



E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2020; 9(1): 09-10
Received: 06-11-2019
Accepted: 10-12-2019

Safia Abidi
Department of Pharmacognosy,
Faculty of Pharmacy and
Pharmaceutical Sciences,
University of Karachi, Pakistan

Umer Gilani
Department of Pharmacognosy,
Faculty of Pharmacy and
Pharmaceutical Sciences,
University of Karachi, Pakistan

Rafia Zehra
Department of Pharmacognosy,
Faculty of Pharmacy and
Pharmaceutical Sciences,
University of Karachi, Pakistan

Extraction and analysis of caffeine from various brands of tea leaves marketed in Pakistan

Safia Abidi, Umer Gilani and Rafia Zehra

Abstract

Caffeine is a xanthine alkaloid originate in a variety of sources such as coca seeds, coffee beans, tea leaves. Its content in tea is predictable by methods like spectroscopy, thin layer chromatography, liquid-liquid extraction. In this study, the content of caffeine was estimated using techniques like liquid-liquid extraction and thin layer chromatography. It is the presences of caffeine in tea that keeps us attentive and energetic and facilitates us to work powerfully when consumed in proper amounts. This aromatic complex, on the one hand, can help fight in opposition to diseases like cancer while on the other hand can trouble one's health by increasing heart rate and blood pressure and causing mental ailments like depression and anxiety when addicted in a small amount.

Keywords: Tea, caffeine, liquid-liquid extraction, TLC, purine alkaloids

1. Introduction

Caffeine which is chemically known as 3, 7-dihydro-1, 3, 7-trimethyl-1H-purine-2, 6-dione or 1, 3, 7-trimethylxanthine having chemical formula $C_8H_{10}N_4O_2$ [1] was first discovered in 1827 belonging to the alkaloid family containing nitrogen in their ring structure [2]. Caffeine widely consumed as a stimulant in the world obtain from different sources such as coffee, tea, cola. Tea leaves are the common source from which caffeine was extracted. Commercially three types of tea are available in the market. Black tea is the fermented type of tea, unfermented tea is green tea, and partially fermented tea is oolong tea. In Pakistani culture usually, black tea is utilised [3].

Caffeine which is the purine-based pseudo alkaloids because they were not directly derived from amino acid [4]. Pharmacologically caffeine acts as CNS stimulant, mild diuretic, a natural pesticide, increase blood pressure, increase heart rate, stimulate gastric motility, algicidal, bactericide [5, 6]. Caffeine is incorporate in different pharmaceutical preparation along with acetylsalicylic acid, ascorbic acid, codeine, and paracetamol and other analgesic and antipyretic preparation [7]. The amount of caffeine in different liquids varied in one cup of tea 60-75 mg of caffeine is present the number of other beverages listed in table-01.

Table 1: Amount of caffeine in differently beverages

Beravages	Amount of caffeine (mgs)
Black tea (one tea bag)	25-110
Oolong tea (one tea bag)	12-55
Green tea (one tea bag)	8-30
Decaffeinated tea (one tea bag)	1-4
Coffee (5.oz. cup)	40-170
Soft drink (12 oz. can)	10-50
Energy drinks (12 Oz. can)	75-90

2. Materials and Methods

Five tea samples (01, 02, 03, 04, 05) purchase from local market each weigh 25g is added to boiling distill water 200ml for preparing infusion (the brand name and sample was present in the herbarium of Department of pharmacognosy faculty of pharmacy and pharmaceutical sciences, university of Karachi), lead acetate, 10% sulphuric acid, chloroform and ethanol utilized during caffeine extraction. All the solvents are of analytical grade.

2.1 Procedure

Accurately weighed tea sample 25 g each with the help of electronic balance. Transfer tea leaves into marked separate beakers for each sample add 200ml of distilling water and start boiling for about 10 minutes with constant stirring. Now with the help of sieves, separate teas

Corresponding Author:
Safia Abidi
Department of Pharmacognosy,
Faculty of Pharmacy and
Pharmaceutical Sciences,
University of Karachi, Pakistan

leave and collect infusion in a separate beaker. Now repeat the above procedure two more times by using the same tea leaves. Collect all the filtrates in the same beaker and add lead acetate in order to precipitate tannins which are present in the tea infusion. Again heat the mixture of tea infusion and lead acetate to complete the process of precipitation. Now filter it with the help of wattman filter paper to separate the precipitated tannins from the infusion. Do washing of tannin precipitate in order to make sure that no caffeine residue left on filter paper. Add 2ml of 10% sulphuric acid into the filtrate.

Now transfer the filtrate into separating funnel and add 30ml of chloroform do proper mixing by inverting the separating funnel, so many times release air and allow to stand to separate caffeine now collect the organic layer same procedure is repeated twice collect all the sample in the same beaker.

For the evaporation of the solvent placed it overnight in the fuming hood next day observe the caffeine crystal now weigh it qualitative analysis of caffeine is carried out.

2.2 Identification of caffeine crystal

Thin layer chromatography adopted for the confirmation of caffeine crystal. Chloroform and methanol, the ratio of 9:1 used as a solvent. By following the procedure of thin layer chromatography, the spots visualised under ultraviolet light at 254 nm wavelength. Mark the spots and calculate the Rf value of caffeine [8, 9, 10].

Table 2: Weight by weight percentage of caffeine content of the various brands of tea

Sample #	% of Caffeine content/25 G OF tea leaves
01	1.1%
02	0.8%
03	0.8%
04	2%
05	0.9%

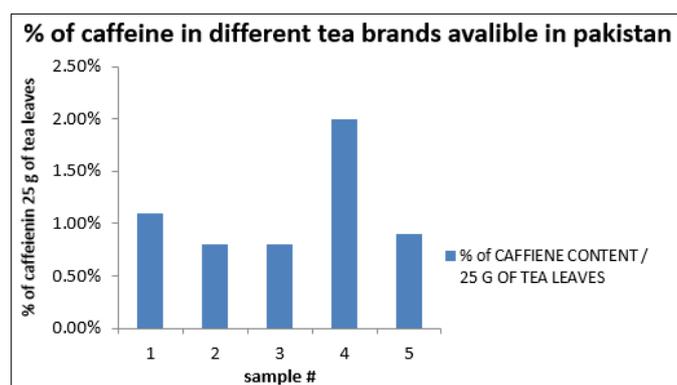


Fig 1: Percentage of caffeine in different kinds of tea

Table 3: Rf Value of isolated caffeine from different tea brands

Sample #	Isolated caffeine Rf value
01	0.79
02	0.8
03	0.76
04	0.8
05	0.8

3. Results and Discussion

Caffeine content among five brands is varying. The % w/w of caffeine content is mention in table 2 and figure 1. The highest caffeine content was present in sample #04, and the

lowest is in sample #02. The extracted caffeine was identified by thin layer chromatography (TLC) by comparing the Rf values of sample and reference which is mention in table 2 it was confirmed that the extracted material is caffeine. Caffeine (1, 3, 5-trimethylxanthine), a mild addicting drug through used for medicinal purposes is the active ingredient that makes tea valuable to humanity. In this study, the levels of caffeine in certain tea brands found in the Pakistan market were determined using extraction and crystallisation method. The order of caffeine concentration in tea samples found as follow (04>01>05>03>02) the level of caffeine in all tea brands are found within the range. Caffeine in low quantity keeps us alert and active and enhances our performance. Increase the speed of perception and decision making; also help in weight loss and cancer prevention. However, in high dose cause insomnia, rising blood pressure, anxiety.

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