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Effect of thermal processing on total phenolic content and antioxidant activity of *Coriandrum sativum* L. leaves

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Coriandrum sativum is a promising functional food which not only provides nutrition, but also has medicinal benefits. It is a widely grown herb and most commonly used spice in India.Total phenolic content and antioxidant activity of ethanolic extracts of coriander leaves at different temperatures were evaluated to determine the effect of thermal processing on potential health benefits of coriander. The leaves were subjected to blanching (80°C), boiling (100°C) as well as storage at refrigerated temperature (4°C). A qualitative phytochemical screening was performed for the presence of phytochemicals. The ethanolic extracts were analyzed for total phenolic content using Folin-Ciocalteau assay and free radical scavenging activity using 2,2-diphenyl-1-picrylhydrazyl (DPPH) radicals. The extracts of fresh leaves showed the highest total phenolic content, which reduced significantly after treatment 100°C and similar trend was observed with antioxidant activity. Increase in temperature reduced the antioxidant activity of coriander leaf extracts. Refrigeration also results in reduction of total phenolic content and antioxidant activity. This indicates that certain bioactive compounds such as polyphenols and phenolic acids are degraded during processing, resulting in reduced antioxidant potential and total phenolic content, thereby decreasing the medicinal value of herb. The study thus, suggests the consumption of fresh coriander leaves to obtain the maximum benefit.

Key words : *Coriandrum sativum* L., Total phenolics, Antioxidant potential, Phytochemical screening

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INTRODUCTION

Coriander is a common food adjunct, which has been used for flavouring and seasoning throughout the world for thousands of years. Coriander (*Coriandrum sativum* L.) is an annual, herbaceous plant belonging to family Apiaceae. It is a flavouring substance used since ancient times and has been enjoyed by many cultures for its culinary and medicinal values (Hill and Sharma, 1998). The plant is widely grown for seed, leaf and essential oil. Seeds are widely used in curry powders, sausages and seasonings. Green leaves have a specific flavour and are used to garnish curries and in chutneys and soups. Leaves spoil quickly when removed from the plant, and lose their aroma when dried or frozen (Brechbill, 2012). Since heating also results in diminished flavour and hence, fresh leaves are preferred. Coriander contains many bioactive components such as linalool, α-pinene, γ-terpinene, cymene along with various non—linalool alcohols and esters. Other constituents include flavonoids, coumarines, isocoumarines, phthalides and phenolic acids (Verma *et al.*, 2011).

Coriander is well known for its antioxidant properties (Wangensteen *et al*., 2004, Diederichsen, 1996) and recent research has indicated that it is a rich source of flavonoids such as quercetin, kaempferol, and acacetin (Nambiar *et al*,. 2010). The polyphenols constitute a wide and complex array of phytochemicals that exhibit antioxidant action and consequently a beneficial physiological effect (Al-Juhaimi and Ghafoor, 2011). They can protect human body from free radicals and could retard the process of many chronic diseases such as cancer, cardio-vascular disease and diabetes; they can also reduce lipid oxidative rancidity in foods (Regnault-Roger *et al*., 2004; Arts and Hollman, 2005; Williamson and Manach, 2005). Polyphenolic compounds have high antioxidant activity due to the reactivity of the phenol ring and are categorized into different classes depending upon the number of phenol rings. The main groups are flavonoids, phenolic acids, phenolic alcohols, stilbenes and lignans. The antioxidant activity of phenolic acids is due to their ability to scavenge free radicals,

donate hydrogen atoms or electrons or chelate metal cations (Tahira *et al*., 2011). Over the past years, researchers and food manufacturers have become increasingly interested in polyphenols. The chief reason for this interest is the recognition of the antioxidant properties of polyphenols, their great abundance in our diet and their probable role in the prevention of various diseases associated with oxidative stress, such as cancer, cardio-vascular and neurodegenerative diseases (Nambiar *et al.*, 2010; Chawla and Thakur, 2013).

