

Development of Resveratrol-Quercetin Face Serum: A Dual Defence against Aging and Acne

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ABSTRACT

Objectives: This study centers on the development and assessment of a facial serum fortified with the innate antioxidant's resveratrol and quercetin, to address skin aging and acne. **Materials and Methods:** The serum was carefully formulated by combining oils and ethanol, resulting in a stable biphasic emulsion with a smooth and uniform texture and a slightly acidic pH of 5.3, making it ideal for everyday application. The serum's small particle size at the nanoscale and quick absorption greatly improves its capacity to effectively target and treat fine lines, wrinkles, and acne. **Results:** The antimicrobial tests showed notable efficacy against *Staphylococcus aureus* and *Pseudomonas sp.*, indicating that the serum has the potential to be a skincare product with dual functions. Additionally, the serum was shown to have potent antioxidant properties as demonstrated by the DPPH and H₂O₂ scavenging tests. These tests confirmed that the serum provides robust protection against oxidative stress, a key contributor to skin aging. **Conclusion:** Although the antioxidant activity of resveratrol and quercetin is slightly lower than that of ascorbic acid, their combined action effectively protects against reactive oxygen species, supporting the serum's ability to preserve youthful and healthy skin. The findings indicate that the RQ face serum has potential as a versatile skincare product, effectively targeting both signs of aging and acne, while also protecting against oxidative damage to the skin.

Keywords: Acne treatment, Anti-aging, Oxidative stress, Quercetin, Resveratrol, Skincare.

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INTRODUCTION

The condition and appearance of one's skin play a crucial role in how individuals perceive their overall well-being and health. Human skin undergoes natural ageing due to internal variables such as hormones, cellular metabolism, heredity, and metabolic processes. There are also exogenous or extrinsic factors that contribute to its development. These include exposure to light, pollution, ionizing radiation, chemicals, and poisons. Both intrinsic and extrinsic factors contribute to progressive alterations in the layers of the skin, as well as cumulative modifications in its structure and physiology. A little subcutaneous deposit that resembles acne is caused by fat. Acne is caused by the blockage of oil glands, which serve as a source of nourishment for bacteria, specifically *Propionibacterium acne*. This leads to irritation and the eventual formation of acne (Nam *et al.*, 2003; Phosri *et al.*, 2022; Gromkowska-Kępką *et al.*, 2021; Prabhu Venkatesh *et al.*, 2023). When it comes to mild acne, topical therapy is the primary

treatment necessary; however, when it comes to moderate and severe acne, systemic medicine is required in addition to topical therapy (de Canha *et al.*, 2020). Topical medication can have negative effects, and certain important drugs, such as benzoyl peroxide and tretinoin, have physical and chemical characteristics that may make it difficult for patients to follow the therapy for acne and alter its effectiveness. In addition to acne, premature aging is also an issue for women's complexion and attention span (Jin and Lee, 2018). Some spermatophytes, such as grapevines, produce resveratrol (3, 4', 5 trihydroxy stilbene) as a natural response to injury. White wine contains significantly lower levels of resveratrol compared to red wine due to its absence in the flesh of grape berries, where it is mostly located, and its presence in the skin (Bhat *et al.*, 2001). A variety of factors determine the quantities of glucosides and aglycone in the form of trans- and cis-isomers. The trans-isomer is the most prevalent type found in red wine, with quantities ranging from 0.1 to 15 mg/L (Oh and Shahidi, 2018). Resveratrol, a phenolic compound, enhances the antioxidant capacity of red wine and has the potential to mitigate cardiovascular diseases in individuals. Studies have shown that resveratrol effectively controls lipid metabolism and inhibits oxidation (Skoza *et al.*, 2015). Quercetin, which is also referred to as 3,3',4',5,7-pentahydroxyflavone, is a member of a diverse



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group of flavonoid compounds that are primarily found in plants and plant-derived foods (Baghel *et al.*, 2012). Quercetin is commonly present in the form of glycosides or sugar derivatives. An example of this is rutin, where a disaccharide replaces the hydrogen atom in the R4 hydroxyl position. Quercetin is a compound that is referred to be the aglycone or sugarless form of rutin (Rashid *et al.*, 2019). The majority of the biological and therapeutic knowledge about quercetin was presented in two extensive publications resulting from significant conferences on plant flavonoids in 1985 and 1987 (Brandi, 1992).

