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Research Paper

Comparative Study on the Isolation and Analysis of Caffeine from Diverse Sources

Prof. Ganesh A. Borawake

Department of Chemistry, Jijamata College of Science and Arts, Bhende, 414605, MS, India

Address correspondence to E-mail: ganesh.borawake77@gmail.com

Abstract

Caffeine is a naturally occurring alkaloid widely distributed in beverages and plant-derived products such as tea leaves, coffee beans, and green tea. The present study focuses on the comparative extraction and analysis of caffeine from different natural sources using efficient and reproducible laboratory methods. Caffeine was isolated through solvent extraction techniques involving aqueous infusion followed by organic solvent separation. The extracts were purified and characterized using physicochemical methods such as melting point determination, UV-Visible spectroscopy FT IR, and thin-layer chromatography to confirm identity and assess purity. Quantitative estimation was carried out to compare caffeine content among the selected sources. The results demonstrated noticeable variation in caffeine yield depending on the plant matrix and processing conditions. This study highlights the effectiveness of conventional extraction methods and provides insight into source-dependent differences in caffeine concentration. The findings contribute to a better understanding of caffeine recovery techniques and support further research on optimizing extraction efficiency for food, pharmaceutical, and analytical applications.

Introduction

Introduction Caffeine is a naturally occurring chemical stimulant found in numerous plants' leaves, seeds, and fruits. Caffeine is the world's most widely consumed psychoactive drug; however, it is legal and unregulated in nearly all parts of the world. It stimulates the central nervous system, reducing fatigue and drowsiness. It can improve athletic performance, muscular strength, and power. Caffeine riches 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 heterocyclic ring structure [2]. Coffee originated in China and was the sole producer until the British Empire colonised India and began large coffee plantations. Coffee drinking is still an important ritual in Japan and England, from the choreographed Japanese coffee ceremony to the daily coffee time held in homes and offices across Brazil. Currently, Brazil is the highest consumer of coffee in the world. Caffeine is a naturally occurring substance found in the leaves, seeds, or fruits of more than 63 plant species worldwide. The most common sources of caffeine are coffee, cocoa beans, cola nuts, coffee leaves, yerba mate, guarana berries and the yaupon holly. The amount of caffeine in coffee varies by brand, but the average in common brands is typically about 30 - 40 mg caffeine/coffee bag. Caffeine is the purinebased Pseudo alkaloids because they were not directly derived from amino acid [3]. Pharmacologically, caffeine acts as a CNS stimulant, mild diuretic, and a natural pesticide, increasing blood pressure, increasing heart rate, stimulating gastric motility, algicides, and bactericides [4, 5]. Caffeine was first isolated from coffee in 1819 by a German chemist, Friedlieb Ferdinand Runge. Its pure form is an odourless white solid with a melting point of 235° - 238° C. Caffeine is classified as a heterocyclic purine base with two or more nitrogen elements in its rings. Caffeine is weakly basic, and its solubility in water at room temperature is quite low (2g/100ml) but increases dramatically in boiling water (66g/100ml).

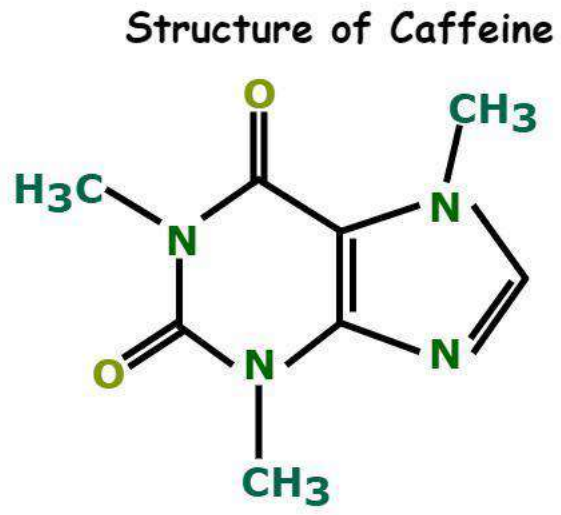
The primary methylxanthine present in tea is the stimulant caffeine. Other methylxanthines occurring in tea include two structurally related compounds, theobromine and theophylline, which significantly contribute to the widespread and lasting popularity of non-alcoholic beverages and food products such as coffee, tea, cocoa, chocolate, and various soft drinks. It is often used in combination with analgesics (such as aspirin or acetaminophen) and a compound known as ergotamine for the treatment of migraine headaches.

