REVIEW ARTICLE

A Review on Phytochemical Analysis and *In-vitro* Antioxidant Activity of Curcumin from *Curcuma longa* L. Rhizomes

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ABSTRACT

This study focused on investigating the phytochemical composition and evaluating the antioxidant capability of an extract obtained from the rhizomes of *Curcuma longa* (turmeric). Preliminary screening tests revealed the presence of various bioactive compounds like alkaloids, flavonoids, phenolic substances, saponins, terpenoids, cardiac glycosides, and fixed oils/fatty acids in the methanolic rhizome extract. The antioxidant potential of the extract was assessed using the widely employed 1,1-diphenyl-2-picryl-hydrazyl (DPPH) free radical scavenging assay. The findings indicated a concentration-dependent enhancement in radical scavenging activity, reaching a maximum of around 81% at the highest concentration evaluated (3 mL). The remarkable antioxidant capacity can be attributed to the unique structural features of curcumin, the principal bioactive compound present, which contains methoxy, phenoxy, and carbon-carbon double bonds that enable efficient neutralization of free radicals and reactive oxygen species. Furthermore, curcumin has been reported to modulate the activity of transcription factors like Nrf2, thereby upregulating the expression of antioxidant enzymes and bolstering the body's defense against oxidative stress. These findings offer empirical support for the historical use of *C. longa* and underscore the diverse potential uses of curcumin as a natural antioxidant across different sectors, including food, pharmaceuticals, and cosmetics.

Keywords: *Curcuma longa*, Curcumin, Phytochemicals, Antioxidant, DPPH, Free radical scavenging.

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INTRODUCTION

Curcuma longa, commonly referred to as turmeric, is an herbaceous plant species belonging to the Zingiberaceae family.[1] It is extensively cultivated across tropical and subtropical regions, particularly in India, where it has been employed as a spice and traditional medicine for centuries.[2] The rhizomes (underground stems) of this plant serve as the primary source of curcumin, a polyphenolic compound responsible for the characteristic yellow color. Curcumin, chemically termed diferuloylmethane, comprises curcumin, demethoxycurcumin, and bisdemethoxycurcumin (Figure 1) as its major curcuminoid constituents.[3] These compounds derived from curcumin display a broad spectrum of biological functions, encompassing scavenging, anti-inflammatory, antibiotic, and antineoplastic properties, attributable to the unique chemical structure featuring two aromatic rings connected by a seven-carbon chain.[4-6]

Oxidative stress, marked by an unequal ratio between reactive oxygen species (ROS) and the body's antioxidant mechanisms, is pivotal in the onset of numerous chronic ailments like cancer, cardiovascular issues, and neurodegenerative disorders.[7] Extensive research has focused on curcumin due to its potent antioxidant properties linked to its ability to neutralize free radicals, induce antioxidant enzyme activation, and inhibit ROS production.[8]

The unique chemical makeup of curcumin, featuring methoxy, phenoxy, and carbon-carbon double bonds, enhances its antioxidant potency by facilitating the transfer of hydrogen

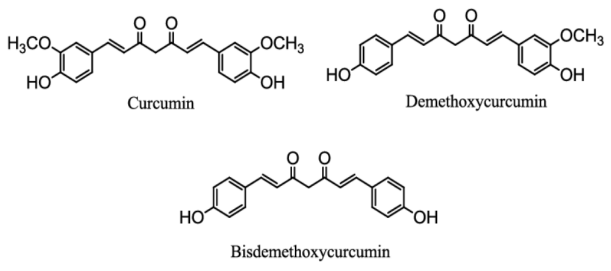

Figure 1: Chemical constituents of *Curcuma longa*. L rhizomes

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atoms and electrons, thereby counteracting free radicals and safeguarding cellular structures from oxidative stress.[9]

This work aimed to conduct a comprehensive phytochemical analysis of the methanolic extract obtained from *C. longa* rhizomes to identify various bioactive compounds, assess the antioxidant potential of the extract through *in-vitro* testing using the DPPH free radical scavenging assay, and explore the potential applications of curcumin as a natural antioxidant source across different industries.[10]

MATERIALS AND METHODS

Materials

Turmeric rhizomes, sourced from a local market in Tirupati, Andhra Pradesh, India, served as the raw material. Methanol of analytical grade was sourced from Merck Life Science Private Limited, Mumbai. DPPH was acquired from Merck Laboratories, Bengaluru. Necessary chemicals, including hydrochloric acid, sodium hydroxide, concentrated sulfuric acid, and glacial acetic acid, were procured from Molychem Chemicals Pvt. Ltd., Bengaluru. Wagner's reagent and Fehling's solutions A and B were prepared in-house following standard protocols. Ferric chloride solution and chloroform were purchased from Molychem, Bengaluru.

