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Phenolic and Flavonoid Content and Antioxidant Activity, of Three Different Extracts of *Tecoma stans* (L.) Kunth and *Zingiber officinales* Roscoe

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Aim: The purpose of this study was to determine the total phenolic and flavonoid content and antioxidant activity in the three medicinal plants *Tecoma stans* and *Zingiber officinales*.
Study Design: Plant extracts were prepared in three different solvents, total phenolic content, total flavonoid content and the antioxidant activity was determined.
Place and Duration of Study: The study was conducted at Banasthali during August-December 2022.
Methods: Aqueous, methanolic, and ethanolic solvent extracts were prepared; total phenolic and flavonoid contents was determined, the DPPH assay was used to measure the extracts' antioxidant activity.
Results: The methanolic extract contained the highest total phenolic (16.67±0.04, 11.56±0.09 mg GAE/g dw) and flavonoid content (14.21±0.10, 7.96±0.06 mg QUE/g,) as well as displayed the

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highest antioxidant activity (68.91%, 53.09% at 300 µg/ml) in *Z.officinales* and *T. stans* extracts respectively.
Conclusion: *Tecoma stans* and *Zingiber officinales* can be used as possible antioxidant agents as well as resource for the production of novel drugs.

Keywords: Antioxidant activity; DPPH assay; total phenolic content; total flavonoid content; Tecoma stans; Zingiber officinales.

1. INTRODUCTION

Plants and natural products constitute the foundation of both contemporary and traditional medicines [1]. Medicinal plants are particularly significant in facilitating the development of alternative remedies that do not have the negative side effects of synthetic pharmaceuticals [2,3]. Herbs are used in approximately 25% of prescription medications globally [4,5].

Ginger (*Zingiber officinale* Roscoe) is a member of the Zingiberaceae family and contains a variety of active compounds including terpenes (sesquiterpene hydrocarbons), alkaloids, and polyphenols [6].

Phenolic compounds are linked to a wide range of biological activities, one of which is their antioxidant ability [7], which may aid in protecting cells from oxidative damage induced by free radicals [8,9]. Studies have reported that ginger has antioxidant properties [10,11]. From ancient times, ginger has been used as a medicine to cure a variety of conditions, including fever, indigestion, hypertension, sprains, sore throats, cramps, and rheumatism [12]. Ginger has a variety of biological properties, including antibacterial activity, antifungal activity, anti-inflammatory activity, and anticoagulant impact [13,14].

A decorative shrub from the *Bignoniaceae* family, is *Tecoma stans* (L.) Kunth also known as yellow elder, has been used in traditional medicine. Aerial parts of the plant are used for the treatment of diabetes, to control fungal infections, and as a tonic, diuretic and dewormer [15]. Teas made from flowers and leaves are commonly used to treat diabetes and digestive problems, and the decoction of flowers has been used to relieve stomach pain [16,17]. The presence of several bioactive compounds in species of *Tecoma* genus have been reported, which include saponins, flavonoids, alkaloids, phenols, steroids, anthraquinones, tannins, terpenes, hydrocarbons, volatile oils, glycosylated flavonoids and phenolic acids [17,18]. These compounds have been shown antioxidant, antibacterial and antifungal activities [19].

These plants have key bio-natural components that are quite effective in giving critical medical advantages. The present study aims to determine the total phenolic and flavonoid content and antioxidant activity in the medicinal plants *Tecoma stans* and *Zingiber officinales.*

2. MATERIALS AND METHODS

The selected medicinal plants/ parts were collected as follows: leaves of *Tacoma stans* were obtained from the plants growing at Banasthali Vidyapith campus, and rhizome of *Zingiber officinale* were procured from the market and kept in a refrigerator at 4ºC.

2.1 Extraction of Leaf Samples

20g of leaf sample was dissolved in 100 ml of solvent (ethanol, methanol, and water) for 24–42 h at room temperature. Then, the extracts were filtered through sterile muslin cloth and Whatman filter paper. The filtrates from each solvent were evaporated in a desiccator. The dried extracts were packaged in airtight containers, labelled, and kept in a refrigerator (2-4°C) for further use. For performing the experiments, the extracts were reconstituted in 5% Dimethyl Sulphoxide (DMSO) to obtain concentrations of 100µg/ml, 150 µg/ml, 200 µg/ml, 250 µg/ml and 300 µg/ml.