Caffeine is widely consumed to enhance mental alertness, but it also has numerous additional applications. The diet supplement and cosmetic industries are among the major users of caffeine. It is utilized in the production of body wash, soap, lip balm, facial scrubs, and other products such as caffeine-infused lipstick. In pharmaceutical formulations, caffeine is combined with acetylsalicylic acid, ascorbic acid, codeine, paracetamol, and other analgesic and antipyretic agents [7].

The caffeine concentration in food and beverage products differs according to serving size, product category, and preparation technique. Tea, a commonly consumed beverage, is prepared from the leaves of the Asian evergreen plant Camellia sinensis. The caffeine level in tea leaves varies depending on the cultivar and cultivation region; typically, tea contains about 3-5% caffeine by weight [8].

The optical absorption characteristics of caffeine have been evaluated in various solvents, including dichloromethane, water, chloroform, and ethyl acetate. Among these, dichloromethane exhibits the highest optical transition values. Caffeine extraction is more efficient at boiling temperature compared to 30°C. It can be isolated from cocoa using different techniques such as aqueous extraction, supercritical carbon dioxide extraction, and organic solvent extraction [9]. Common solvents employed for caffeine extraction include chloroform, methylene chloride, ethanol, acetone, and ethyl acetate [10].

Several extraction approaches are available, including Soxhlet extraction, ultrasonic-assisted extraction, and heat reflux extraction [11,12]. Heat reflux extraction is a frequently applied laboratory-scale technique for isolating caffeine from cocoa beans.



Adventages of Caffeine 1. Enhanced Mental Alertness and Focus • Improved concentration :- Caffeine helps increase alertness and sharpens focus, making it easier to stay attentive, particularly in situations that require prolonged concentration. • Memory improvement :- Some studies suggest that caffeine can enhance short-term memory, improving learning performance in the short run. 2. Boost in Physical Performance • Increased stamina :- Caffeine stimulates adrenaline production, which can improve physical endurance, making it a popular aid for athletes and fitness enthusiasts. • Reduced perception of effort:- It reduces the feeling of fatigue, making physical activities seem less tiring. 3. Mood Enhancement • Improved mood :- Caffeine can increase the levels of dopamine and serotonin, the brain chemicals responsible for mood regulation. This can lead to feelings of happiness and well-being. • Reduced risk of depression: Some research suggests that moderate caffeine intake is associated with a lower risk of depression and a better overall mood. 4. Cognitive Protection • europrotective benefits :- Long-term consumption of caffeine has been linked to a reduced risk of developing neurodegenerative diseases such as Alzheimer's and Parkinson's. It may help protect brain function as you age. 5. Faster Metabolism • Increased calorie burn:- Caffeine can boost metabolism by 3-11%, promoting fat burning in the body. This can make it helpful for weight loss when combined with a healthy diet and exercise. • Appetite suppression*: It can act as a mild appetite suppressant, which may help with controlling calorie intake. 6. Pain Relief • Headache reduction: Caffeine has vasoconstrictive properties, meaning it narrows blood vessels, which can help relieve headaches or migraines. It's commonly found in pain relief medications for this reason. • Post-workout muscle recovery: Caffeine may reduce the perception of muscle pain and improve recovery after intense workouts. 7. Enhanced Reaction Time and Accuracy • Better decision-making: In tasks that require quick decisions or fast reflexes, caffeine can improve reaction times and make performance more efficient, especially in low-energy situations. 8. Antioxidant Properties • Rich in antioxidants: Coffee and tea, two common sources of

caffeine, are also rich in antioxidants that help reduce inflammation and may lower the risk of chronic diseases such as certain types of cancer and heart disease. When used in moderation, caffeine can be a helpful tool for improving mental and physical performance, enhancing mood, and even offering some long-term health benefits

Disadventages of Caffeine 1. Sleep Disruption • Caffeine can interfere with the ability to fall asleep or stay asleep, leading to poor sleep quality and insomnia. 2. Anxiety and Jitters • High doses of caffeine can cause feelings of nervousness, restlessness, and an increase in heart rate, which may contribute to anxiety. 3. Dependence and Withdrawal • Regular caffeine consumption can lead to dependence. When trying to reduce or stop caffeine intake, withdrawal symptoms such as headaches, fatigue, irritability, and difficulty concentrating may occur. 4. Digestive Issues • Caffeine can stimulate the production of stomach acid, which may cause acid reflux or worsen conditions like gastritis or ulcers. 5. Increased Blood Pressure • Caffeine can cause a temporary increase in blood pressure, which might be concerning for individuals with hypertension or heart problems. 6. Dehydration • Caffeine has a diuretic effect, meaning it can increase urine production and potentially lead to dehydration if fluid intake isn't adequate. 7. Addiction Potential • The regular use of caffeine can lead to psychological dependence, where a person feels they need it to function normally.