The quantity of phenolic compounds present in plants is influenced by genotype, storage method and the environmental conditions. Thus, it is important to determine the level of these compounds present in the plant after different thermal treatments. The present paper highlights the effect of refrigeration and conventional cooking methods such as blanching and boiling on the total phenolic content and antioxidant activity of coriander leaves as well as correlation between total phenolic content and antioxidant activity of coriander leaves. The plant extracts were also screened for qualitative phytochemical screening.

RESEARCH METHODOLOGY

Chemicals :

Ethanol, methanol, sodium carbonate, gallic acid, Folin-Ciocalteu reagent, and DPPH (1,1-diphenyl-2-picrylhydrazyl) were used. All the chemicals and reagents used were of analytical grade.

Collection of plant material and sample preparation :

Fresh leaves of *Coriandrum sativum* were purchased from a local vendor in Delhi, India and washed with tap water. 200 grams of leaves were taken and divided into 4 equal parts (50 grams each). One portion was retained fresh; others were given different thermal treatments, as given below.

Blanching :

Coriander leaves (50 g) were blanched at 80°C for 1 min. The sample was drained off and cooled rapidly with cold water.

Boiling :

Leaves (50 g) were boiled for 15 min, drained off and cooled rapidly.

Refrigeration :

Coriander leaves were kept at 4°C in refrigerator for 5 days.

Preparation of extracts :

Leaves were extracted with ethanol at room temperature prior to removal of the solvent. Coriander leaves were soaked in 500 ml of 99.9 per cent ethanol for 2-3 days separately. The soaked material was filtered and the extracts were collected. This process was repeated thrice and filtrates were collected. The filtrates obtained were concentrated under vacuum on a rotary evaporator (Buchi Rotary Evaporator, Model R-124) and stored at 4°C for further use (Song *et al*., 2010).

Phytochemical screening :

Ethanolic extracts of fresh coriander leaves were used for qualitative screening of phytochemicals as per standard biochemical procedures. The tests were performed to confirm the presence of alkaloids, carbohydrates, proteins and amino acids, glycosides, flavonoids, tannins, phenolics, terpenoids and steroids (Tiwari *et al.*, 2011).

Estimation of total phenolic content :

The total phenolic content in ethanolic extracts of coriander leaves was estimated by Folin-Ciocalteu reagent, as described by Singleton and Rossi (1965). Gallic acid stock solution (1000 µg/ml) was prepared by dissolving 100 mg of gallic acid in 100 ml ethanol. Various dilutions of standard gallic acid were prepared from this stock solution. Calibration curve was plotted by mixing 1 ml aliquots of 1.0, 2.5, 5.0, 10, 25, 50 and 100 µg/ml of gallic acid solution with 5.0 ml of Folin-Ciocalteu reagent (diluted ten-fold) and 4.0 ml of sodium carbonate solution (75 g/l). The absorbance was measured after 30 min at 20°C at 765 nm. 1 ml of ethanol extract was mixed separately with the same reagents and absorbance was measured at 765 nm after 1 hour. The total phenolic compound in all the extracts was determined using the formula:

$$C = C_1 \times V/M$$

where, C= Total content of phenolic compounds in mg/g in GAE (Gallic acid equivalent); C_1 = The concentration of gallic acid established from the standard curve in mg/ml; V=The volume of extract in ml, M=Weight of plant extract in grams.

Determination of free-radical scavenging activity using DPPH method :

The free radical scavenging activity of test samples was measured by 1, 1- diphenyl-2-picrylhydrazyl (DPPH) (Kaur and Arora, 2011). A 0.1 mM solution of DPPH in methanol was made and 1.5 ml of this solution was added to 0.5 ml of extract solution in methanol at different concentrations (100-500 µg/ml). The mixture was shaken vigorously and allowed to stand at room temperature in dark for 30 min. The absorbance was then measured at 517 nm using a spectrophotometer. A blank without DPPH was used to remove the influence of the colour of samples. A methanolic solution of DPPH was used as negative control. The DPPH radical scavenging activity was calculated using the following equation :

$$\text{DPPH Scavenging effect (\%)} = \frac{A_0 - A_S}{A_0} \times 100$$

where A_o is the absorbance of negative control and A_S is the absorbance of sample.