Quercetin and resveratrol, two potent polyphenols, exhibit promising synergistic effects in combating acne and signs of aging. Their antibacterial properties specifically target bacteria that cause acne, while their combined anti-inflammatory properties help to reduce inflammation, which is a significant contributor to the development of acne (Kleinedler *et al.*, 2011). Additionally, they may assist in regulating sebum production, which, when excessive, can exacerbate acne. Resveratrol and quercetin are potent antioxidants that protect the epidermis from free radicals and oxidative stress, which accelerate the ageing process (Mikstacka *et al.*, 2010). In addition to fortifying the skin barrier and stimulating the production of collagen, which is crucial for maintaining skin firmness and elasticity, they also improve the overall resilience and well-being of the skin. Supplementing skincare products or diets with resveratrol and quercetin may enhance their efficacy in treating acne and age-related skin issues (Riya *et al.*, 2019).

This study work focuses on the production of a face serum containing natural antioxidants, namely quercetin and resveratrol. These compounds possess potent anti-aging and anti-acne properties, promoting the maintenance of healthy skin.

MATERIALS AND METHODS

The RQ face serum was formulated using high-purity resveratrol and quercetin from Yucca Enterprises, Mumbai. The oils used in the formulation were of cosmetic grade, sourced from Saveetha College of Pharmacy, Chennai.

Formulation of Face Serum

The designated amount of resveratrol and quercetin was quantified and subsequently dissolved in ethanol. To create a consistent solution, a blend was created by blending oils such as olive oil, coconut oil, and almond oil (Savic *et al.*, 2021). The oil phase was gradually added in small amounts to the liquid phase to form a biphasic emulsion. The mixture is ground using a mortar and pestle until a distinct clicking sound is heard. The serum that has been created is assessed for its efficacy in combating acne and reducing signs of aging (Amnuaitik *et al.*, 2019). The ingredients used in the preparation of RQ Face serum were tabulated in Table 1.

Evaluation of Face Serum

Physical Assessment

The color, odor, taste and appearance of the formulation were assessed, revealing a uniform distribution of the extraction throughout the formulation step. Touch and look were used as confirmation for this examination (Sasidharan *et al.*, 2014; Sao and Pounikar, 2021).

pH determination

The pH meter was calibrated using a standardized buffer solution. The pH of the mixture was determined by accurately measuring and dissolving approximately 1 mL of the facial serum in 50 mL of distilled water. The pH of the skin falls within an acidic range; hence the skin serum should have a pH level ranging from 4.1 to 6.7 (Ashwini *et al.*, 2023).

Determination of Spreadability

Spread ability is the measure of how effortlessly the serum can be distributed across the skin or affected areas. After applying cosmetic items, they should be evenly distributed across the skin without any restrictions. Spread ability is influenced by various elements, including viscosity and temperature. The duration of spreading should be quite brief (Thorat *et al.*, 2023).

Stability studies

An extensive stability analysis is necessary to assess the physical and chemical stability and the safety of a pharmaceutical product's formulation and development. Stability investigations adhere to the ICH criteria. An expedited evaluation of the stability of the formulated product was conducted over a brief period of several months. The samples were maintained under three distinct storage conditions: 3-5°C, 25°C with 60% relative humidity, and 40°C with 2% relative humidity (Stability Testing of New Drug Substances and Products, 2003).

Globule size determination

The serum was analyzed using a microscope to ascertain the size of the globules. A small amount of serum was mixed with water, placed onto a glass slide, covered with a glass cover, and observed using a microscope (Surber and Kottner, 2017; Zasada and Budzisz, 2019; Waghaye *et al.*, 2018).