Experimental

Materials And Chemicals

Extraction and Characterization of Caffeine From Different Sources.

Chemicals:

Tea leaves, Coffee powder, APC tablet, Green tea, Dichloromethane, lead acetate (10%), sulphuric acid (2 N), activated charcoal, petroleum ether, calcium carbonate, acetone, sodium sulphate.

Glassware and Apparatus:

Separating funnel, filtration setup, beakers etc.

Procedure:

Caffeine is a minor constituent of tea, coffee, and other natural plant materials. 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. The latter two components can be converted to their calcium or lead salts which are insoluble in water. The caffeine can then be extracted from the water by Dichloromethan in almost pure form.

Procedure I: Extraction using water, Lead acetate and chloroform

1. Boil 25 gm of powered leaves in 250 ml of water in a beaker for 15-20 min. Filter the aqueous solution while hot. Collect the filtrate in beaker.
2. Boil the residue on filter paper again with water (100 ml) for 15-20 minutes and Filter the aqueous solution while hot. Collect the filtrate in same beaker.
3. Combine both the aqueous solution and add lead acetate solution (10%, 55 ml) slowly until no more precipitate is formed. Filter the solution while hot to separate the precipitate. The precipitated material consists of naturally occurring tannins (poly phenols). Collect

1. the filtrate in beaker.

4. To the clear solution (filtrate) add dilute sulphuric acid until the precipitation of lead as lead sulphate is complete. Filter the precipitate of lead sulphate
5. Boil the filtrate with a little activated charcoal, filter and concentrate on a sand bath to
2. about 50 ml. 6. Cool the solution to room temperature and extract with chloroform (3 x 30 ml). Combine 13 the chloroform layers and dry using sodium sulphate.
6. Remove the chloroform from the dried solution by distillation using water bath. The residue of crude caffeine will obtain.
7. Add petroleum ether (30 ml) to the residue and stir the mixture. Filter the solid product of crude caffeine.
8. Recrystallize the extract caffeine using and petroleum ether. Record its yield M.P and TLC.

Solvent Extraction (Liquid–Liquid Extraction):

This is one of the most widely used methods in laboratories for isolating caffeine. In this technique, an aqueous solution containing caffeine (such as brewed tea or coffee) is shaken with an immiscible organic solvent like chloroform, dichloromethane, or ethyl acetate. Caffeine preferentially partitions into the organic phase. The organic layer is then separated, dried to remove moisture, and the solvent is evaporated to recover caffeine.

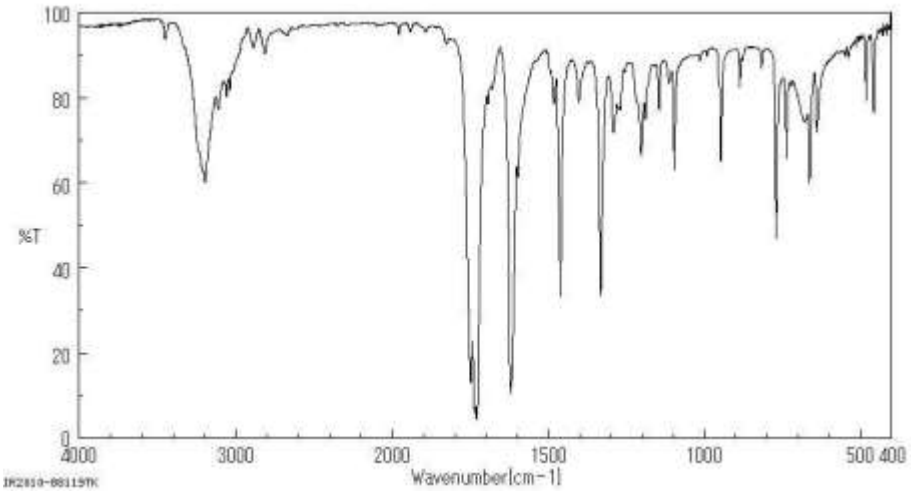
Advantage: The method is straightforward and efficient.