RESEARCH FINDINGS AND ANALYSIS

The results of qualitative phytochemical analysis of ethanolic extracts of fresh coriander leaves showed the presence of proteins and amino acids, carbohydrates, glycosides, phenolics, tannins, saponins, terpenoids, sterols, and flavonoids (Table1).

Table 1: Qualitative phytochemical screening of coriander extracts

Sr. No.	Phytochemical	Coriander
1.	Alkaloids	-
2.	Proteins and amino acids	+
3.	Carbohydrates	+
4.	Phenols	+
5.	Terpenoids	+
6.	Sterols	+
7.	Saponins	+
8.	Glycosides	+
9.	Flavanoids	+
10.	Tannins	+

The amount of total phenols was determined using Folin-Ciocalteu reagent. Gallic acid was used as standard compound. The standard curve of gallic acid concentrations and absorbance is shown in Fig 1. Found standard curve equation was :

$y = 0.0106x + 0.041$

$R^2 = 0.996$

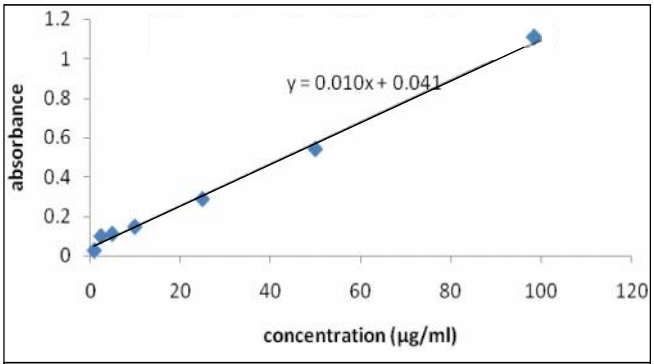

Fig. 1 : Gallic acid standard curve

Table 2: Total phenolic content of ethanol extracts of coriander

Sr. No.	Sample	Absorbance at 765nm (Mean ± Standard error)	Total phenolic content (mg gallic acid equivalents per gram weight)
1.	Fresh coriander leaves	0.866±0.0015	128.83 ± 0.23
2.	Refrigerated coriander leaves	0.649±0.0008	95.03 ± 0.14
3.	Blanched coriander leaves	0.096±0.0012	8.81 ± 0.19
4.	Boiled coriander leaves	0.065±0.0017	3.91 ± 0.27

The total phenolic content of ethanol extracts of coriander is given in (Table 2 and Fig. 2). Data expressed as mean ± standard error of three samples analyzed separately.

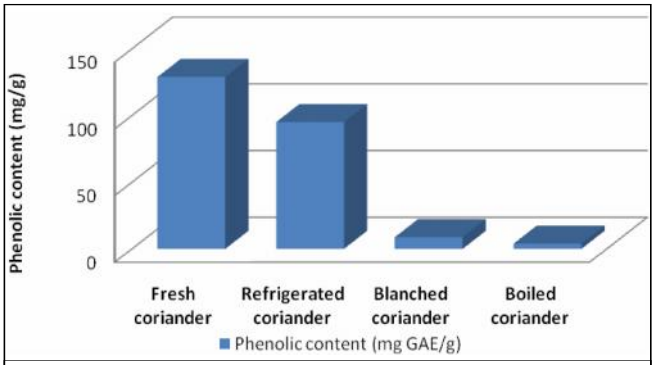

Fig. 2 : Effect of thermal processing on phenolic content of coriander leaves

The reduction capability of DPPH was determined by the decrease in its absorbance at 517 nm induced by the antioxidants. DPPH reacts with the antioxidants and gets converted into 1, 1-diphenyl-2-picrylhydrazine by accepting a hydrogen atom and hence, shows a decrease in absorbance. The methanolic extracts of coriander showed a concentration-dependent DPPH radical scavenging activity. Results are presented in (Table 3 and Fig. 3).

