IJBPAS, May, 2013, 2(5): 1141-1150

ISSN: 2277–4998

International Journal of Biology, Pharmacy and Allied Sciences (IJBPAS)

'A Bridge Between Laboratory and Reader'

www.ijbpas.com

# ANTIOXIDANT PROFILE OF ORGANIC AND CONVENTIONAL GROWN GREEN TEA (*CAMELLIA SINENSIS*)

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## ABSTRACT

Tea is the most widely consumed beverage in the world. Green tea (*Camellia sinensis*) is a good source of bioactive compounds and it is gaining interest due to its health benefits. In the present study, a comparison of antioxidant profile of organic and conventional green tea was carried out. For this, polyphenol, flavonoid, and total antioxidant capacity by DPPHRSA and FRAP were analysed using standard methods. Results from this study showed that organic green tea showed a significant higher values of total phenol, flavonoid, FRAP and DPPHRSA. Regression analysis revealed that total phenol content showed a positive and significant correlationship with DPPHRSA and FRAP in organic green tea. In conventional green tea, total phenol showed it with FRAP. It revealed that the total antioxidant capacity could be due to its total phenol content. The study concludes that organic green tea offers higher amount of antioxidant properties without any adverse effects of synthetic pesticides.

**Keywords: Green Tea (*Camellia sinensis*), Organic, Conventional, Antioxidant Capacity**

## INTRODUCTION

Tea is the most widely consumed beverage in the world. The type and quantity of tea taken varies in different countries **[1, 2]**. A recent awareness of health benefits has increased consumers' interest in this beverage especially green tea. Green tea is derived from drying and steaming the fresh tea leaves and thus no oxidation occurs, resulting in high levels of catechins **[3]**. Traditionally, Green tea was used to improve blood flow, eliminate alcohol and toxins, improve resistance to disease, relieve joint pain and to clear urine and improve its flow **[4]**. Tea contains large amounts of

polyphenolic compounds with antioxidant properties, and these may prevent oxidative damage of DNA **[5]**. Tea is also rich in flavonoids and other polyphenol compounds which have different beneficial activity such as anticarcinogenic **[6]** cholesterol lowering **[7]**, antiviral, antibacterial **[8, 9]**. Green tea catechins have the potential to alleviate symptoms of the Metabollic syndrome **[3]**.

However, these benefits of green tea could be affected by its cultivation practices. Use of pesticides and chemical fertilizers in cultivation may affect its properties negatively. Many pesticides are toxic substances and persistent in character **[10]**.

Tea plants are sprayed with endosulfan, dimethoate, fenpyroximate, hexythiazox, oxyflurofen **[11]**. The perceived potential hazards of modern agricultural practices, such as the use of pesticides and their residues in food, are likely to be associated with long-term and unknown effects on health **[12, 13]**. For quite some time now, there has been a growing social desire reflected to reduce the risk of pesticide by the society. Organically grown foods are an alternate to the potential threat posed by excessive use of the pesticide.

Organic foods are foods that are produced using methods that do not involve modern synthetic inputs such as synthetic pesticides and chemical fertilizers, do not contain genetically modified organisms, and are not processed using irradiation, industrial solvents, or chemical food additives **[14]**. Nowadays, many consumers prefer organic to nonorganic food because of developing greater interest in both a healthier-safer diet and a better environment **[15, 16, 17]**. Therefore, the study was planned with objectives to analyze nutrients and bioactive compounds of organically and conventionally grown green green tea.

## MATERIALS AND METHODS

### Samples

Green tea samples from organic and conventional source were procured. 500 grams of certified organic tea was purchased from organic food store and conventional green tea was purchased from local grocessary store of Ahmedabad, Gujarat, India. Samples were powdered using kitchen grinder (Philips Ltd.) to obtain a fine powder..

### Chemicals Used

Trolox, Gallic acid, Rutin, Folin cio-calteu, 1,1-Diphenyl-2-picrylhydrazyl (DPPH), and 2,4,6- Tris (2-pyridyl)-s-triazine (TPTZ) were purchased from Sigma-Aldrich Ltd. (India) and methanol was of analytical grade.

### Preparation of the Sample for Total Phenol, Flavonoid and Total Antioxidant Capacity

Tea extract was prepared by infusing 1 gram of green tea in 100 ml of boiling distilled water. Each sample was boiled for 2 minutes. The extracts were centrifuged at 7000 rpm for 10 minutes and supernatant were collected in separate flask. The green tea sample from organic and conventional sources was extracted twice with the same procedure. After cooling the extracts were used for total phenol, flavonoid and antioxidant activity analysis.

