coli* using ciprofloxacin (5 µg/mL) as positive standard. Results: DPPH scavenging increased from 22.56% at 10 mg/mL to 85.51% at 50 mg/mL. The calculated IC₅₀ values were 25.18 mg/mL for the syrup and 37.92 µg/mL for ascorbic acid. The syrup produced zones of inhibition of 22.4 ± 0.5 mm against *S. aureus* and 18.6 ± 0.8 mm against *E. coli*. Net zones were 16.4 and 12.6 mm, and relative antibacterial efficacies were 73.87% and 61.46%, respectively. Conclusion: The optimized formulation demonstrated concentration-dependent DPPH radical-scavenging activity and antibacterial activity against both test organisms, with greater antibacterial activity against *S. aureus*.

Keywords: Dragon fruit; Kiwi fruit; Fruit peel; Herbal syrup; DPPH; Antioxidant activity; Antibacterial activity; *Staphylococcus aureus*; *Escherichia coli*; IC₅₀.

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1. INTRODUCTION

Plant-derived materials are important sources of phytochemical constituents, including polyphenols, flavonoids and pigments with potential biological activity. Fruit-processing residues, particularly peels, may retain substantial amounts of these constituents and can therefore be considered for value-added herbal formulations.

Dragon fruit (*Hylocereus polyrhizus*) contains polyphenols, flavonoids and betacyanins, while its peel has been recognized in the source research as a phenolic-rich botanical material [2-4]. Kiwi fruit (*Actinidia chinensis* var. *deliciosa*) also contains biologically relevant constituents, and studies cited in the source document describe its antioxidant and related biological properties [6-9].

The optimized formulation used in this study incorporated both ethanolic and aqueous peel extracts of dragon fruit and kiwi fruit. It contained a total peel-extract load of 5.0% w/v and had previously been optimized to provide suitable physical properties. The biological component was designed to determine whether the optimized syrup retained measurable antioxidant and antibacterial activity.

2. LITERATURE REVIEW

Paško et al. [4] described dragon fruit as a reservoir of natural polyphenolics with chemopreventive properties, while Jadhav and Jadhav [3] reviewed its pharmacognostic and phytochemical relevance. Nur et al. [10] reported in vitro antioxidant, anti-inflammatory, antibacterial, and cytotoxic effects of colorants extracted from dragon fruit, supporting biological investigation of dragon-fruit-derived materials.

For kiwi fruit, Lee et al. [6] investigated antioxidant and glycation-inhibitory activities of gold kiwifruit. Brevik et al. [7] evaluated golden kiwifruit supplementation in relation to biomarkers of oxidative damage and antioxidant protection, while Hunter et al. [8] discussed gold kiwifruit for immune support. Khutare and Deshmukh [9] reviewed medicinal applications of kiwi fruit.

The biological testing of herbal syrups. Ugwoke et al. [21] evaluated a bi-herbal syrup against selected respiratory tract pathogens, while Akharaiyi and Maliki [22] investigated inhibition of bacterial growth by a syrup prepared from *Vitex doniana* fruits. These studies provided a basis for evaluating antibacterial activity of the optimized fruit-peel syrup.

Table 1. Summary of Literature Relevant to the Antioxidant and Antibacterial Evaluation of Dragon Fruit and Kiwi Fruit Herbal Syrup

Reference	Plant/Formulation	Main focus	Relevance
Paško et al. [4]	Dragon fruit	Natural polyphenolics	Supports phytochemical basis
Lee et al. [6]	Gold kiwifruit	Antioxidant activity	Supports antioxidant evaluation
Brevik et al. [7]	Golden kiwifruit	Oxidative damage/antioxidant protection	Supports antioxidant background

Nur et al. [10]	Dragon fruit	Antioxidant and antibacterial activity	Supports biological testing
Ugwoke et al. [21]	Bi-herbal syrup	Antibacterial screening	Supports syrup antibacterial evaluation
Akharaiyi & Maliki [22]	Vitex doniana fruit syrup	Bacterial growth inhibition	Supports fruit-syrup antimicrobial evaluation

3. AIM AND OBJECTIVES

The aim was to evaluate the in vitro antioxidant and antibacterial potential of the optimized dragon fruit and kiwi fruit peel herbal syrup. The objectives were to determine DPPH free-radical scavenging activity, calculate the IC₅₀ and compare it with ascorbic acid, determine antibacterial activity against *S. aureus* and *E. coli*, calculate net zone of inhibition and relative antibacterial efficacy against ciprofloxacin, and compare the responses of the Gram-positive and Gram-negative test organisms.