Table 3 : Per cent scavenging activity in coriander extracts at different concentrations

Concentration (µg/ml)	Per cent scavenging activity				
	Ascorbic acid	Fresh coriander	Refrigerated coriander	Blanched coriander	Boiled coriander
100	51.5	74.4	61	48.4	26.3
200	61.6	75.1	65.6	53.3	28.2
300	77.3	76.8	73.9	53.7	32.8
400	78.6	86.6	78	60.2	36.2
500	88.46	92.6	89	62.1	38.9

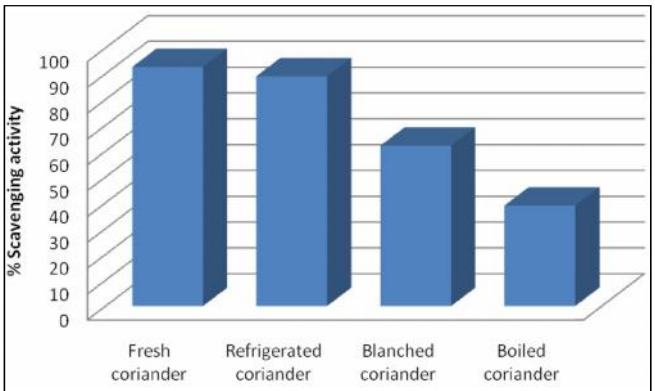

Fig. 3 : Per cent scavenging activity in coriander leaves at 500 µg/ml

Highest concentration of polyphenols was found in fresh coriander leaves (128.83 ± 0.23 mg GAE/g) and hence, the antioxidant activity was the highest in fresh leaves (92.6%) at 500µg/ml concentration (Fig. 4). Randhir *et al.* (2007) suggested that processing of any kind can alter the content, activity and bioavailability of bioactive compounds. Heat treatment has found to decrease the total phenolic content as well as antioxidant potential of coriander leaves. Results show that refrigeration also affects the antioxidant activity but the reduction is less as compared to thermal treatment. Blanching drastically reduces the phenolic content and thus, the antioxidant activity. In boiled samples, the total phenolic content as well as the scavenging activity of DPPH has been reduced drastically indicating the destruction of phenolic compounds. The total phenolic content and antioxidant activity followed the order:

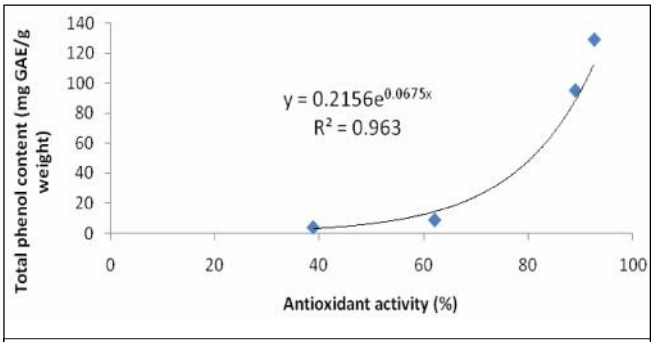

Fig. 4 : Correlation between antioxidant activity and total phenolic content

Fresh leaves > Refrigerated leaves > Blanched leaves > Boiled leaves

Conclusion :

Phytochemical screening of fresh leaf extracts revealed the presence of phenolic compounds, which are responsible for potential health benefits of coriander such as antioxidant and antimicrobial activity. Due to the changes in the lifestyle, the consumption of fresh leaves has been minimized. The above results indicate that thermal processes like blanching and boiling as well as storage under refrigerated conditions for longer periods leads to destruction of phenolic compounds and other phytochemicals and hence, reduces the antioxidant potential of coriander. The study suggests the use of fresh coriander leaves in order to obtain the maximum potential.

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