**Total Phenol Content Assay**

Total phenol content was measured by the spectrophotometric method **[18]**. Aliquots (0.1ml and 0.2 ml) sample extracts of the sample were mixed with remaining volume of Distilled water to made the total volume 1 ml which were further mixed with 1ml of folin cio-calteu (50% v/v with D/W) and 1 ml of 35 % sodium carbonate reagents. All the tubes were vortexed (Gilson Ltd.) and kept for 1 hour incubation at 37°C. 2 ml of distilled water was added to all the tubes after incubation and the absorbance was measured at 620 nm against D/W as a blank. Gallic acid was used as a standard. Total Phenol content was expressed as Gallic acid Equivalent.

**Flavonoid Assay**

The concentrated extracts of the samples were used to analyse flavonoid content by spectrophotometric method **[19]**. 0.1 and 0.2 ml aliquot of the sample were taken and made 1 ml with 95% methanol. Tubes were further added with 0.1 ml of 10 % aluminium chloride, 0.1 ml of 1M Potassium acetate and 2.8 ml of D/W. After incubation of 30 minutes at 37°C, absorbance was measured at 415 nm against 95% methanol as a blank. 10 mg% Rutin was used as a standard. Flavonoid content was expressed as rutin equivalents.

**Total Antioxidant Capacity Assay by Ferric Reducing Antioxidant Power**

Total antioxidant capacity of extracts was determined by using the method **[20]**. Suitable aliquote of extracted sample was taken in clean and dry test tube and volume was made 300 µl with distilled water and 1.8 ml of freshly prepared FRAP reagent (Acetate buffer (pH- 3.6) + TPTZ solution+ 20 mM Ferric chloride) was added and vortexed. After incubation of 10 minutes at 37C absorbance was read at 593nm spectrophotometrically. Different aliquots of Trolox were treated as standard and results were expressed in terms of TEAC (mg of Trolox equivalent/100g).

**Total Antioxidant Capacity Assay DPPH Radical Scavenging Activity of Fresh Green Tea**

1, 1- diphenyl, 2-picrylhydrazyl (DPPH) scavenging activity was measured by the spectrophotometric method **[21]**. A solution of DPPH in methanol was prepared freshly. 3 ml aliquot of this solution was mixed with

0.1 ml of the samples. The solutions in the test tubes were shaken well and incubated in the dark for 15 min at room temperature. The absorbance was measured at 517 nm against methanol as blank. Control tube containing 1 ml of methanol and 3 ml of DPPH reagent was also noticed for absorbance. Gallic acid was used as a standard and Total antioxidant capacity was expressed as Gallic acid Equivalent

**Statistical Analysis**

Mean, standard error of mean, t –test and regression analysis was done to determine the difference between the means and significance was defined at $P < 0.05$ and $P < 0.01$ using SPSS (version-16).

**RESULT AND DISCUSSION**

Phenolic compounds are a class of free radical terminator. The antioxidant activity of phenolic compounds is mainly due to redox properties, which allow them to act as reducing agents, hydrogen donors, singlet oxygen quenchers, heavy metal chelators, and hydroxyl radical quenchers **[22]**. Radical scavenging activity toward either reactive oxygen species or toward lipid peroxidising radicals proceeds *via* hydrogen atom transfer or electron donation; prevention of the transition metal−catalyzed production of reactive species, interaction with other antioxidants, localization, and mobility of the antioxidant at the microenvironment

Total phenol content, total flavonoid content and antioxidant capacity by FRAP, DPPHRSA of organic and conventional green tea are shown in **Table 1**. Total phenol content of organically grown green tea was 147.576 mg GAEqui/gram which was significantly ($P < 0.01$) higher than conventionally grown green tea (122.568 mgGAEqui/gram). Similarly flavonoid content was also significant ($P<0.01$) higher in organic green tea (84.691 mgRutinEq/gram) than conventional green tea (51.542 mgRutinEq/gram).

Total antioxidant capacity by DPPHRSA was also found significant ($p<0.01$) higher in organic green tea (7.238 mgGAEqui./gram) compared to conventional green tea (4.292 mg GAEqui/gram). Similarly, Ferric reducing power activity of organic green tea was significantly ($p<0.01$) higher than conventional green tea. Organic green tea has ferric reducing antioxidant power of organic green tea was 8.1197 gTEqui../gram and same of conventional green tea was 6.077 gTEqui./gram.

Looking at correlation study, in organically grown green tea, total phenol has positive and significant correlation with FRAP ($R^2 = 0.929$, $p=0.036$) and DPPHRSA ($R^2 = 0.989$, $p=0.011$). It also showed positive but nonsignificant correlationship with flavonoid ($R^2 = 0.830$, $p=0.089$). Flavonoid

content showed positive and non significant correlationship DPPHRSA ($R^2$ = 0.863, p=0.071) and FRAP ($R^2$ = 0.871, p=0.067) as shown in **Figure 1(a)** and **1(b)**.