Disadvantage: It involves the use of potentially hazardous organic solvents.





Characterization



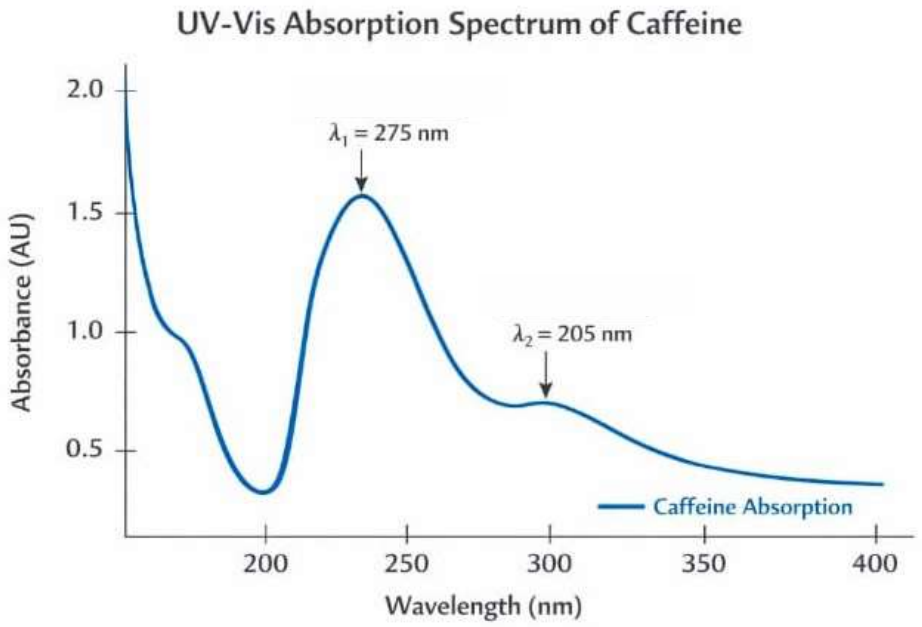
FT-IR Spectra of Caffeine

Caffeine ($C_8H_{10}N_4O_2$) is a methylated xanthine derivative containing two carbonyl (C=O) groups, C=N bonds, C-N bonds, and methyl (-CH_3) groups. Its FT-IR spectrum shows characteristic absorption bands due to these functional groups.

Important FT-IR Peaks of Caffeine

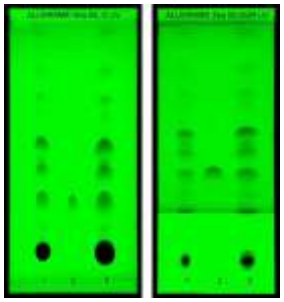
Wavenumber (cm^{-1})	Functional Group	Conclusion
3150–2950	=C–H stretching	Aromatic/heterocyclic C–H stretch
1705-1660	C=O stretching	carbonyl absorption
1610–1560	C=N stretching	Heterocyclic ring vibration
1500–1450	C=C stretching	Ring skeletal vibration
1310–1210	C–N stretching	Tertiary amine vibration
1100–950	C–N / ring vibrations	Fingerprint region

Two strong C=O peaks confirm the presence of two carbonyl groups.



UV-vis absorption of caffeine in water and dichloromethane

The UV-vis absorption spectrum of caffeine in water is found to be in the region of 243-302 nm at room temperature. It is clearly shown in Fig. 1 that the spectral intensity of caffeine drops to zero at wavelength greater than 302 nm, on the other hand a new peak absorbance is noticed at a wavelength below 243 nm. This new spectrum is expected to be the peak absorbance due to the solvent. The peak absorbance of the solution is found to be A = 1.224 at the maximum wavelength λmax=272nm.



Thin Layer Chromatography (TLC) is one of several chromatographic techniques, along with paper chromatography and gas chromatography. All these methods are based on a common principle: different solutes vary in their solubility in a given solvent and in their ability to adsorb onto a stationary phase.