4. MATERIALS AND METHODS

Table 2. Composition of Optimized Dragon Fruit and Kiwi Fruit Herbal Syrup

Ingredient	Quantity/100 mL	Role
Dragon Fruit Peel (Ethanolic Extract)	1.5 g	Active ingredient
Kiwi Fruit Peel (Ethanolic Extract)	1.5 g	Active ingredient
Dragon Fruit Peel (Aqueous Extract)	1.0 g	Active ingredient
Kiwi Fruit Peel (Aqueous Extract)	1.0 g	Active ingredient
Sodium CMC (Na-CMC)	0.32 g	Suspending agent
Sucrose	66.62 g	Vehicle/sweetener
Methylparaben	0.1 g	Preservative
Propylparaben	0.02 g	Preservative
Purified Water	q.s. to 100 mL	Solvent

The total fruit peel extract load in the optimized formulation was 5.0% w/v.

In Vitro Antioxidant Activity: DPPH Assay

A 0.1 mM DPPH reagent was prepared by dissolving exactly 3.94 mg of DPPH in 100 mL of analytical-grade methanol. The flask was protected from light with aluminium foil and stored at 4°C until use. An ascorbic acid stock solution of 1000 µg/mL was prepared in methanol and diluted to 20, 40, 60, 80 and 100 µg/mL. The optimized syrup was diluted in methanol to 10, 20, 30, 40 and 50 mg/mL.

For the assay, 1.0 mL of each syrup dilution or ascorbic acid standard was placed in separate foil-wrapped test tubes and 3.0 mL of 0.1 mM DPPH solution was added. A negative control contained 1.0 mL methanol and 3.0 mL DPPH solution. The tubes were incubated in the dark at 27°C for 30 minutes. Absorbance was measured at 517 nm using a UV-visible spectrophotometer, with methanol as blank.

DPPH scavenging (%) = $[(\text{Abscontrol} - \text{Abssample}) / \text{Abscontrol}] \times 100$. A concentration-versus-percentage-scavenging plot was generated, the regression equation $Y = mX + c$ was obtained, and IC₅₀ was calculated as $\text{IC}_{50} = (50 - c) / m$.

In Vitro Antibacterial Activity: Agar Well Diffusion

The antibacterial potential was evaluated against *Staphylococcus aureus* and *Escherichia coli*. Nutrient Agar was used as the medium, ciprofloxacin (5 µg/mL) as positive standard, and sterile distilled water as negative control.

Nutrient Agar (28 g) was suspended in 1000 mL distilled water and sterilized at 121°C and 15 psi for 15 minutes. Twenty millilitres of sterilized agar was poured into Petri dishes. After solidification, plates were lawn-inoculated with bacterial suspension. Four wells of 6 mm diameter were bored with a sterile cork borer. A 100 µL quantity of optimized syrup was added to the test wells and positive and negative controls were added to their respective wells. Plates were left for 1 hour at room temperature for pre-diffusion and then incubated at 37°C for 24 hours.

Net ZOI (mm) = Total diameter of clear zone - 6 mm. Relative antibacterial efficacy (RAE) was calculated as: $RAE (\%) = (\text{Net ZOI of optimized syrup} / \text{Net ZOI of standard antibiotic}) \times 100$. Mean ZOI was calculated from three independent replicates ($n = 3$).

5. RESULTS

Table 3. DPPH Radical-Scavenging Activity of Optimized Dragon Fruit and Kiwi Fruit Herbal Syrup and Ascorbic Acid

Concentration	Syrup Absorbance (Mean ± SD)	Scavenging (%)	Ascorbic Acid Absorbance (Mean ± SD)	Scavenging (%)
Control (0)	0.842 ± 0.004	0.00	0.842 ± 0.004	0.00
10 mg/mL syrup / 20 µg/mL AA	0.652 ± 0.012	22.56	0.582 ± 0.008	30.88
20 mg/mL syrup / 40 µg/mL AA	0.488 ± 0.009	42.04	0.395 ± 0.011	53.09
30 mg/mL syrup / 60 µg/mL AA	0.315 ± 0.015	62.59	0.224 ± 0.005	73.39
40 mg/mL syrup / 80 µg/mL AA	0.204 ± 0.007	75.77	0.112 ± 0.009	86.69
50 mg/mL syrup / 100 µg/mL AA	0.122 ± 0.010	85.51	0.065 ± 0.003	92.28

The optimized syrup demonstrated a concentration-dependent increase in DPPH scavenging from 22.56% at 10 mg/mL to 85.51% at 50 mg/mL. Ascorbic acid increased from 30.88% at 20 µg/mL to 92.28% at 100 µg/mL.

