



## A Review on Antioxidant Property of Herbal Tea

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### ABSTRACT

Antioxidants are substances found in the medicinal plants which may have a protective role to play in certain conditions such as heart disease, stroke and some cancers. Polyphenols, which are antioxidant components, can be found in tea extracts. Natural antioxidants have gotten a lot of attention and research since they are effective due to free radical scavenger activity and are thought to be less hazardous than synthetic antioxidants. Tea, along with water, is one of the world's most popular beverages. tea (Camellia sinensis) have classified into green tea, black tea, oolong tea. Spectroscopy is a common method for evaluating antioxidants. It is a hydrogen atom transfer (HAT) based approach that uses the following methods for antioxidant assay: DPPH, ABTS, FRAP, PFRAP and it shows that the tea plant possesses the antioxidant property of tea by using various extracts of tea and shows the green tea has more antioxidant property than black tea. The antioxidant capacity measured by two techniques electrochemical technique and chromatographic technique. The actual antioxidant content is given directly in the amperometry technique.

**Keywords:** Antioxidant, DPPH, ABTS, FRAP, PFRAP, Antioxidant capacity

### Antioxidant:

Antioxidants are compounds found in medicinal plants that may have a preventive impact in illnesses such as heart disease, stroke, and cancer.<sup>1</sup> Mechanisms of antioxidants like a chain-breaking process and quenching chain-initiating catalyst to remove reactive oxygen species.<sup>2</sup> Polyphenols, which are antioxidant components, can be found in tea extracts. Natural antioxidants have gotten a lot of attention and research since they are effective due to free radical scavenger activity and are thought to be less hazardous than synthetic antioxidants.<sup>3</sup> Antioxidant activity and total antioxidant content in foods, beverages, dietary supplements, and herbal extracts have become increasingly popular. Regular consumption of antioxidant-rich foods and beverages can help to minimize the harmful effects of free radicals and oxidative stress.<sup>4</sup>

Any substance capable of stabilising or deactivating free radicals before they assault cells is referred to as an "anti-oxidant." Antioxidants function as radical scavengers, hydrogen donors, electron donors, peroxide decomposers, singlet oxygen quenchers, enzyme inhibitors, synergists, and metal chelating agents. An antioxidant is a chemical that can prevent other molecules from oxidising. Oxidation is a chemical reaction in which electrons are transferred from a material to an oxidizer. Free radicals are produced by oxidation events, and free radicals can launch chain reactions that harm cells. Antioxidants stop these chain reactions from continuing by eliminating free radical intermediates and preventing additional oxidation processes.

Antioxidants are generally reducing agents since they do this by getting oxidised themselves. radicals such as superoxide (O<sub>2</sub>•), hydroxyl (OH•), peroxy (RO<sub>2</sub>•), hydroperoxyl (HO<sub>2</sub>•), alkoxyl (RO•), peroxy (ROO•), nitric oxide (NO•), nitrogen dioxide (NO<sub>2</sub>•), lipid peroxy (LOO•), nitric oxide (NO•), nitrogen dioxide (NO<sub>2</sub>•), and lipid peroxy (LOO•). Free radicals harm cells at various levels: they attack lipids and proteins in the cell membrane, preventing the cell from performing its essential duties (transport of nutrients, waste disposal, cell division, etc.)<sup>1</sup>

### Antioxidant Mechanism of Action:

1) The first is a chain-breaking process in which the principal antioxidant donates an electron to the system's free radical.

2) The second method includes quenching chain-initiating catalyst to remove reactive oxygen species ROS/reactive nitrogen species initiators (secondary antioxidants).<sup>2</sup>

Spectroscopy is a common method for evaluating antioxidants. It is a hydrogen atom transfer (HAT) based approach that uses the following methods for antioxidant assay: DPPH, ABTS, FRAP, PFRAP.<sup>3</sup>

**TEA:**

Tea, along with water, is one of the world's most popular beverages.<sup>6</sup> Flavan-3-ols, phenolic acids, purine alkaloids, condensed tannins, hydrolysable tannins, saponins, flavonols, and their glycoside derivatives are the primary secondary metabolites found in fresh tea leaves. Caffeine is the second major contents are flavonol and flavonol act as antioxidant[12].caffine do not affect on the antioxidant affect of teas.

Many of the health benefits claimed to tea may be due to flavonoids, a wide category of phenolic products of plant metabolism with a range of phenolic structures and distinct biological activities. Flavonoids may be responsible for many of the health advantages attributed to tea.<sup>5</sup>Polyphenols, which are antioxidant components, are found in tea extracts.<sup>2</sup>Green and black tea include antioxidants that may protect against cancer, cardiovascular disease, and neurological illnesses, among other ailments.<sup>1</sup>

Black tea is preferred by 76-78 percent of consumers worldwide, followed by green tea (20-22 percent) and oolong tea (2 percent).



