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Determination of Total Phenolic and Tannins in different Varities of tea by Colorimetry

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A B S T R A C T

Camelia Sinensis Linn., commonly known as 'Tea, is widely distributed in most tropical countries and have been used for varied purposes; to treat different kinds of pathologies, particularly as antioxidant owing to the presence of numerous phytochemical constituents residing in the plant. The powerful biological activities as exhibited by plant phenolics and flavonoids posed the need of determining their contents in *Camelia sinensis*. The present study was aimed at estimation of total phenolics, flavonoids and tannins in alcoholic and aqueous extracts of the different varities of tea. The contents were determined by spectrophotometric assays by measuring the absorbance at different wavelengths. Total phenolic content were estimated by the Folin–Ciocalteu colorimetric method; the total tannin content was estimated by Folin-Denis method whereas the total flavonoid content was estimated by lithium chloride colourimetric method. The alcoholic extract showed highest concentration of phenolics, flavonoids and tannins; ranging between 15-89 µg/mg of gallic acid equivalent, and 15-102 µg/mg of tannic acid equivalents respectively. The results clearly indicate that different varities of tea is a rich source of phenolics, the basis of its traditional use in different systems of medicines.

Keywords: *Camellia sinensis*, Gallic acid, Tannic acid, Colorimetric method

A R T I C L E I N F O

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1. Introduction

Plants synthesize compounds with biological activity, namely antioxidant, as secondary products, which are mainly phenolic compounds serving in plant defense mechanisms to counteract reactive oxygen species (ROS) in

order to avoid oxidative damage. Phenolics are secondary plant metabolites ranging from simple to highly polymerized compounds[1]. Many epidemiological studies have shown that the consumption of phenolics-rich foods is associated with the prevention of chronic diseases[2]. In addition to their antioxidant properties, these compounds have been reported to be potential candidates in lowering cardiovascular diseases [3] and anticarcinogenic activities [4,5] antiallergenic, anti-arthrogenic, anti-inflammatory, antimicrobial and antithrombotic effects [6] Green tea [7] is the most popular form of tea in China. Chinese green teas are made from over 600 different cultivators of the camellia sinensis plant, giving plenty of variety and regional teas. Chinese green teas are traditionally pan-fired, unlike the Japanese steaming process. Other processes in China include oven-dried and sun-dried. Due to the different production process, Chinese teas are said to have a more "earthy" taste than Japanese teas.

2. Materials and Methods

Procurement of material:

The samples of different tea varieties were collected from the local shops of Kurnool. The samples of the tea include Green tea, Black tea and yellow Label tea.

Preparation of extract:

The collected materials were air-dried for twodays at 35-40°C and pulverized in electric grinder. The powdered material was subjected to successive extraction with water for 2 hours by boiling on a water bath.The extracts were concentrated under reduced pressure in a rotary evaporator and dried. The percentage extractive values were recorded.

Determination of Total Phenolic Content

The total phenolic content of the extracts was evaluated by spectrophotometric method measuring the absorbance at 765 nm. One milliliter of sample (concentration 1 mg/mL) was mixed with 1 mL of Folin and Ciocalteu's phenol reagent. After 3 min, 1 mL of saturated sodium carbonate solution was added to themixture and made up to 10 mL with distilled water. The reaction was kept in the dark for 90 min, after which the absorbance was read at 725 nm. Gallic acid was used for constructing the standard curve and the results were expressed as µg of gallic acid equivalents/ mg of extract (GAE).

Determination of Total Tannin Content:

Tannin content of the extracts was measured by Folin–Denis method. The various extracts (50 lL) were made up to 7.5 mL by adding double distilled water. 0.5mL Folin–Denis reagent and 1 mL of Na2CO3 were added to it. Again the volume was made up to 10 mL with double distilled water. Absorption was recorded at 700 nm. Tannic acid was used to calculate the standard curve and the results were expressed as µg of tannic acid equivalents (TAE) per mg of extract.

3. Results and Discussion

1. Extraction Efficiency:

The Extracts obtained Maceration with water and alcohol under different conditions to prevent oxidation of active constituents showed that normal condition extraction with water and alcohol yielded extract and extraction by alcohol using yielded a thick green colour extract with green tea. Different varities of tea showed highest extractive value with water.

2. Phytochemical Screening:

Preliminary phytochemical screening of the different extracts at different varieties of tea reveals the presence of Alkaloids, Saponins, Carbohydrates, Tannins, Amino acids, Flavonoids and Glycosides.

3. Total Phenolic Content:

The Aqueous extract of green tea recorded the highest content of about 59µg/mg of extract. The next highest was observed in aqueous extract of yellow labeled tea. The least content was reported in black tea.