Rheological studies

The Brookfield Viscometer, namely the spindle-type model S64, was employed to measure the viscosity of the formulation at a rotational speed of 100 rpm. After immersing the spindle in 5 mL of the serum in a beaker for approximately 5 min (Yeskar *et al.*, 2023).

Pharmacological Activity

Anti-Microbial Activity

The antimicrobial activities of quercetin and resveratrol face serum were evaluated using the agar well diffusion method. The Mueller-Hinton agar plates underwent sterilization using autoclaving at a temperature of 121°C for a period of 15 to 20 min after their preparation. Following the sterilizing process, the medium was transferred onto a sterile Petri dish and allowed to cool down to the ambient temperature. The bacterial suspension containing *S. aureus* and *Pseudomonas* sp. was uniformly spread across the agar plates using sterile cotton brushes (Kunjan *et al.*, 2024). A sterile polystyrene tip was used to partition the agar plates into wells with a diameter of 9 mm. The wells were subsequently filled with varying concentrations of face serum (25 µg/mL, 50 µg/mL, and 100 µg/mL). The plates were incubated with bacteria for 24 hr. at a temperature of 37°C. The diameter of the inhibitory zone surrounding the wells was measured to evaluate the antibacterial activity. Once the width of the zone of inhibition in Millimeters (mm) was established, the computation of the zone of inhibition was completed (Tharani *et al.*, 2023).

Antioxidant Activity

DPPH Assay

A stock solution of DPPH in methanol was prepared, at a concentration of 0.1 mM. Each experiment consisted of creating a new solution by mixing the current solution with methanol until it reached a final concentration of 20 M (Amnuakit *et al.*, 2022). A 96-well plate was used to combine 200 µL of DPPH working solution with different amounts of facial serum (10-50 µg/mL). The plate was left in a dark, ambient temperature environment for 30 min without disturbance (Bondet *et al.*, 1997). A microplate reader was used to measure the absorbance at 517 nm. The subsequent equation was utilized to determine the fraction of DPPH-scavenging activity:

The DPPH scavenging activity can be calculated using the formula:

$$= [(A_{\text{control}} - A_{\text{sample}}) / A_{\text{control}}] \times 100$$

A control refers to the measurement of absorbance in the control group, which is composed of a DPPH solution without the

sample. On the other hand, A sample refers to the absorbance measurement of the experimental group, which contains a DPPH solution mixed with face serum. Methanol served as the solvent. The positive control group consisted of a solution containing a concentration of 1 mg/mL of ascorbic acid (Üstün *et al.*, 2022).

H₂O₂ Assay

The antioxidant activity was evaluated in this experiment using the H₂O₂ assay. A reaction mixture of 1 mL (1 mL) was created by adding 100 microliters (µL) of 28 millimolar (mM) 2-deoxy-2-ribose. Varying doses of (10-50 µg/mL) were introduced (Chatterjee, 2023). Furthermore, 200 µL of Ethylenediaminetetraacetic Acid (EDTA), 200 µL of ascorbic acid, and 200 µL of 200 µM ferric chloride were included. The optical density at a wavelength of 532 nm was measured after incubating at a temperature of 37°C for one hr, using the blank solution. Vitamin E was used as the positive control.

The hydroxyl radical scavenging activity can be calculated using the formula:

$$= [(A_{\text{blank}} - A_{\text{sample}}) / A_{\text{blank}}] \times 100$$

A blank sample's absorbance is denoted by A blank, A sample's absorbance by A sample. A blank is the absorbance of the control reaction in the absence of the sample; whereas A sample is the absorbance of the reaction with the sample (Fernando and Soysa, 2015).

RESULTS

In this study, a comprehensive analysis of the face serum's physical, chemical and mechanical properties was conducted to evaluate its potential applications. The key parameters assessed include color, odor, taste, texture, pH, viscosity, spread ability, stability and particle size. The results, as summarized in Table 2, provide valuable insights into the sample's characteristics and their implications for delivering active principles effectively to the skin.