Methods

Preparation of the extract

Fresh *C. longa* rhizomes were procured from a local market, cleaned, sliced into small pieces, and dried under sunlight. The dried rhizome pieces were ground into a fine powder using a grinder. For extraction, 1 g of the powdered sample was mixed with 25 mL of methanol and placed in a shaking water bath at room temperature for 2.5 hours. After extraction, it was centrifuged at 6000 rpm for fifteen minutes to obtain the solid residue. The supernatant liquid was carefully decanted and filtered through Whatman filter paper to obtain a clear methanolic extract.[11]

Phytochemical screening

The methanolic rhizome extract underwent various qualitative tests to identify the presence of different phytochemical constituents according to standard procedures. Specific tests included Wagner's test for alkaloids, ferric chloride test for phenolic compounds and tannins, lead acetate test for tannins, Salkowski's test for terpenoids, foam test for saponins, ferric chloride test for flavonoids, glacial acetic acid test for cardiac glycosides, and spot test for fixed oils and fatty acids. The observations were recorded to determine the phytochemical composition.[12]

DPPH free radical scavenging assay

The methanolic solution of DPPH (0.1 mM) was shielded from light. The extract was then diluted with methanol to yield various concentrations (1, 2, and 3 mL). For each concentration, 3 mL of the DPPH solution was added to the diluted extract, and the total volume was adjusted to 10 mL using methanol. The reaction mixtures were incubated in darkness at room temperature for 30 minutes. Following incubation, the absorbance was recorded at 517 nm using a UV-visible spectrophotometer. Additionally, a control solution containing only DPPH was prepared.[13] The percentage of DPPH radical scavenging activity (%RSA) was subsequently determined using the formula:

%RSA = [(Absorbance of control - Absorbance of sample)/ Absorbance of control] × 100.

Table 1: Results of the preliminary phytochemical analysis

Phytochemical	*Observation*	*Inference*
Alkaloids	Reddish-brown precipitate	Present
Tannins and phenols	Green color	Present
Terpenoids	Reddish-brown color	Present
Saponins	Stable froth/foam	Present
Flavonoids	Blackish-red color	Present
Cardiac glycosides	Brown ring	Present
Fixed oils and fatty acids	Translucent greasy stain	Present

Table 2: Radical scavenging activity of curcumin

Concentration (mL)	*Control absorbance*	*Sample absorbance*	*%RSA*
1	0.52	0.278	46.53
2	0.52	0.197	62.11
3	0.52	0.943	81.34

RESULTS

Phytochemical Analysis

The phytochemical screening tests showed the presence of alkaloids, tannins, phenols, terpenoids, saponins, flavonoids, cardiac glycosides, and fixed oils/fatty acids in the methanolic rhizome extract.[14,15] The results of the qualitative tests are presented in Table 1.

Antioxidant Activity (DPPH Assay)

The findings demonstrated a concentration-dependent increase in radical scavenging activity. At 1-mL concentration, the %RSA was moderate. However, as the concentration increased to 2 and 3 mL, a significant enhancement in radical scavenging ability was observed, reaching around 81% at the highest concentration of 3 mL. The results of this assay, expressed as the percentage of radical scavenging activity (%RSA), are presented in Table 2.

DISCUSSION

The phytochemical screening of the *C. longa* rhizome extract showed the presence of several bioactive compounds, including alkaloids, tannins, phenolic compounds, terpenoids, saponins, flavonoids, cardiac glycosides, and fixed oils/fatty acids.[16] This remarkable phytochemical diversity contributes to the multitude of pharmacological activities and therapeutic potential associated with turmeric.[17]

Alkaloids, known for their antimicrobial, analgesic, and antioxidant properties, may play a crucial role in the traditional use of turmeric for treating various infectious diseases

and inflammatory conditions. The presence of tannins and phenolic compounds, which exhibit potent antioxidant, anti-inflammatory, and antimutagenic effects, may contribute to the chemopreventive and anticancer properties attributed to turmeric.[18]

The terpenoid class, encompassing the curcuminoids (curcumin, demethoxycurcumin, and bisdemethoxycurcumin), has garnered significant attention for its anticancer, anti-inflammatory, and neuroprotective activities.[19] These bioactive compounds may be responsible for the observed therapeutic effects of turmeric in various chronic diseases, including cancer, inflammatory disorders, and neurodegenerative conditions.[20]

