



DETERMINATION OF CAFFEINE CONTENT IN TEA BEVERAGES AND ITS EFFECTS

Chemistry

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ABSTRACT

Caffeine is the main component and characteristic indicator of tea beverages, and it is also a stimulant of the central nervous system. In this paper, the content of caffeine in tea beverage samples in the market were determined by the methylene chloride method, which was convenient, rapid, effective and low cost. The results showed that caffeine was detected in all samples as an inherent component of tea itself. The content of caffeine in the tea samples are vary because of different type of raw tea and the difference in the production process and formula, the caffeine content of the beverages from different manufacturers of the same tea varieties was also significantly different.

KEYWORDS

Caffeine, tea beverages and methylene chloride

INTRODUCTION

Tea is one of the most consumed beverages in the world because of its pleasant taste, aroma, flavor and potential health-promoting properties. It gained its popularity in recent years mostly by the increased range of new flavors in the market and the several studies of tea healthy properties¹. The Botanical name of tea is *Camellia Sinensis*². Tea health benefits have been widely studied and there are scientific evidences that tea intake can reduce cholesterol levels; reduce the risk of developing hypertension, immune disorders and Parkinson's disease; and protect against cancer, cardiovascular and nervous system diseases. Additionally, it consist anti-oxidants, anti-microbial, anti-inflammatory, anti-allergic and anti- obesity effects. Tea is most commonly and widely used soft beverage in the household it act as a stimulant for central nervous system and skeletal muscles. That's why tea removes fatigue, tiredness and headache. It also increases the capacity of thinking. As per history of tea drinking in India, common people used to brew and drink tea using the leaves of the wild native tea plants. Since the time, different varieties of tea have emerged; the most famous among them is the Darjeeling tea.

The important constituent which influence the taste and flavor in tea brew are ploy-phenol, caffeine, sugar, organic acid, volatile flavor compounds and amino acids. Phenolic compounds of such as the a flavins and the arubigins are very important from intrinsic quality point of tea; these are responsible for the colour, flavor and brightness of tea.

The composition of tea varies with type of processing, leaves age, species, origin and climate. Moreover, each type of tea has its characteristic taste, health properties and different level of caffeine.

Green tea is not fermented and it is prepared from young leaves that are exposed to steam and high temperatures with the purpose to inactive the oxidizing enzymes. Black tea is fully fermented and has a high level of caffeine. Mate tea is also known as yerba mate, is a traditional south America tea made from leaves of *Ilex paraguariensis*, and its process involves three steps:- crushing, drying and toasting of the leaves³.

Black tea⁴ has historically been India's most common tea, but there has been a considerable rise in demand for green and white teas in recent years. As a result you can choose from a rich variety of flavors and blends, including spiced chai tea, strong black tea and the fresh and fruity flavored white varieties. While *masala chai* is the perhaps most loved and widely consumed form of Indian tea, there is no end to the

variation available. In fact, different part of country known for their unique brews, with some of the most popular options being Mumbai's cutting chai, the rich Irani chai of Hyderabad, and the delicate pink Kashmiri chai.

Clinical studies have demonstrated that one of the harmful effect of tea is over consumption at least in sensitive people is due to its caffeine content. Caffeine is a methylated xanthine alkaloid derivate identified as 1,3,7-trimethylxanthine and it is naturally present in several foods. According to a caffeine consumption review, it is one of the most consumed dietary ingredients the world for which tea and coffee are the main prominent sources. Caffeine can affect the tea quality characteristics, such as taste, contributing to the acidity and conferring astringency and bitterness. Several studies have reported the ability of caffeine to improve mood, attention, performance, alertness, speed at which information is processed and reaction time.

Moderate caffeine intake may bring some health benefits; however excessive intake of caffeine can increase the risk of tachycardia, dehydration, anxiety, headaches, sleep disorders, nervousness, indigestion and dizziness. Over the years, different studies have been raising concerns related to which ingestion by specific groups, especially pregnant women, children and heavy coffee consumers. In healthy adults, a daily caffeine intake of up to 400 mg per day is not associated with adverse effect.

There are approximately 600 traces of aroma compounds in tea leaves, some of which are lost and some that are released during the manufacturing process. Once plucked, tea leaves begin to wither their cell walls begin to break down and chemical compounds begin to form new chemical compounds not all of which are water soluble.

Carbohydrates, which add to the sweetness of tea help to fuel the enzymatic reaction that take place during oxidation and are also responsible for the correction of polyphenols in young tea leaves.