The RQ serum has a uniform texture and smooth consistency, which allows for even application. Its mildly acidic, pH of 5.3 matches the natural environment of the skin, making it suitable for daily use. With a viscosity of 1050 cps, the serum has a medium-thick consistency that is easy to apply without leaving

Table 1: Formulation of RQ face serum.

Sl. No.	Ingredients	Standard Formula	Working Formula
1	Resveratrol and Quercetin	50%	10 mL
2	Coconut oil	2%	0.4 mL
3	Olive oil	9%	1.8 mL
4	Almond oil	0.1%	0.02 mL
5	Glycerine	25%	5 mL
6	Ethanol	Q. S	q. s

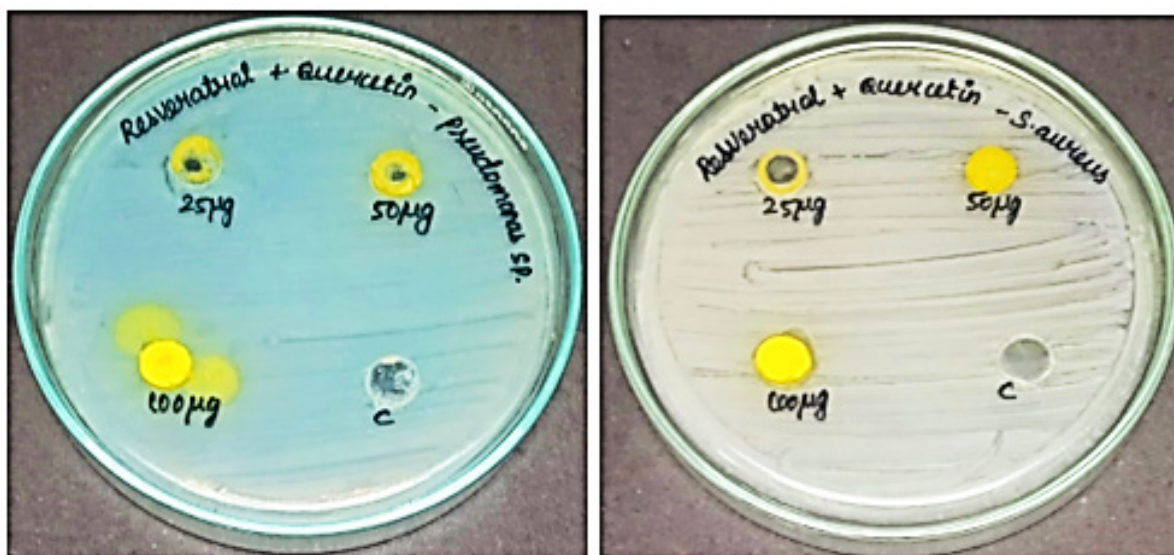


Figure 1: The agar well plate represents the anti-microbial activity.