In conventional green tea, total phenol showed a positive but non significant correlationship with DPPHRSA ($R^2$ = 0.850, p=0.078) and FRAP ($R^2$ = 0.530, p=0.272). Flavonoid content showed a significant and positive correlation with FRAP ($R^2$ = 0.914, p=0.044) but non significant correlation with DPPHRSA ($R^2$ = 0.849, p=0.079) as shown in **Figure 2 (a)** and **2(b)**.

Phenolic content of green tea was ranged from 0.030GAg/g to 0.196 gGA/g where as flavonoid content ranged from 2.898 mg/g to31.152 mg/g. Similarly, ferric reducing antioxidant potential of green tea ranged from 0.554 mmol/gram to 3.082 mmol/gram **[23]**. FRAP of green tea 272- 1144 micromol/g and it was stongly correlated with total phenol content **[24]**. The health-promoting effects of regular green tea consumption are mainly attributed to its polyphenol content, which represents 35% of the dry weight **[25, 26]**. These polyphenols in turn consist of several fractions such as flavanols and flavonols **[4]**.

Two major hypotheses explaining the possible increases in organic acids and polyphenolics in organic versus conventionally grown foods have been proposed. One hypothesis considers the impacts of different fertilization practices on plant metabolism. In conventional agriculture, synthetic fertilizers frequently make nitrogen more available for the plants than do the organic fertilizers and may accelerate plant growth and development. Therefore, plant resources are allocated for growth purposes, resulting in a decrease in the production of plant secondary metabolites (compounds not essential to the life of the plant) such as organic acids, polyphenolics, chlorophyll, and amino acids **[27]**.

The second hypothesis considers the responses of plants to stressful environments such as attacks from insects, weeds, and plant pathogens. It has been argued that organic production methods which are limited in the use of insecticides, herbicides, and fungicides to control plant pests may put greater stresses on plants and may require plants to devote greater resources toward the synthesis of their own chemical defence mechanisms. Increases in antioxidants such as plant polyphenolics have been attributed to their production in plant defense **[28]**.

**Table 1: Phenolic Profile of Organic and Conventional Green Tea**

| Type of green tea | Organic Green Tea | Conventional Green Tea |
|---|---|---|
| Total Phenol (mgGAEq/gm) | 147.576 ± 3.055* | 122.568 ± 1.190 |
| Flavonoid (mgRutin Eq/gm) | 84.691 ± 1.460* | 51.542 ± 1.314 |
| DPPHRSA (mgGAEq/gram) | 7.238 ± 0.687* | 4.292 ± 0.538 |
| FRAP (gTroloxEq/gram) | 8.506 ± 0.685* | 6.005 ± 0.104 |

