



## Determination of tannin content by titrimetric method from different types of tea

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### ABSTRACT

Tannins are water soluble polyphenols. Tea leaves are abundant natural sources of polyphenols which get oxidized during processing to prepare the final products. Of the three main types of tea, black tea is produced by complete oxidation (fermentation) while oolong tea is semi-oxidized. Green tea production bypasses oxidation step. Tannins are antioxidant but at high concentration may act as antinutritional. In this study tannin content in various types of tea has been assessed by titrimetric method. Of the tea samples studied black tea was found to contain highest amount of tannin while green tea contained lowest amount of tannin.

**Key words:** Black tea, oolong tea, green tea, tannins, polyphenols

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### INTRODUCTION

Tea is the most widely consumed non-alcoholic beverage in the world. The origin of tea as a beverage is so old that it is lost in legend. Tea is derived from terminal leaves of the shoots from the plant *Camellia sinensis* L. (Kuntz) native to Southeast China. *C. sinensis* var. *sinensis* is indigenous to South east China, Darjeeling and Japan. *C. sinensis* var. *assamica* is indigenous to Assam, Thailand and Sri Lanka [1]. Catechin is the most abundant polyphenol in fresh tea leaves. Three main types of tea are black tea, oolong tea and green tea. Black tea is produced by complete fermentation of tea leaves resulting in black or brown colour of tea where catechins are oxidized by polyphenol oxidases. Oolong tea is basically incompletely fermented black tea. Green tea is produced by inactivating polyphenol oxidase enzymes by steaming or roasting freshly plucked leaves to bypass fermentation step resulting in weaker flavoured green colour of tea.

When any type of tea leaf is steeped in hot water it brews a "tart" (astringent) flavor i.e. dry and pucker feeling characteristic of tannins [2]. Tannins (commonly referred to as tannic acid) are water-soluble polyphenols that are present in many plant foods. Tannins are a diverse class of compounds and have various effects on health. The antioxidant and antimicrobial activities of tannin are well documented. They are also used as antiseptics and astringents. Antioxidant activities confer upon the anti-carcinogenic and anti-mutagenic properties of tannins. Antimicrobial properties of tannins are shown against diverse group of bacteria, fungi and viruses. However, these beneficial effects may be negated by some undesirable attributes. These include precipitation of protein, inhibition of digestive enzymes and prevention of vitamin and mineral utilization [3]. Tannins are often considered antinutritional, as animal studies suggest their role in reduction of net metabolizable energy and protein digestibility [4]. However, the dosage of tannins is critical to these effects. Therefore, foods rich in tannins are considered to be of low nutritional value and are harmful for health [5]. Recent findings indicate that the major effect of tannins is not

due to their inhibition on food consumption or digestion but rather the decreased efficiency in converting the absorbed nutrients to new body substances.

Tea “tannins” are chemically distinct from other types of plant tannins such as tannic acid. Tannic acid is absent in tea extract [2]. In black tea, the major polyphenols are thearubigins and theaflavins [6, 7]. The major theaflavins of black tea are theaflavin, theaflavin 3-gallate, theaflavin 3'-gallate, and theaflavin 3,3'-gallate [8, 9]. Green tea contains mainly flavanols or catechins of epigallocatechin gallate (EGCG), epigallocatechin (EGC), epicatechin gallate (ECG), and epicatechin (EC) [6, 10].

Tea polyphenols are rich natural source of antioxidant. It has broad-spectrum and specific curative effects in antioxidation, anti-atherosclerosis, resistance to dental caries, antitumor, anti-radiation, anti-aging, antimicrobial and in reducing blood pressure, hematic fat, and blood sugar, and even in anti-HIV [11]. Extracts of tea leaves are also sold as dietary supplements [12]. A number of beneficial effects have been attributed to tea, including the prevention of oral cancer and tooth decay [13]. Preliminary results from an intervention study have shown that oral and topical administration of a tea preparation significantly reduced the size of oral lesions and the incidence of micronucleated oral mucosa cells in leukoplakia patients compared with a non-treated group [14]. However, tea during meals significantly inhibits the absorption of both food iron and medicinal iron [4, 15].

Keeping these effects in view, our objective was to analyze tannins contents in several samples of black tea, oolong tea and green tea.

## EXPERIMENTAL SECTION

### Collection of sample

A total of 17 samples belonging to three different types of tea (Table 1) were collected from tea merchants during March 2014 to June 2014 for analysis.

**Table 1. Samples of tea for analysis**

Type	No. of samples	Place of collection	Loose/packed
Black tea (B)	11	West Bengal	Packed (3), Loose (8)
Oolong tea (O)	3	West Bengal (2), USA (1)	Packed
Green tea (G)	3	West Bengal (1), Assam (1), USA (1)	Packed

### Preparation of tea extract

1g of tea sample was added to 25 ml of distilled water and heated in a magnetic stirrer at 70°C for 5 minutes. The decoction was cooled and filtered through Whatman No.1 filter paper. The filtrate was then centrifuged at 10000 rpm for 15 minutes. The supernatant was collected in a sterile clean screw capped tube and stored at 4°C for future analysis.

### Qualitative estimation of tannin

2-3 drops of 5% (w/v) aqueous solution of ferric chloride was added to 1 ml of extract to observe formation of greenish precipitate indicating the presence of tannins in the sample.