Table 4. IC₅₀ Values of Optimized Dragon Fruit and Kiwi Fruit Herbal Syrup and Ascorbic Acid

Sample	IC ₅₀
Optimized Dragon Fruit-Kiwi Fruit Syrup	25.18 mg/mL
Ascorbic Acid	37.92 µg/mL

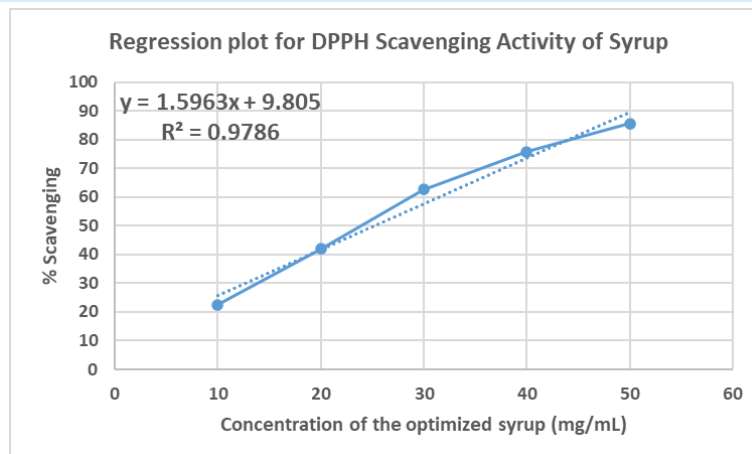


Figure 1. Regression Plot for DPPH Radical-Scavenging Activity of Optimized Dragon Fruit and Kiwi Fruit Herbal Syrup

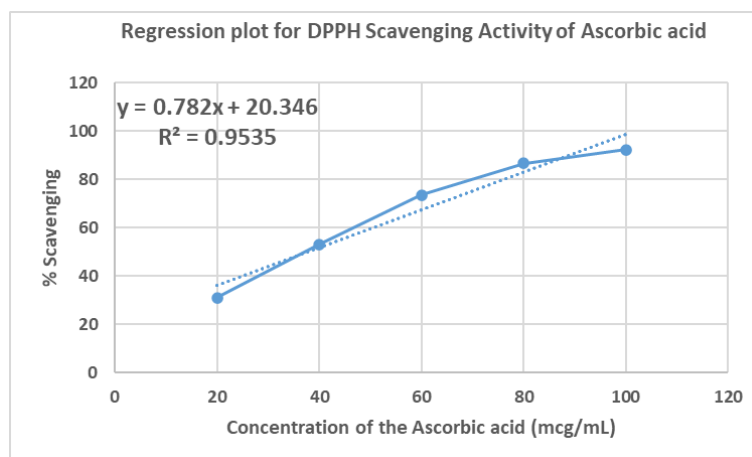


Figure 2. Regression Plot for DPPH Radical-Scavenging Activity of Ascorbic Acid

Table 5. Zone of Inhibition (ZOI) and Relative Antibacterial Efficacy (RAE) of Optimized Dragon Fruit and Kiwi Fruit Herbal Syrup Against *Staphylococcus aureus* and *Escherichia coli*

Test Organism	ZOI of Optimized Syrup (mm)	Net ZOI (mm)	ZOI of Ciprofloxacin (mm)	Net ZOI Standard (mm)	RAE (%)	Susceptibility
S. aureus	22.4 ± 0.5	16.4	28.2 ± 0.3	22.2	73.87	Susceptible
E. coli	18.6 ± 0.8	12.6	26.5 ± 0.4	20.5	61.46	Intermediate

The optimized syrup inhibited both test organisms. Greater activity was observed against *S. aureus*, with a net ZOI of 16.4 mm and RAE of 73.87%, compared with a net ZOI of 12.6 mm and RAE of 61.46% against *E. coli*.