Fig 1 Tea plant

Kingdom Plantae - plantes, Planta, Vegetal, plants

Sub Kingdom -Viridiplantae

Infra Kingdom Streptophyta - land plants

Super Division -Embryophyta

Division- Tracheophyta vascular plants, tracheophytes j

Sub Division -Spermatophytina spermatophytes, seed plants, phanérogames

Class -Magnoliopsida

Super Order- Asteranae

Order- Ericales

Family -Theaceae –tea

Genus Camellia L. –tea

Species -Camellia sinensis (L.) Kuntze – tea.<sup>2</sup>

Camellia sinensis is a plant belonging to the Camellia sinensis genus. The Sinensis species is split into two types: sinensis and assamica. South east China, Darjeeling, and Japan are home to Camellia sinensisvar.sinensis. Assam, Thailand, and Sri Lanka are home to Camellia sinensis var assamica.<sup>6</sup>

China, India, Sri Lanka, Kenya, and Indonesia are the leading tea-producing countries, accounting for 80 percent of global production. India is a significant tea producer, consumer, and exporter, accounting for 31% of global tea production.<sup>6</sup>

The methods by which different tea are made and these teas are used for further study of antioxidant in various ormulation.

1. Green tea
2. Black tea.
3. Oolong tea.
4. White tea

## Tea manufacturing process

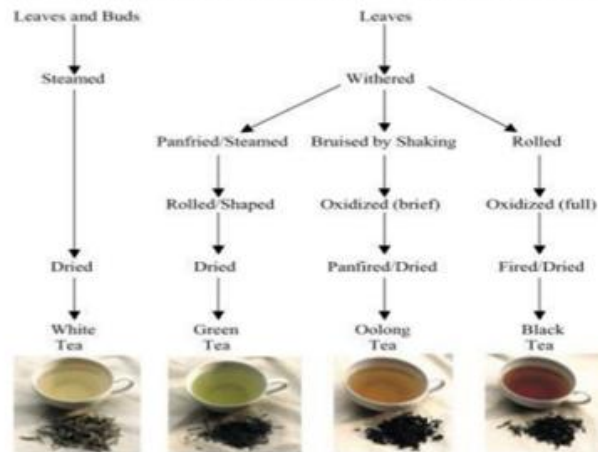


Fig no 2 methods of preparing tea

Teas are classified into the following categories:

**1) Green tea** :is made from unfermented leaves, unlike oolong tea leaves, which are partially fermented, and black tea leaves, which are fully fermented. Green tea is high in catechins, which are flavanol monomers, which are a type of flavonoid. Epicatechin (EC), epigallocatechin (EGC), epicatechin-3- gallate (ECG), and epigallocatechin-3- gallate (EGG) are among the catechins.<sup>6</sup>

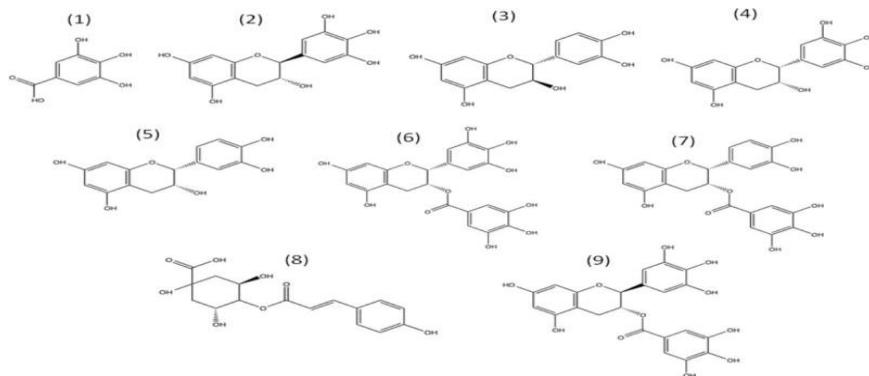


Figure no-3 structure of polyphenols in tea

Chemical structures of the major green tea polyphenols. Structures shown: (1) Gallic acid, (2) (-)-gallocatechin, (3) (+)-catechin, (4) (-)-epigallocatechin, (5) (-)-epicatechin, (6) (-)-epigallocatechin gallate, (7) (-)-epicatechin gallate, (8) p-coumaroylquinic acid, and (9) (-)-gallocatechin gallate.