2.2 Total Phenolic and Total Flavonoid Content

The determine the total phenolic content (TPC), aliquots of 1ml of plant extract (0.5g/20ml) were placed in test tubes, mixed with 4 ml of Folin-Ciocalteau reagent, and after 5-7 min, 5 ml of sodium carbonate (20%) was added and mixed thoroughly [20,21]. The mixture was incubated for 30 mins at room temperature in complete darkness. A spectrophotometer was used to measure absorbance at 740nm. Gallic acid was used as a calibration curve standard and TPC concentration was expressed as gallic acid equivalent (GAE).

To determine total flavonoid content (TFC), aliquots of 1ml of plant extract (0.5g/20ml) were placed in test tubes, mixed with 4 ml of distilled water and subsequently with 0.30ml of $NaNO_2$ solution (10%) [22,23]. After 5 min, 0.30ml $AlCl_3$ solution (10%) was added followed by 2.0ml of NaOH solution (1%) to the mixture. Immediately the mixture was thoroughly mixed and absorbance was then determined at 510 nm versus blank. Standard curve of quercetin was prepared (0-210 µg/ml) and the results were expressed as quercetin equivalents (mg QUE/ g dried extract).

2.3 Determination of Antioxidant Activity

To determine DPPH (1, 1 dihydroxy 2-picrylhydrazyl, Sigma Aldrich) free radical scavenging activity, 1.0ml of DPPH solution (0.135mM DPPH in methanol) was added to (100-300µg/ml) plant extract [19]. The reaction mixture was kept in the dark for 30 min and its absorbance at 517nm was measured with a spectrophotometer. Ascorbic acid was used as a reference.

2.4 Statistical Analysis

The experiments were repeated three times, and the findings were provided as Mean± S.E. All tests were designed using a randomised block-design and ANOVA at $p<0.05$. Appropriate post-hoc tests were performed, including the "Duncan Multiple Range Test" (DMRT) and the "Least Significant Difference" (LSD).

3. RESULTS AND DISCUSSION

3.1 Total Phenolic and Flavonoid Contents

Table 1 shows the amounts of phenolic and flavonoid content in several medicinal plants. Among these three solvents, MetOH extraction yielded the most phenolic acid in all three extracts, but Aq. solvent extraction yielded the least phenolic content. TPC (16.67, 17.78 and 10.59 mg GAE/g extract respectively) and TFC (14.21, 12.75 and 8.10 mg QE/g dw extract respectively) are abundant in *Z. officinales* extracts in MetOH, EtOH, and Aq. Solvent.