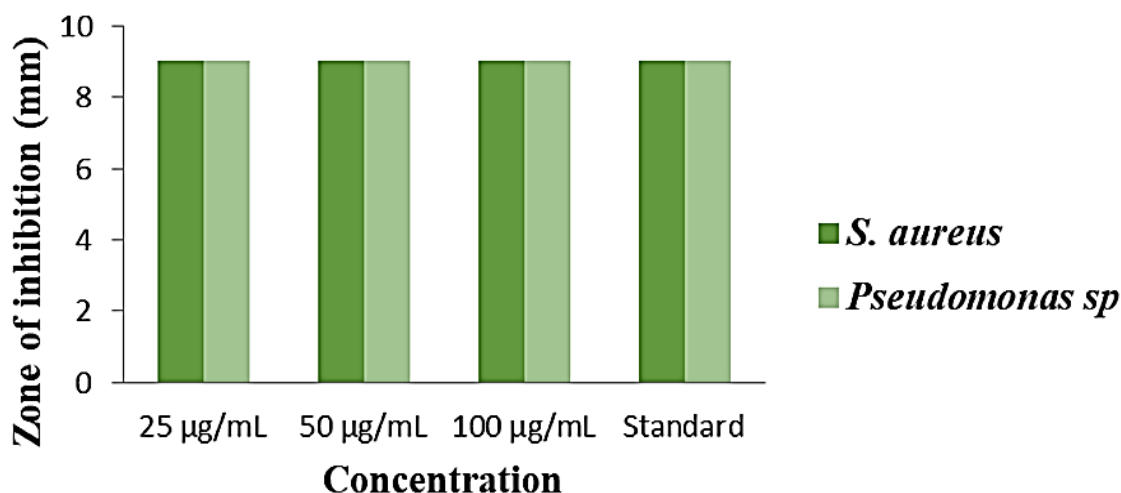


Figure 2: The graph represents the anti-microbial activity of RQ face serum.

a greasy residue. Its rapid absorption is particularly beneficial for targeting fine lines, wrinkles, and acne, as it ensures that active ingredients penetrate the skin quickly and effectively. The stable formulation and nanoscale particle size (ranging between 0.1 and 0.3 µm) further enhance the serum's anti-aging and anti-acne efficacy by facilitating deeper skin penetration and more targeted action. These findings highlight the serum's potential as an effective treatment for aging and acne-prone skin.

Antimicrobial Study

The face serum's antimicrobial activity was assessed by evaluating the zone of inhibition against 2 prevalent skin pathogens: *Staphylococcus aureus* and *Pseudomonas sp.* The findings, depicted in Figures 1 and 2, indicate that the serum displays a noteworthy inhibitory impact on both bacterial strains.

The serum, at concentrations of 25, 50, and 100 µg/mL, consistently resulted in inhibition zones of around 9 mm for both *S. aureus* and *Pseudomonas sp.*, which were like those produced by the standard antimicrobial agent employed as a control. The serum's consistent antimicrobial impact at all tested concentrations demonstrates its broad-spectrum activity, which is crucial for its dual function as an anti-aging and anti-acne treatment. The serum's capacity to suppress the growth of *S. aureus* is very crucial in the treatment of acne, as this bacterium is frequently linked to acne breakouts. Moreover, the suppression of *Pseudomonas sp.* a bacterium frequently associated with skin infections-emphasizes the serum's capacity to safeguard the skin against a wider array of microbiological dangers. The findings indicate that the face serum not only tackles ageing concerns but also offers efficient protection against microbial infections, positioning it as a viable multi-functional skincare solution.