The term **Polyphenol** refers to a categorization of compounds composes of many phenolic groups-up to 30,000. These compounds are plant metabolites produce as a defense against insects and other animals; such as the alkaloid caffeine and to similar compounds: Theobromines and thophylline, which add to the bitterness of tea alkaloid leaves depend on the varietal and cultivar of the camellia sisensis used along with climate geology geography age of the leaves and the propagation method used. The bud and first leaf have the highest concentration polyphenols 2-3 times more caffeine and

polyphenol leaves decrease in each leaf moving down the plant.

One compound, **Favanoids** (also known as Tannins) are responsible for the health claims of the tea. The major flavanols in tea are: catechin(C), epicatechin (EC), epicatechingallate (ECG), galocatechin (GC), epigallocatechin (EGC) and epigallocatechingallate(EGCG). EGCG is the most active of these catechins and is obtain the subject of studies regarding tea anti-oxidants especially as it relates to cancer research.

The amino acid L-Theanine found in tea has been shown to reduce cognitive stress and used in conjunction with caffeine show a possible improvement in memory and learning tasks.

The consumption volume of tea in India was approximately one billion kilograms during the financial year 2020.

Consumption Of Tea In Different States Of India⁵

Table-1: State Wise And Month Wise Tea Production Data Of Year 2021 – Qty. In M. Kgs.

State	JAN-21*			FEB-21*			JAN-FEB 21*		
	BG	SG	TOT AL	BG	SG	TOT AL	BG	SG	TOT AL
Assam valley	0.00	0.00	0.00	0.19	0.02	0.21	0.19	0.02	0.21
Cachar	0.00	0.00	0.00	0.02	0.00	0.02	0.02	0.00	0.02
Total Assam	0.00	0.00	0.00	0.21	0.02	0.23	0.21	0.02	0.23
Dooars	0.00	0.00	0.00	0.68	0.81	1.49	0.68	0.81	1.49
Tarai	0.00	0.00	0.00	0.14	0.29	0.43	0.14	0.29	0.43
Darjeeling	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Total West Bengal	0.00	0.00	0.00	0.82	1.10	1.92	0.82	1.10	1.92
Others	0.00	0.00	0.00	0.01	0.02	0.03	0.01	0.02	0.03
Total North India	0.00	0.00	0.00	1.04	1.14	2.18	1.04	1.14	2.18
Tamilnadu	4.50	5.93	10.43	4.55	5.69	10.24	9.05	11.62	20.67
Kerala	4.07	1.17	5.24	3.57	1.06	4.63	7.64	2.23	9.87
Karnataka	0.36	0.02	0.38	0.40	0.03	0.43	0.76	0.05	0.81
Total South India	8.93	7.12	16.05	8.52	6.78	15.30	17.45	13.90	31.35
All India	8.93	7.12	16.05	9.56	7.92	17.48	18.49	15.04	33.53

Table-2 Estimated Apparent Consumption Of Tea In India.⁶

YEAR	DOMESTIC CONSUMPTION (Million kg)	YEAR	DOMESTIC CONSUMPTION
2014	927	2014-15	932
2015	948	2015-16	951
2016	965	2016-17	973
2017	1059	2017-18	1066
2018	1084	2018-19	1090
2019	1109	2019-20	1116
2020*	1135	2020-21*	1145

*The actual consumption during 2020 and 2020-21 may vary due to lockdown conditions and availability of tea

Worldwide there are 25000 cups of tea consumed every second, about 2.16 billion cups per day. Today India is one of the largest producers in the world, with over 70% of domestic tea being consumed within India itself. The Indian tea industry has grown to own many global tea brands, and has evolved to one of the most technologically equipped tea industry in the world.

According to National Sample Survey Organization data, India consumes 15 times more tea than coffee. Tea is made both at home and outside the home, tea is most commonly and easily found at the tea stalls about every street of India.

India is the largest producer and consumer of tea in the world, accounts about 29% of world's population. Tea has been accepted all over the world especially in the Indian sub continent as the most favoured beverage. It is popular among all the state of society. India earns substantial revenue in the form of foreign exchange from the export of Indian tea in the world trade.

There is no restriction on export of tea, although import of tea is under the restricted list, but is freely importable from SAARC countries with effect from 1-8-1998.

Up to 400 milligrams of caffeine a day appears to be safe for most healthy adults. That's roughly the amount of caffeine in 4 cups of brewed tea. It's said, it worth nothing that fatal overdoses have been reported with a single doses of 500 mg of caffeine⁷.