**NOTE: Values are Mean ± SEM of Four Replicates; * Significant Difference at P <0.01**

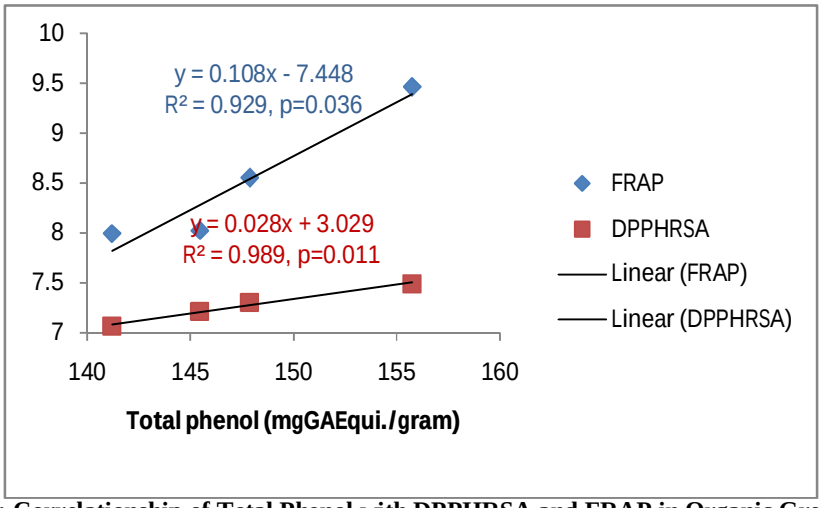


**Figure 1(a): Correlationship of Total Phenol with DPPHRSA and FRAP in Organic Green Tea**

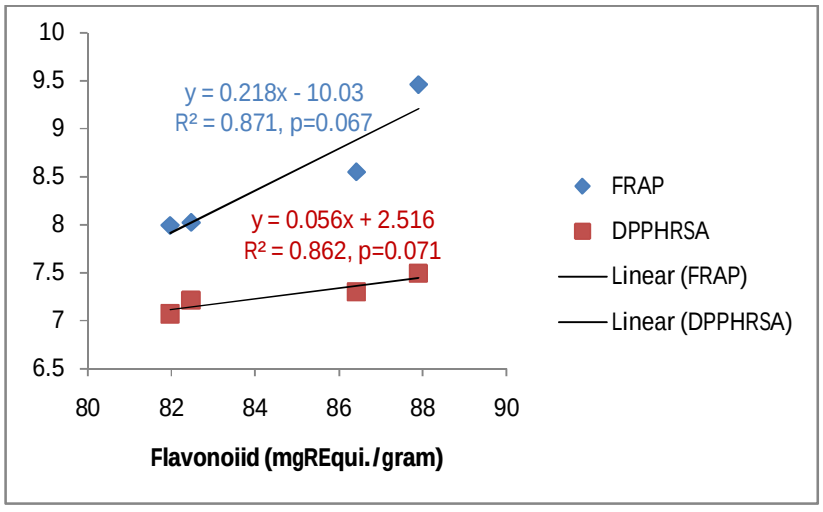


**Figure-1(b): Correlationship of Flavonoid with DPPHRSA and FRAP in Organic Green Tea**

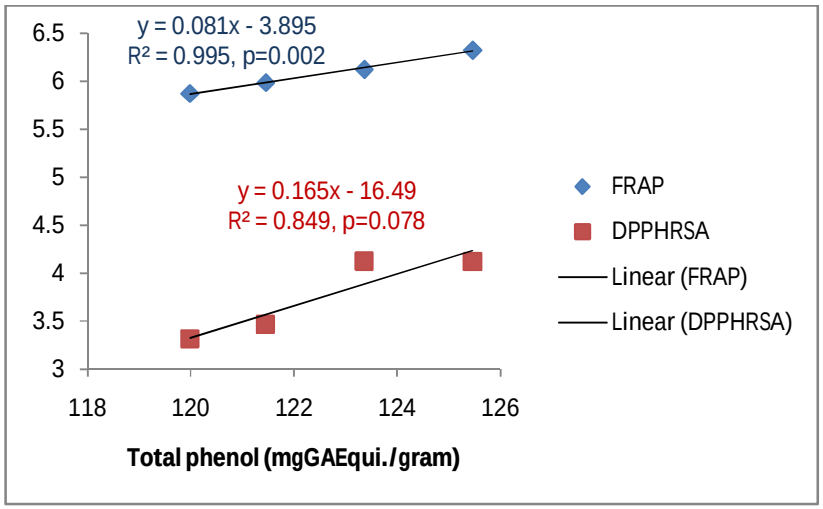


**Figure 2(a): Correlationship of Total Phenol with DPPHRSA and FRAP in Conventional Green Tea**

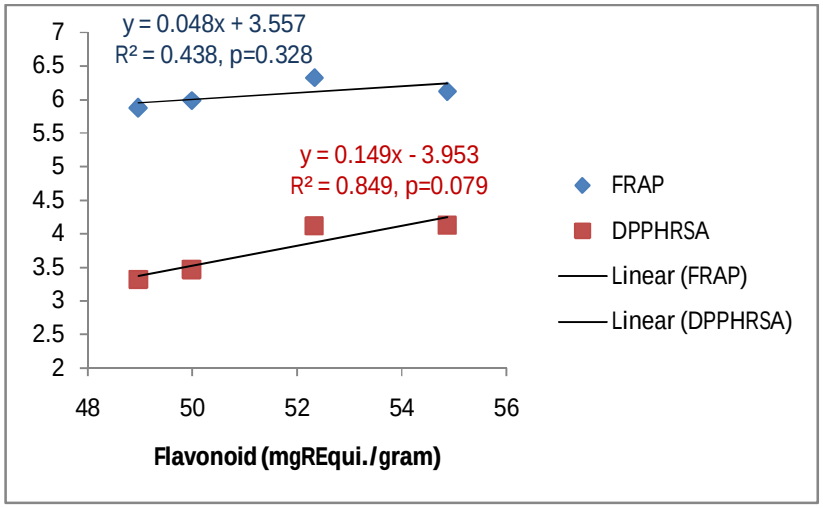


**Figure 2(b): Correlationship of Flavonoid with DPPHRSA and FRAP in Conventional Green Tea**

## CONCLUSION

Organic gren tea showed a significant higher concentration of polyphenol, flavonoid as well as ferric reducing activity and free radical scavenging activity. Total phenol content of tea determines its total antioxidant capacity. In organic production, synthetic chemicals in the form of pesticide and fertilizers are not permitted. Therefore, organically grown plants are usually exposed to different forms of stress, which induce accumulation of phenolic compounds.

In recent time, it has been observed that phytochemicals with functional properties and safety are in great demand in foods and pharmaceuticals sectors. Hence it could be concluded that organically grown green tea

has high antioxidant potential with reduced threat of adverse effects of pesticide, can be considered as a functional food which is needed for the maintenance of good health.

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