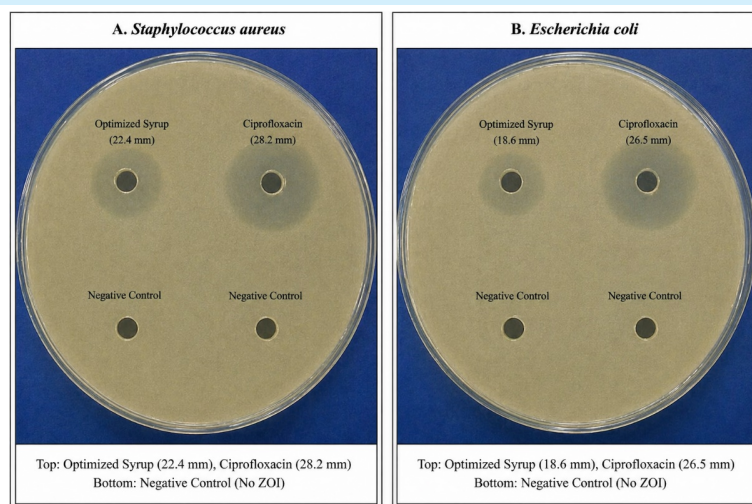


Figure 3. Agar Well Diffusion Assay Showing Antibacterial Activity of Optimized Dragon Fruit and Kiwi Fruit Herbal Syrup Against *Staphylococcus aureus* and *Escherichia coli*

6. DISCUSSION

Antioxidant Potential and IC₅₀ Analysis

The DPPH assay demonstrated a clear dose-dependent radical-scavenging response. As the syrup concentration increased from 10 to 50 mg/mL, inhibition increased from 22.56% to 85.51%. The calculated IC₅₀ of the optimized syrup was 25.18 mg/mL. Ascorbic acid showed an IC₅₀ of 37.92 µg/mL. The pure ascorbic acid standard showed greater potency on a common concentration basis, while the complex herbal syrup nevertheless demonstrated substantial concentration-dependent radical-scavenging activity.

The antioxidant activity of the 5% total peel-extract load and the phenolic and flavonoid constituents of dragon fruit and kiwi fruit peels. The progressive reduction in syrup absorbance from 0.652 ± 0.012 at 10 mg/mL to 0.122 ± 0.010 at 50 mg/mL corresponded with increasing radical scavenging.

Antibacterial Spectrum and Efficacy

The optimized syrup showed antibacterial activity against both Gram-positive *S. aureus* and Gram-negative *E. coli*. The total ZOI was 22.4 ± 0.5 mm against *S. aureus* and 18.6 ± 0.8 mm against *E. coli*. After correction for the 6-mm well diameter, net ZOI values were 16.4 and 12.6 mm, respectively.

Relative antibacterial efficacy also showed greater activity against *S. aureus*. The syrup achieved 73.87% of the net inhibitory activity of ciprofloxacin against *S. aureus* and 61.46% against *E. coli*. The source study associated the greater response of *S. aureus* with differences between Gram-positive and Gram-negative bacterial envelope structures and with the potential interaction of fruit-peel polyphenolic constituents with bacterial cells.

7. CONCLUSION

The optimized dragon fruit and kiwi fruit peel herbal syrup exhibited in vitro antioxidant and antibacterial activities. DPPH radical-scavenging activity increased from 22.56% at 10 mg/mL to 85.51% at 50 mg/mL, and the calculated IC₅₀ was 25.18 mg/mL. The formulation inhibited both bacterial test organisms, producing net zones of inhibition of 16.4 mm against *S. aureus* and 12.6 mm against *E. coli*, with relative antibacterial efficacies of 73.87% and 61.46%,

respectively. The results supported the biological potential of the optimized 5.0% w/v fruit-peel extract formulation, with greater antibacterial activity against *S. aureus* than *E. coli*.

8. LIMITATIONS

The biological evaluation was limited to the DPPH radical-scavenging assay and agar well diffusion against two bacterial species.

9. ACKNOWLEDGEMENT

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10. CONFLICT OF INTEREST

The authors declare no conflicts of interest related to the publication of this article.

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