Composition

Tea contain catechins a type of anti- oxidant. Tea also contains the amino acids L-theanine which modulates caffeine's psychoactive effect and contributes to tea's umami taste^{9,10}. Caffeine constitutes about 3% of tea's dry weight, depending upon type, brand and brewing method⁸. Tea also contains small amounts of theobromine and theophylline, which are stimulants and xanthenes similar to caffeine. Because of modern environmental pollution, fluoride and aluminum also sometime occur in tea, certain types of brick tea made from old leaves and stem having the highest levels. Although tea contains very poly phenols and tannins, it does not contain tannic acid¹⁰. The principal constituent of tea which is responsible for all these properties is the alkaloid- caffeine. The major constituent of tea is cellulose which is not water soluble. Caffeine is water soluble but so are some tannins and gallic acid which is formed in the process of boiling tea leaves.

Table-3 Caffeine Contents In Different Samples Of Tea¹¹

Sr. No.	Sample Name	Quantity of Caffeine in gms.	Percentage Weight of Caffeine
1.	Lajawab Tea	0.69	1.38
2.	Maharaja Tea	0.61	1.22
3.	Marvel Gold Tea	0.64	1.28
4.	Red Label Tea	0.55	1.10
5.	Swad Gold Tea	0.70	1.40
6.	Swastik Tea	0.74	1.48
7.	Taaza Tea	0.60	1.20
8.	TajMahal	0.58	1.16
9.	Tata Agni Tea	0.66	1.32
10.	Tata Tea Gold	0.62	1.24
11.	Tata Tea Premium	0.56	1.12
12.	WaghBakri Tea	0.54	1.08

MATERIALS AND METHODS

There are many methods of removing caffeine from naturally occurring tea plant products.

a. Methylene Chloride Method

Caffeine can be removed from local tea with the help of methylene chloride. We have soften the material in a water bath or by exposing it to steam, this process of extraction also extract the oil and the flavor and all the good stuff. Take the solution out of the extraction and exposed that to methylene chloride and it removes the caffeine. Now put the solution back again on water to get back oil and flavor. Methylene chloride molecule and caffeine molecule get bonded with each other. So by this process we get decaffeinated tea or coffee¹².

Take 10 mg of tea in 30 ml of water and boiled about 15-20Minutes on burner. Allowed the solution to cool. Poured this solution in dichloromethane, Put the solution in separating funnel to separate caffeine from tea. Now caffeine is extracted from tea with dichloromethane. When the solution of tea in water to regain its flavours and oils. Caffeine is extracted from tea successfully. it will result in bond formation between Caffeine and Dichloro-methane, but it will also extract natural flavours and oils from tea. (Fig.1-4)



Fig.-1

Fig.-2

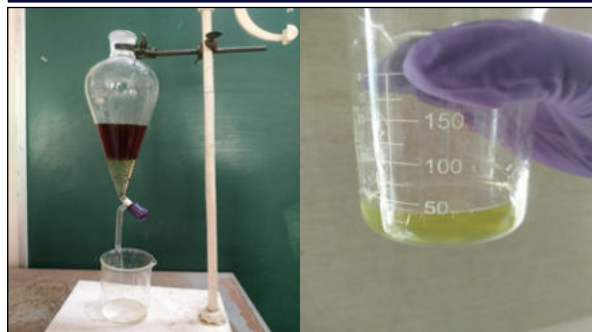


Fig:-3

Fig:-4

B. Thin Layer Chromatography (TLC) –

Different solutes have different solubility in a solvent/different solutes have different degrees of tendency to be dissolved in the same solvent. As the solution (contains the solvents with the dissolve solutes) move along a stationary solid surface, different solutes adsorb on to the solid surface in different extent as they have different degree of adsorption characteristics (due to the different degrees of dissolve tendency). The “less soluble solute” will be retained first, and the “more soluble solute” will be retained after wards.(No two substance have the same solubility and adsorption characteristics). Different solutes will then be separated on the different positions of the solid surface.

OBSERVATION

We have taken 3 different local tea samples of tea Table-4 (Sample 1 from TonkPhatak Jaipur), Sample 2 from Mansarovar (Jaipur) and Sample 3 from Sitapura (India gate, Jaipur). in which we have tried find out Melting point and presence of Caffeine with different Observations.

(Table-4) Observations And Melting Point Of Different Samples Of Tea

S. No.	Sample	Melting Point	Observation	Percentage of Caffeine
1.	Sample 1	226°C	Light Green Colour Present, Caffeine is Present	2.28%
2.	Sample 2	218°C	Light Green Colour Present, Caffeine is Present	2.36%
3.	Sample 3	220°C	Light Green Colour Present, Caffeine is Present	1.78%

RESULT AND DISCUSSION

We have taken 3 samples of tea from different places in Jaipur (Table-4) and we have performed experiment in our lab and we have found that significant amount of caffeine is present in all the 3 samples of tea. Caffeine is very potent, yet unrecognized drug. Although there are beneficial effects to caffeine intake, the negative effect clearly indicates that one should limit their caffeine consumption. Many major organ systems are adversely affected by high amount of caffeine consumption, including the heart, stomach, respiratory and reproductive organs. Caffeine affects our body according to both age and diet. Young children and older people must be much more careful in monitoring in their caffeine intake and should limit themselves to less than 100 mg of caffeine per day. Those in between can take slightly more, but 200 mg of caffeine each day is about the maximum that the average person can ingest without adverse side effects.