### Quantitative estimation of tannin

Quantitative estimation of tannin was performed by titrating the extract with standard potassium permanganate solution following the method of AOAC [16]. (1980). Briefly 5 ml aliquot of the extract was mixed with 12.5 ml of indigo-carmin solution and 375 ml of distilled water. This mixture was titrated against  $\text{KMnO}_4$  solution (“Y” mL). As titration proceeded the blue colour of the indigo-carmin passes through many shades to a final yellow with a faint pink tint at the rim. It was taken as the end-point. This volume of  $\text{KMnO}_4$  was used to titrate total tannin plus all other related compounds. To determine the volume of  $\text{KMnO}_4$  (“X” mL.) used to titrate non tannin (related) compound, another aliquot of 50 ml extract was mixed with 25 ml of gelatin solution (25 g. gelatin was soaked for 1 hr. in saturated NaCl solution. The mixture was then warmed until the gelatin has dissolved and after cooling the solution was made up to 1liter with saturated NaCl), 50 ml of the acidic NaCl solution (25 ml of concentrated  $\text{H}_2\text{SO}_4$  was added to 975 ml of saturated NaCl solution.) and 5 g powdered kaolin. The mixture was shaken for 15 minutes and filtered through Whatman No. 1 filter paper. 12.5 ml of the filtrate was mixed with same volume of indigo carmine solution and 375 ml of distilled water. This mixture was again titrated against  $\text{KMnO}_4$  solution until colour

changed to faint pink as earlier. The volume of  $\text{KMnO}_4$  used to titrate true tannin was calculated by the values of Y and X. The concentration of tannin was estimated using the following relationship:

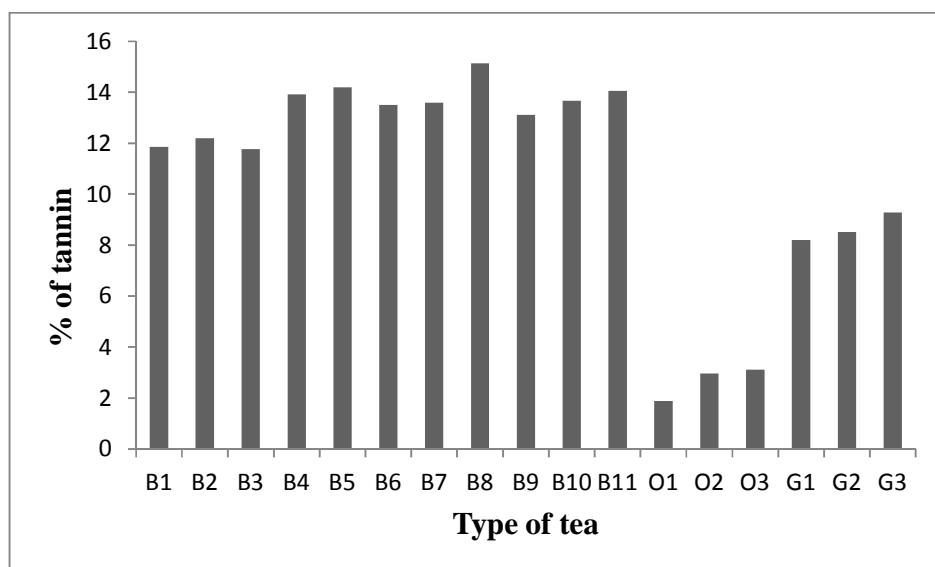
1 ml of standard  $\text{KMnO}_4$  solution = 0.595 ml of 0.1N Oxalic acid

1 ml of 0.1 N Oxalic acid = 0.0042 g of tannin

## RESULTS AND DISCUSSION

Results of the tannin content in different samples of tea have been presented in figure 1.

Figure 1. Percentage of tannin present in different types of tea samples



It was found that samples of black tea had highest tannin content while green tea had lowest tannin content. Tannin content in black tea ranged from 11.76 to 15.14% with an average of 13.36%. Highest tannin content in the studied samples of green tea was 3.11% with an average of 2.65%. Oolong tea had average tannin content of 8.66% lying between black tea and green tea. Tea tannin, different from tannic acid, is a type of polyphenol present in tea leaves. Kaur *et. al.* [17] estimated total polyphenolic content of 10 samples of black tea and 6 samples of green tea. They found that the total polyphenol content in green tea ( $3.066 \pm 1.911$  mg TAE/g) was significantly higher than in black tea ( $0.72 \pm 0.55$  mg TAE/g) ( $p < 0.05$ ). The higher levels of polyphenols in green tea (unfermented tea) than black tea could be due to conversion of the tea polyphenols into thearubigins and theaflavin during the fermentation process of black tea [18]. Tabasum *et. al.* [19] reported lower tannin content (0.18%) in green tea as well as Supreme Brooke Bond tea than in Tapal Danedar (0.38%) and Lipton Yellow label (0.48%) tea while Atanassova and Christova-Bagdassarian [20] reported higher tannin content in green tea (55.89%) than that of in black tea (10.23%). Ushir *et. al.* [5] reported tannin content in four brands of tea viz., Wagh bakri, Rad label, Taj Mahal and Mili at a level of 9.37, 9.82, 7.99 and 7.99%, respectively. These very much differences in tannin contents of different tea samples may be due to the difference in the process of manufacture, aging of tea leaves or the differences in climate and soil texture.

## CONCLUSION

It has been reported that tannin decrease feed intake, growth rate, feed efficiency and protein digestibility. Tannins are known to bind to dietary iron and prevent its absorption specifically of 'nonheme' iron found in plant foods. So it can be concluded that lesser tannin content observed in our study supports the fact that green tea is better for health than black tea and oolong tea.

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