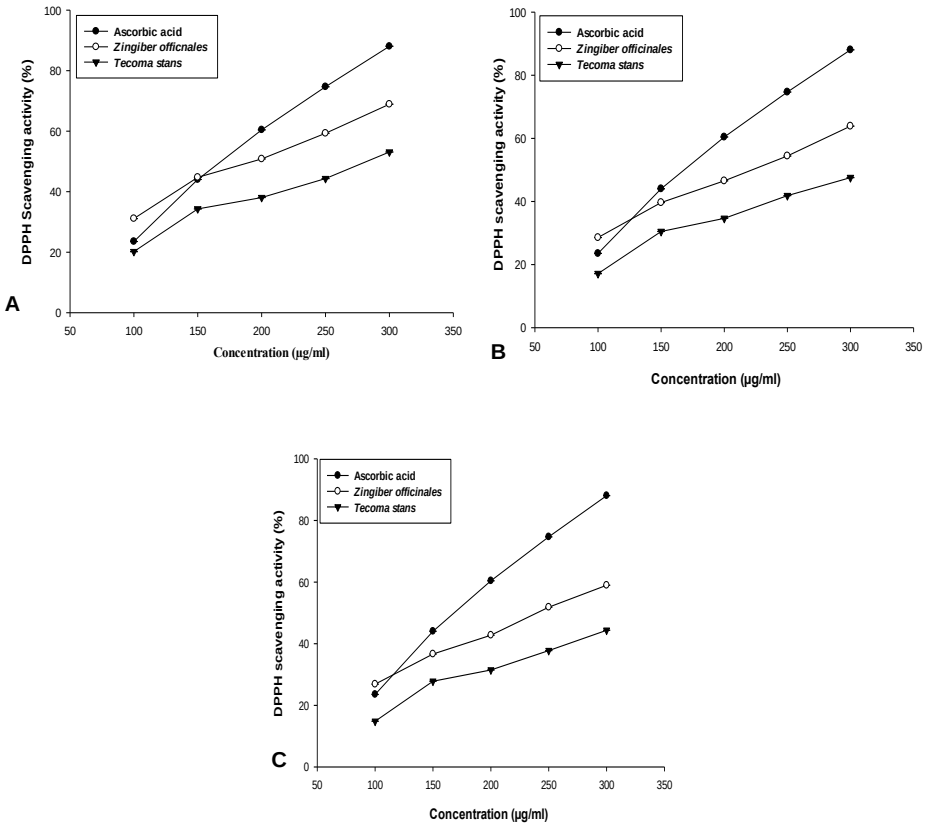

Fig. 1. Total antioxidant activity of *Z. officinales* and *T. stans* in different solvents (A) methanolic (B) ethanolic (C) aqueous

Table 1. Total phenolic and flavonoid content of plant extracts

Plants/ Extract	Total Phenolic Content (mg GAE/g dw)			Total Flavonoid Content (mg QUE/g dw)		
	MetOH	**EtOH**	**Aq.**	**MetOH**	**EtOH**	**Aq.**
TS	11.56±0.09^{c}	9.73±0.07^{b}	4.30±0.14^{a}	7.96±0.06^{c}	6.20±0.11^{b}	3.66±0.06^{a}
ZO	16.67±0.04^{c}	12.78±0.06^{b}	10.59±0.07^{a}	14.21±0.10^{c}	12.75±0.13^{b}	8.10±0.10^{a}

Results are mean ± standard deviation (SD), values followed by the same letter for a plant and a particular metabolite separately are not significant at *p=0.05* following ANOVA and LSD; EtOH= Ethanolic extract, MetOH= Methanolic extract, Aq.= Aqueous extract; *T. stans*=TS; *Z. officinales*= ZO.

3.2 Total Antioxidant Activity

The efficacy of three distinct solvent extracts to scavenge DPPH free radicals was tested and compared to the standard, ascorbic acid. Fig. 1 shows the antioxidant properties of medicinal plant extracts as assessed by DPPH scavenging experiments. The present study's results demonstrated that *Z. officinales* sample DPPH• scavenging activity considerably increased in methanol solvent. However, the extracts' DPPH radical scavenging capacities were lower than those of ascorbic acid (82.29%) at 300µg/ml. This result clearly shows that the extracts have proton donating potential and might be used as free radical inhibitors or scavengers, possibly functioning as primary anti-oxidants.

In the current investigation, high TPC and TFC values, as well as antioxidant activity, were found in methanolic solvent extract. Plant phenolic compounds are potent antioxidants and antimicrobials [24,25]. Several investigations have shown that flavonoids in plant extracts have antioxidant and antifungal properties [26,27].

The presence of phenolic and flavonoid compounds in these plants may enhance their biological capabilities when compared to other therapeutic plants evaluated. The antioxidant capabilities of phenolic and flavonoids compounds are determined by the structural interaction between various elements of their chemical structure. Natural polyphenols have the ability to eliminate free radicals, bind metal prooxidants, decrease radicals, inhibit oxidases, and terminate the oxidation chain reaction [28]. These metabolites are reported to have considerable antioxidant activity [29]. For better understanding of the plant metabolites, evaluation of such metabolites is recommended [30].

Based on in vitro findings, the studied medicinal plants are intriguing and promising as a possible source for new antioxidant medications.

4. CONCLUSION

The selected medicinal plant parts *Tacoma stans* leaves and rhizome of *Zingiber officinale* contain considerable amount of polyphenolic compounds and flavonoids which show high antioxidant activity. As such, *T stans* and *Z officinales* can be further explored as possible antioxidant agents as well as resource for the production of novel drugs.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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