Effect Of Tea On Human Health

A. Reduced Iron Absorption

Tea is a rich source of a class of compound called tannins. Tannins can bind to iron in certain foods, rendering it unavailable for absorption in your digestive tract. Iron deficiency is one of the most common nutrient deficiencies in the World, and if you have low iron levels, excessive tea intake may exacerbate your condition. Research suggested that tea tannins are more likely to hinder the absorption of iron from plant source than from animal-based foods. Thus, if you follow a strict vegan or vegetarian diet, you may want to pay extra close attention to how much tea you consumed¹³.

B. Increase Anxiety, Stress And Restlessness

Tea leaves naturally contains caffeine. Over consuming caffeine from

tea, or any other source, may contribute to feeling of anxiety, stress and restlessness. An average cup (240 ml) of tea contains about 11-61mg of caffeine, depending upon variety and brewing method. The longer you steep the tea, the higher the caffeine content. If you notice your tea habit is making you feel jittery or nervous, it could be a sign you have had too much and may want to cut back to reduce symptoms¹⁴.

C. Poor Sleep

Because tea naturally contains caffeine, excessive intake may disrupt your sleep cycle. Melatonin is a hormone that signals your brain that it's time to sleep. Some research suggests that caffeine may inhibit melatonin production, resulting in poor sleep quality. Inadequate sleep is linked to a variety of mental issues, such as fatigue, impaired memory, and reduced attention span. What's more, chronic sleep deprivation is associated with an increased risk of obesity and poor blood sugar control¹⁵.

D. Nausea

Tannins in tea leaves are responsible for the, bitter, dry taste of tea. The astringent nature of tannin can also irritate digestive tissue, potentially leading to uncomfortable symptoms such as nausea or stomach ache. If you notice any of these symptoms after drinking tea, you may want to consider reducing the total amount you drink at any one time. You can also try adding a splash of milk or having some food with your tea. Tannins can bind to proteins and carbs in food, which can minimize digestive irritation¹⁶.

E. Heartburn

The caffeine in tea may cause heartburn or aggravate preexisting acid reflux symptoms research suggest that caffeine can relax the sphincter that separates your esophagus from your stomach, allowing acidic stomach contents to more easily flow into the esophagus. Caffeine may also contributes to an increased in total stomach acid production¹⁷.

F. Pragnancy Complications

Exposure to high levels of caffeine from beverages like tea during pregnancy may increase your risk of complication, such as, miscarriage and infant birth rate. Data on the dangerous of caffeine during pregnancy is mixed, and it's still unclear exactly how much is safe. However, most research indicates that the risk of complications remains relatively low if you keep your daily caffeine intake under 200-300mg.

The total caffeine content of tea can vary but usually falls between 20-60mg per cup (240ml); this is on the side of caution, it's best not to drink more than about 3 cups (700ml) per day. Some people preferred to drink caffeine-free herbal teas in place of regular teas to avoid caffeine exposure during pregnancy. However, not all herbal teas are safe to use during pregnancy¹⁸.

G. Headache

Intermittent caffeine intake may help relive certain types of headache. However, when used chronically, the opposite effect can occur. Routine consumption of caffeine from tea may contribute to recurrent headache. Some research suggest that as little as 100mg of caffeine per day could contribute to daily headache recurrence, but the exact amount required to triggered a headache can vary based on an individual's tolerance. Tea tends to be lower in caffeine than other popular types of caffeinated beverages, such as soda or coffee, but some types can still provides as much as 60mg of caffeine per cup(240ml). If you have recurrent headache and think that may related to your tea intake, try reducing or eliminating this beverage from your diet for a while to see if your symptoms improve¹⁹.

H. Dizziness

Although feeling light-headed or dizzy is a less common side effect, it could be due to consuming too much caffeine from tea this symptoms is typically associated with large doses of caffeine, typically those greater than 400-500mg, or approximately 6-12 cups (1.4-2.4litres) worth of tea. However, it could occur with small doses in people who are particularly sensitive. Generally, it's not recommended to continue that much tea in one sitting. If you notice that you often feel dizzy after drinking tea, opt to for lower caffeine versions or consult your healthcare providers²⁰.

I. Esophageal Cancer

While some studies have associated tea consumption in an increased risk of esophageal cancer, this effect appears due to the scalding beverage, temperature common to these specific cohorts²¹.

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