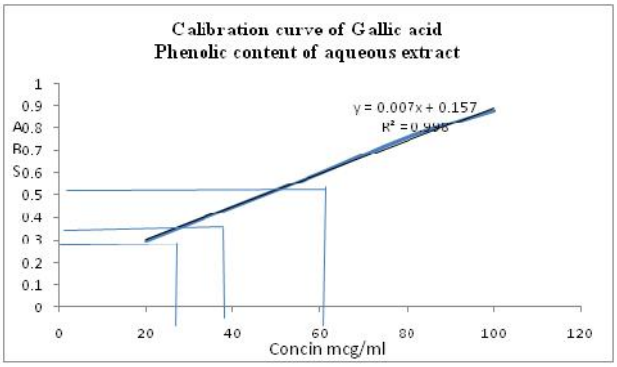

Figure 1: Calibration curve of gallic acid 20-100µg/ml

4. Estimation of Tannins:

The highest value was reported for the aqueous extract of black tea followed by yellow labeled tea, and Green tea.

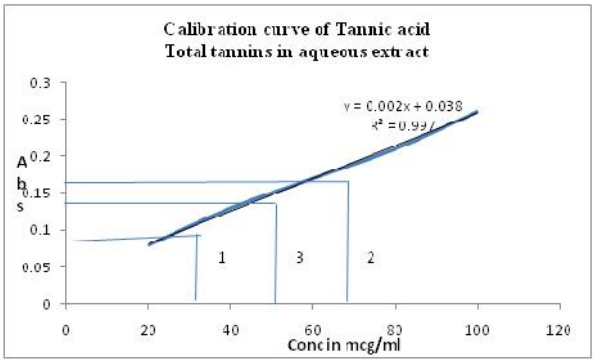

Figure 4: Determination of total tannins in aqueous extracts of tea varities

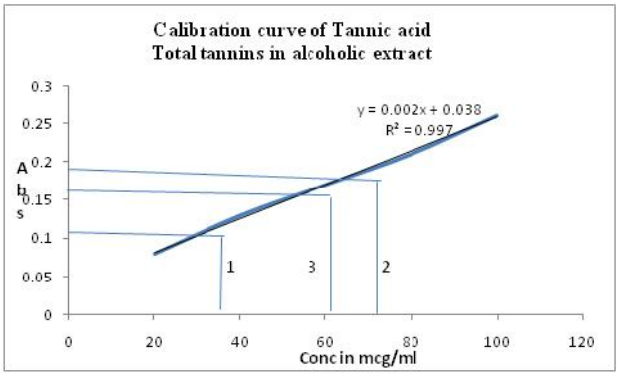

Figure 5: Determination of total tannins in aqueous extracts of tea varities

Discussion:

Phenolics are important plant secondary metabolites with antioxidant activity owing to their redox potential, which play an important role in absorbing and neutralizing free radicals, quenching singlet and triplet oxygen, or decomposing peroxide. In previou studies the aqueous and ethanolic extracts of Tea varities have shown significant antioxidant activity. Antioxidant activity was directly correlated with the amount of total phenolic contents in the extracts. These findings indicate that free radical scavenging in part has immense value in the prevention and treatment of deadly diseases and holds good only if the tea contains phenolics in an appreciable amount .The results obtained a high yield of total phenolics from the green tea pointing that it can be utilised as are markable source for the preparation of not only neutraceuticals as potent antioxidants but also for the treatment of other major health problems. Approaches can be made to identify the individual polyphenolic compounds that are responsible for various pharmacological effects.

Table 1: Extractive values of Different Varities of tea in aqueous media and in alcoholic media

S.No	Name of the Varities of Tea	Extract Extractive Value (%w/w)	
		Aqueous	Alcoholic (Ethanol)
1	Green tea	09.34	09.10
2	Black tea	12.20	10.22
3	Yellow label tea	10.80	09.22

Table 2: Phytochemical screening of different extracts of Tea Varieties

Name of Phytoconstituent	Extract by maceration (Aqueous)			Extract by maceration (Alcoholic)		
	Green tea	Black tea	Yellow tea	Green tea	Black tea	Yellow tea
Alkaloids	+	+	+	+	+	-
Carbohydrates	+	+	+	+	+	+
Amino acids	+	+	+	+	+	+
Tannins	+	+	+	+	+	+
Steroids	+	+	+	+	+	-
Saponins	-	-	-	-	-	+
Flavanoids	+	+	+	+	+	+
Glycosides	+	+	+	+	+	+
Mucilages	+	+	+	+	+	+
Gelatin	+	+	+	+	+	+

Table 3: Total phenolic content of various extracts of varities of tea

S.NO	Tea Varities	Total Phenolic Content* (µg/mg of extract -GAE)	
		Aqueous	Alcoholic
1	Green tea	59	56
2	Black tea	23	34
3	Yellow label tea	38	42

Table 4: Total tannin content of various extracts of Tea varities

S.No	Tea Varities	Total Tannin Content (µg/mg of extract - TAE)	
		Aqueous	Alcoholic
1	Green tea	32	38
2	Black tea	68	72
3	Yellow label tea	54	62

4. Conclusion

The results obtained from this study demonstrated that a considerable variation is present among both various tea samples determined The total phenol content of tea increased significantly (about two times) from green tea to black tea and black tea wastes (fully fermented). Interestingly, tea factory wastes showed good phenolic and tannic acid contents Considering the very low price of wastes, the comparatively clean conditions they are processed and separated from various varities of tea and their significant antioxidant capacity, they could b considered for used in pharmaceutical applications.

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