Saponins, known for their ability to enhance immune function and exhibit hypocholesterolemic effects, may contribute to the traditional use of turmeric in promoting overall health and well-being.[21] Flavonoids, another class of polyphenolic compounds present in the extract, possess remarkable antioxidant, anti-inflammatory, and anticancer properties, further enhancing the therapeutic potential of turmeric.[22]

The diverse phytochemical composition of *C. longa* rhizomes provides a strong scientific rationale for its widespread traditional use in various therapeutic applications and highlights the potential for further exploration and development of novel therapeutic agents derived from this plant source.[23] The DPPH free radical scavenging assay is a widely accepted and reliable method for evaluating the antioxidant capacity of various compounds. In this study, the *C. longa* rhizome extract exhibited a concentration-dependent increase in radical scavenging activity, reaching a maximum of around 81% at the highest concentration tested (3 mL). This potent antioxidant activity can be attributed to the unique chemical structure of curcumin, the principal bioactive compound present in the extract.

Curcumin's remarkable antioxidant potential is attributed to its ability to donate hydrogen atoms and electrons efficiently, thereby neutralizing free radicals and reactive oxygen species (ROS). The presence of methoxy, phenoxy, and carbon-carbon double bonds in the curcumin molecule facilitates this process, enabling the formation of relatively stable phenoxy radicals and preventing further oxidative damage to cellular components.[24]

Additionally, curcumin has been reported to chelate metal ions, which can catalyze the formation of free radicals through the Fenton reaction. By sequestering these metal ions, curcumin prevents the generation of harmful free radicals, further contributing to its overall antioxidant activity.

Moreover, curcumin exhibits indirect antioxidant mechanisms by modulating the activity of transcription factors like nuclear factor erythroid 2-related factor 2 (Nrf2). Nrf2 regulates the expression of various antioxidant enzymes, including superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx). By activating the Nrf2 signaling pathway, curcumin can enhance the levels of these enzymes, thereby strengthening the body's endogenous antioxidant defense system against oxidative stress.

The potent antioxidant potential exhibited by the *C. longa* rhizome extract in this study highlights its potential applications across various industries. In the food industry, curcumin can be explored as a natural antioxidant additive to extend the shelf life of food products by preventing oxidative deterioration, rancidity, and discoloration. Additionally, its antioxidant properties may contribute to preserving the nutritional value and sensory qualities of food products during storage and processing.

In the pharmaceutical and nutraceutical industries, curcumin can be incorporated into formulations as an antioxidant and chemopreventive agent, owing to its ability to neutralize free radicals and protect against oxidative stress-related diseases. Curcumin's anti-inflammatory and anticancer properties, combined with its antioxidant activity, make it a promising candidate for the development of novel therapeutic agents targeting chronic diseases associated with inflammation and oxidative stress, such as cancer, cardiovascular disorders, and neurodegenerative diseases.

Furthermore, the antioxidant properties of curcumin may find applications in the cosmetic industry, where it can be utilized as a natural ingredient in skincare products to combat oxidative stress, promote skin health, and potentially mitigate the effects of aging. The ability of curcumin to neutralize free radicals and protect cellular components from oxidative damage may contribute to its potential use in anti-aging and skin-rejuvenating formulations.

It is important to note that while the findings of this study provide valuable insights into the antioxidant potential of *C. longa* rhizomes, further research is warranted to explore the bioavailability, pharmacokinetics, and long-term safety of curcumin and its derivatives. Additionally, clinical studies are necessary to validate the therapeutic efficacy and potential applications of curcumin in various disease contexts.

Overall, this study demonstrates the remarkable phytochemical diversity and antioxidant capacity of *C. longa* rhizomes, providing scientific validation for its traditional use and highlighting its potential as a valuable source of natural antioxidants and therapeutic agents in various industries.

CONCLUSION

This study investigated the phytochemical constituents and antioxidant potential of curcumin extracted from the rhizomes of *C. longa* (turmeric). The phytochemical screening revealed the existence of various phytochemical compounds, including alkaloids, phenolic compounds, flavonoids, terpenoids, saponins, cardiac glycosides, and fixed oils/fatty acids. These phytochemicals contribute to the diverse pharmacological activities associated with turmeric. The antioxidant activity assessment using the DPPH free radical scavenging assay demonstrated a concentration-dependent increase in radical scavenging ability, with curcumin exhibiting potent antioxidant potential. The unique chemical structure of curcumin, featuring methoxy, phenoxy, and carbon-carbon double bonds, enables it to neutralize free radicals and reactive oxygen species effectively. Additionally, curcumin can modulate the activity

of transcription factors like Nrf2, enhancing the expression of antioxidant enzymes and reinforcing the body's defense against oxidative stress.

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