Figure no-4 Green tea

**2) Black tea:** While most EGCG antioxidants are oxidised during the fermentation process, black tea retains a significant percentage of antioxidant polyphenols such as flavonoids. When the catechins in black tea are oxidised, they are transformed into theaflavins and thearubigins, which are still antioxidants.<sup>6</sup>



Fig no-5 Black tea

3) **White Tea:** Before the buds fully open, the buds and immature tea leaves are harvested. The leaves are then steamed and dried with as little processing as possible. As a result, white tea has the highest antioxidant content and the lowest caffeine content of all the teas.<sup>7</sup>

4) **Oolong Tea:** Oolong tea is a partially fermented tea and has the flavour and health features of teas, both green and black. It contains a high number of antioxidants

## Methods for evaluating antioxidants

Spectroscopy is a common method for evaluating antioxidants. It is a hydrogen atom transfer (HAT) based approach that uses the following methods for antioxidant assay: DPPH, ABTS, FRAP, PFRAP, etc.

Various tea samples were prepared by adding 0.2 g of tea in 10 ml of distilled water and steeped for 5 minutes at 95-100°C. The hot water samples were filtered using Whatman's filter paper and the filtrate was used for further investigations

### 1) DPPH:

The stock solution was made by dissolving 25 mg of DPPH in 100 mL of methanol and storing it at -20°C until needed.<sup>8</sup>

Due to the delocalization of the spare electron on the entire molecule, DPPH• (2,2-diphenyl-1-picrylhydrazyl) is a stable free radical. As a result, unlike other free radicals, DPPH• does not dimerize. The presence of a purple colour is determined by delocalisation on the DPPH• molecule, having an absorption band with a maximum about 520 nm.

When DPPH• combines with a hydrogen donor, it produces the reduced (molecular) form (DPPH), which results in the violet colour disappearing. As a result, the reduction in absorbance is proportional to the antioxidant content.<sup>11</sup>

To make the stock solution (5 mg/mL), the tested material was combined with 95 percent methanol. A 2.0 mL solution of various doses of plant extract was added to 1.0 mL of DPPH (0.25 mM) in methanol in tubes. After that, the reaction mixture was allowed to sit at room temperature for 30 minutes in a dark laboratory. In an ultra violet spectrophotometer, the change in colour from deep violet to light yellow was detected at 518 nm.<sup>1</sup>

The reference standard was ascorbic acid. As a blank, 95 percent methanol was used. The percent inhibition of DPPH =  $[\text{Abs control} - \text{Abs sample} / \text{Abs control}] \times 100$  was used to calculate the percentage of DPPH scavenging activity.

When we calculate the % inhibition then plot the graph % inhibition versus concentration mg/ml. Calculate IC<sub>50</sub> maximum antioxidant capacity of herbal extract test solution. When we compare the IC<sub>50</sub> of the green tea, black tea and oolong tea we found that the green tea has the more antioxidant effect. At less concentration it shows IC<sub>50</sub> effect.<sup>1</sup>

### 2) ABTS:

The ABTS radical cation decolorization assay was used to test the free radical scavenging activity of plant materials. [8] When an antioxidant is applied to the blue-green chromophore ABTS + (2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)), this method uses spectrophotometry to detect the loss of colour. The antioxidant decolorizes ABTS<sup>•+</sup> by reducing it to ABTS. (11). ABTS<sup>•+</sup> cation radical was generated by the reaction between 7 mM ABTS in water and 2.45 mM potassium persulfate (1:1), stored in the dark at room temperature for 12-16 h before use. After diluting the ABTS<sup>•+</sup> solution with methanol, an absorbance of 0.700 at 734 nm was obtained. The absorbance was measured 30 minutes after the initial mixing of 5 µl of plant extract with 3.995 ml of diluted ABTS<sup>•+</sup> solution. The absorbance of ABTS decreased using the 734 nm technique, which was monitored using spectroscopy.

Percent inhibition of absorbance at 734 nm was calculated using the formula:

ABTS<sup>•+</sup> scavenging effect (percent) =  $((\text{AB} - \text{AA}) / \text{AB}) \times 100$ .<sup>2</sup> where AB is absorbance of ABTS radical + methanol and AA is absorbance of ABTS radical + sample extract/standard.

Take various concentrations and observe the antioxidant effect calculate the graph and % inhibition vs concentration..we have result the more % of inhibition at lower concentration then the more antioxidant effect.<sup>12</sup>