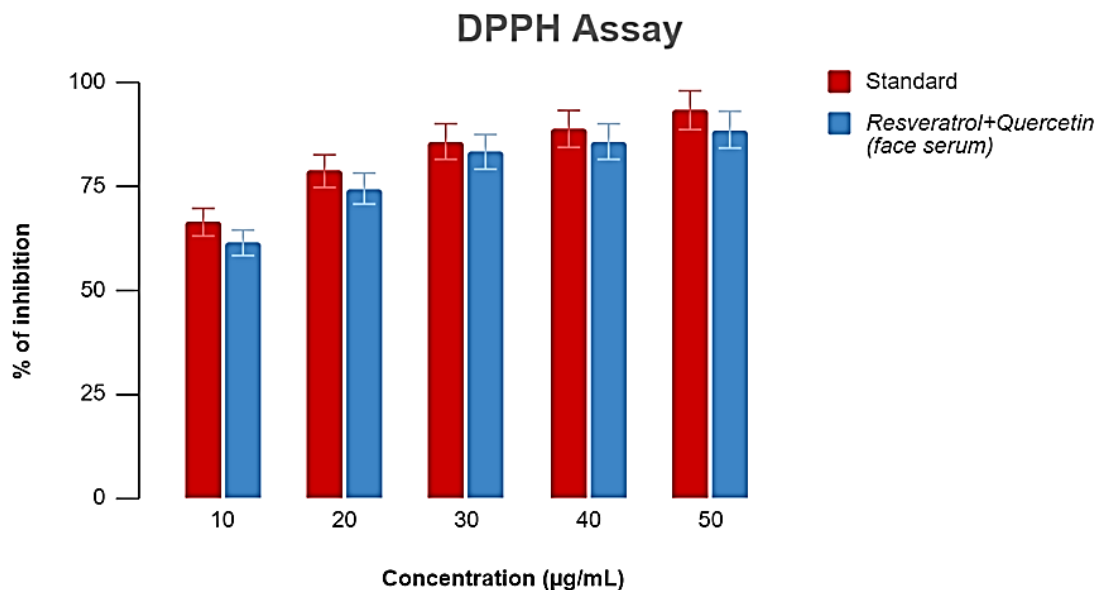


Figure 3: Graphical representation of the free radical scavenging activity of RQ face serum.

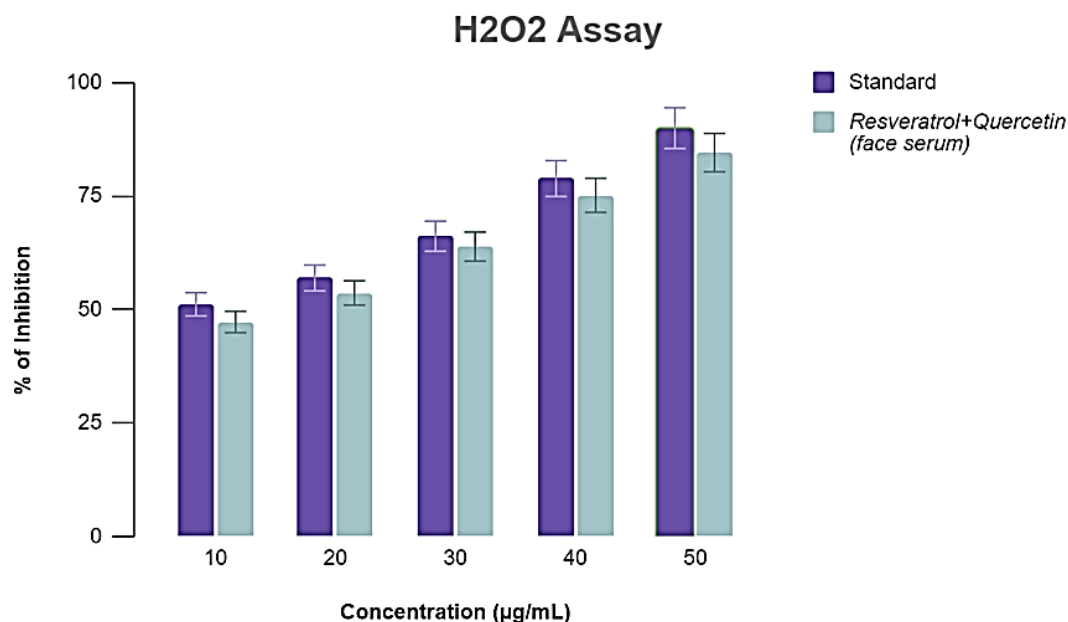


Figure 4: Graphical representation of the free radical scavenging activity of RQ face serum.

Antioxidant Activity

The antioxidant efficacy of the facial serum, which contains resveratrol and quercetin, was evaluated using the DPPH and H₂O₂ assay, a recognized technique for measuring the ability to neutralize free radicals. The findings, depicted in Figure 3, demonstrate that the serum has significant antioxidant activity at various doses, which is essential for its anti-aging characteristics.

In the DPPH assay, the face serum contains five different concentrations, ranging from 10-50 µg/mL, which were included. The different concentrations of face serum, which are 10 µg/mL,

20 µg/mL, 30 µg/mL, 40 µg/mL, and 50 µg/mL, correspond to inhibitions of 61.29%, 74.34%, 83.15%, 85.62%, and 88.48% respectively. The percentage of inhibition increased with the concentration of the serum, reaching up to approximately 85% inhibition at 50 µg/mL. Although the inhibition levels of the serum were slightly lower than those of ascorbic acid, the standard used in this assay, the serum still displayed robust antioxidant activity.

Figure 4 summarizes the results, which show that the face serum exhibits significant H₂O₂ scavenging activity at the tested concentrations. Here, the face serum contains five different

Table 2: Evaluation of RQ Face serum.

Sl. No.	Characteristics	Observation
1	Color	Yellow
2	Odor	Aromatic
3	Taste	Bitter
4	Texture	Smooth
5	Homogeneity	Homogenous
6	Ph	5.3
7	Spread ability	Absorbed rapidly
8	Stability	No variations
9	Particle size	0.1 and 0.3 µm
10	Viscosity	1050 cps.

concentrations, ranging from 10-50 µg/mL, which were included. The different amounts of face serum, namely 10 µg/mL, 20 µg/mL, 30 µg/mL, 40 µg/mL, and 50 µg/mL, correspond to inhibitions of 47.2%, 53.6%, 63.8%, 75.1%, and 84.5% respectively. With an increase in the concentration of the face serum, its capacity to remove hydrogen peroxide improved, reaching a peak scavenging activity of around 80% at the highest concentration tested (50 µg/mL). Despite the serum's scavenging activity being slightly lower than that of ascorbic acid, the standard employed in the study, it nevertheless demonstrated a robust ability to counteract hydrogen peroxide.

The face serum's robust radical-scavenging and antioxidant actions, which effectively neutralize free radicals and alleviate oxidative stress, can be attributed to the presence of resveratrol and quercetin. This is significant as oxidative stress is a crucial contributor to skin aging. These data indicate that the serum provides robust defence against oxidative damage, amplifying its anti-aging advantages and demonstrating similar performance to ascorbic acid. The synergistic effect of resveratrol and quercetin offers a strong protection against reactive oxygen species, rendering the serum a helpful remedy for maintaining youthful and healthy skin.

CONCLUSION

Ultimately, the research emphasizes the creation and assessment of a facial serum that incorporates the powerful natural antioxidants resveratrol and quercetin, which are recognized for their abilities to combat aging and acne. The serum exhibited a consistent texture, smooth composition, and a slightly acidic pH that is appropriate for everyday application to the skin. The quick absorption, stable formulation, and small particle size at the nanoscale improve its efficacy in specifically targeting fine lines, wrinkles, and acne. The serum's antibacterial efficacy against *Staphylococcus aureus* and *Pseudomonas* sp. highlights its promise as a dual-purpose skincare product, successfully addressing both acne and microbial infections.

In addition, the serum had notable antioxidant activity, as demonstrated by its capacity to eliminate free radicals in both DPPH and H₂O₂ tests. While the serum's antioxidant efficacy was significantly inferior to that of ascorbic acid, it still offered significant defense against oxidative stress, a crucial contributor to skin aging. The synergistic impact of resveratrol and quercetin provides a strong protection against reactive oxygen species, making the serum a highly effective treatment for preserving youthful and healthy skin. The findings indicate that the RQ face serum shows great potential as a versatile skincare product, effectively targeting both signs of ageing and acne, while also providing protection against oxidative damage to the skin.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

RQ: Resveratrol and quercetin; **Pseudomonas sp.:** *Pseudomonas* species; **DPPH:** 1,1-diphenyl-2-picrylhydrazyl; **H₂O₂:** Hydrogen peroxide; **ICH:** International Council for Harmonization.

AUTHORS CONTRIBUTION

Abida HariPriya and Dharshini wrote the manuscript, Udhaya Kumar contributed to the research work, and Vandhana and Thirumalaikumaran reviewed and corrected the manuscript. Every author oversees the content and similarity index of the document and has carefully examined and approved the last draft.

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