### **3)FRAP**

Another approach for determining antioxidants is the FRAP (ferric reducing antioxidant power) method, which is based on the reduction of ferric – tripyridyltriazine caused by antioxidants present in the sample.[3] The approach is based on the action of electron donating antioxidants at low pH to reduce Fe<sup>3+</sup> TPTZ complex (colourless complex) to Fe<sup>2+</sup>-tripyridyltriazine (blue coloured complex). The change in absorbance at 593 nm is used to monitor this process. At 37°C, the Ferric reducing antioxidant power (FRAP) reagent was made by combining 300 mM acetate buffer, 10 ml TPTZ in 40 mM HCl, and 20 mM FeCl<sub>3</sub>.6H<sub>2</sub>O in a 10:1:1 ratio. Using a 1-5 ml variable micropipette (3.995 ml), freshly made working FRAP reagent was pipetted into 5 µl of the correctly diluted plant sample and thoroughly mixed. When ferric tripyridyl triazine (Fe<sup>3+</sup> TPTZ) complex was reduced to ferrous (Fe<sup>2+</sup>) form, a strong blue colour complex was generated, and the absorbance at 593 nm was measured.<sup>10</sup> Plotting the absorbance at 593 nm versus different concentrations yielded the curve. The higher the absorbance, the greater antioxidant power...and the higher the Fe<sup>2+</sup> concentration. The higher the absorption value, the more powerful the anti-oxidant. The ferric to ferrous ion was decreased in the antioxidant assay.[3] The absorbance may be used to determine the quantity of iron that has been decreased, and it may also be used to determine the quantity of antioxidants present<sup>11</sup>. When we evaluate the antioxidant effects of green tea and black tea using the FRAP assay, we find that green tea has a higher antioxidant effect when the concentrations of both teas are equal

### **4) PFRAP**

This approach was used in the reduction of potassium ferricyanide with the help of potassium ferrocyanide Fe<sup>3+</sup> in antioxidant determination.

The antioxidant extract is measured using a calorimetric approach in the PFRAP method.

This approach involves first reacting any chemical having antioxidant capability with potassium ferricyanide, then altering with ferrocyanide, and then reacting with ferric trichloride to create ferric ferrocyanide, which results in a blue colour complex with the greatest absorbance at 700 nm.

By using different concentration of tea extract we determine their antioxidant study.<sup>10</sup>

## **ANTIOXIDANT CAPACITY :**

Measured by two techniques electrochemical technique and chromatographic technique

### **1)ELECTROCHEMICAL TECHNIQUE:**

#### **a)VOLTAMETRY:**

At the appropriate applied voltage, a chemical is reduced or oxidised at the surface of a working electrode, causing mass transport of fresh material to the electrode surface and the creation of a current.<sup>13</sup>

The current of the cathodic/anodic peak is measured.

#### **b)AMPEROMETRY;**

In comparison to a reference electrode, the working electrode's potential is set at a predetermined value.<sup>14</sup>

The current generated by the oxidation/reduction of an electroactive analyte is measured.

#### **c) BIAMPEROMETRY:**

The analyte (antioxidant) reacts with the oxidised form of a reversible redox couple, suggesting a redox couple.<sup>15</sup>

Measurement of current flowing between two identical working electrodes immersed in a solution containing the examined sample and a reversible redox couple at a minor potential difference.

### **2)CHROMATOGRAPHIC TECHNIQUE**

#### **a)GAS CHROMATOGRAPHY**

The partition between a liquid stationary phase and a gas mobile phase is used to separate the components in a mixture.

Thermal conductivity detection or flame ionization

**b) HIGH PERFORMANCE LIQUID CHROMATOGRAPHY:**

The separation of compounds in a mixture is based on the partitioning of a solid stationary phase and a liquid mobile phase with different polarities, at high flow rates and pressures of the mobile phase UV-Vis (e.g., diode array), fluorimetric detection, mass spectrometry, or electrochemical detection.<sup>16</sup>

**HEALTH BENEFIT OF TEA –**

Tea is high in antioxidants, which are disease-fighting substances that aid in the prevention of illness. These antioxidants' detoxifying properties protect cells from free radical damage, which can cause blood clots, atherosclerosis, and cancer. Tea consumption has also been shown to be useful for prevention of many debilitating human diseases that include maintenance of cardiovascular and metabolic health.<sup>17</sup> The majority of research indicate that those who consume two cups or more of tea per day have a lower risk of heart disease and stroke, have lower total and LDL (commonly referred to as "bad") cholesterol, and recover from heart attacks faster. "It has no calories and a lot of polyphenols," says the tea drinker. Black and green tea can help you lose weight, block allergic reactions, limit tumour growth, protect your bones, battle bad breath, enhance your skin, guard against Parkinson's disease, and even delay the beginning of diabetes. Tea's antioxidants may aid in the prevention of skin cancer. Tea extracts applied to the skin (as a lotion) have also been shown to protect against sun damage, which can lead to skin cancer.<sup>18</sup>

**CONCLUSION:**

Tea having antioxidant effect seen by the DHAP, FRAP, PFRAP, ABTS methods. The tea have antioxidant effect that is more safe than synthesized antioxidant. The green tea having high antioxidant effect than other black tea and oolong tea. The polyphenols in tea plant shows antioxidant property.

The content of antioxidant in various formulations of tea can be determine by amperometry method. The antioxidant content is different in different formulation of tea due to different processing is for make different tea formulation. Antioxidants are beneficial for health and tea is the good source of antioxidant.

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