In TLC, a solution containing dissolved substances moves upward over a solid stationary surface. As the solvent travels, the individual components interact differently with the solid surface because each has a unique balance between solubility in the solvent (mobile phase) and adsorption onto the solid (stationary phase). Substances that are less soluble in the solvent or more strongly adsorbed onto the solid surface move more slowly and remain closer to the baseline. In contrast, substances that are more soluble in the solvent travel further up

the plate. Since no two compounds have identical solubility and adsorption properties, they separate at different positions on the stationary phase.

The retention factor (Rf) for each component is calculated using the formula:

Pure caffeine and the extracted sample were applied to the same TLC plate, and their Rf values were compared to determine any differences between them.

Physical Properties

Here are some physical constant of caffeine:

- Molecular mass: **194.194** g/mol
- Density: **1.23 g/cm3** in solid form
- Melting point: **227-228°C** for anhydrous form, **234-235°C** for monohydrate form
- Boiling point: **178°C**
- Solubility: Soluble in water, milk, and other substances
- Ultraviolet absorption spectrum: Maximum at **274** nm and an absorption coefficient of **9700** in aqueous solution

Caffeine is a white crystalline substance that is commonly found in the form of fine needles or powder.

It is a naturally occurring alkaloid and stimulant compound with the chemical formula $C_8H_{10}N_4O_2$.

Caffeine is found in various , such as coffee, sting, tea leaves, green tea, and APC Tablet.

Result & Discussion

Source	Caffeine per Serving
Green Tea (1 Cup)	30 mg
Black Tea (1 Cup)	50 mg
Instant Coffee (1 Cup)	80 mg
Sting (250 ml Bottle)	85 mg
APC Tablet (1 Tablet)	40 mg

Green Tea = Low Caffeine

Herbal Tea = No Caffeine

Conclusion

caffeine content measured in all brands was found to lie within an acceptable range. In moderate amounts, caffeine promotes alertness, increases activity levels, and improves overall performance. It enhances perception and accelerates decision-making, and it may also contribute to weight management and potential cancer-preventive effects. However, excessive intake can lead to adverse effects such as insomnia, elevated blood pressure, and anxiety. Tea and coffee, among the most widely consumed beverages globally, are rich sources of antioxidants and possess notable medicinal properties. Caffeine, the world's most extensively used psychoactive substance, exhibits pharmacological activity and functions as a mild central nervous system stimulant, depending on the dosage.

The isolated caffeine was subsequently characterized using analytical techniques such as IR UV vis,Thin Layer Chromatography (TLC), Gas Chromatography (GC), iodometric and the spike test. Owing to concerns regarding its possible pathogenic effects, caffeine has become one of the most thoroughly investigated compounds. The findings indicate that coffee

contains a comparatively higher concentration of caffeine than other beverages. Furthermore, caffeine demonstrates greater solubility in dichloromethane than in many other solvents.

References

1) Wanyika, H. N., 2010 "Determination of the caffeine content of tea and instant coffee brands found in the Kenya market", Afr. J. Food Sci. 4(6), 353-358,
2) Halligudi,N., 2012."Analysis of various brands of tea powder marketed in Oman",International Journal of Nutrition Pharmacology, Neurological Diseases, 2(2),
3) Cordell, G.A.,1981. "Introduction to alkaloids", Jhon Wiley, Sons, Inc. Canada, 6
4) JinnoD., 1996. "Comprehensive medical chemistry", Pergamon Press
5) Eaton, K., 2010. "Caffeine could be helpful Las Vegas Review-Journal", 23,
6) Rebecca, Jeyanthi & Seshiah, Candace & Tissopi, Trisha, 2014."Extraction of caffeine from used tea leaves", The Annals of "Valahia" University of Targoviste, 9
7) Hamad,M.N., 2010. "Gravimetric estimation of caffeine in different commercial kinds of tea found in the Iraqi Market", Iraqi J. Pharm. Sci., 19(2),
8) Venkatesh, S.,Swamy, M. M., Reddy,Y. S. R., SureshB. and Sethuraman,M.,1994. "A Simple method for determination of caffeine content in tea samples" Journal Ancient Science of Life, 14(1-2), 35-38,
9) Aniket Chaugule, Hitesh Patil, Shreyans Pagariya, and Pradnya Ingle, 2018."Extraction of caffeine: A Review", Journal of emerging technology and innovative research, ISSN- 2349-5162, 5(9)
10) Postu, A. and Wilson,S., 2013. "Isolation of caffeine from tea leaves via Acid-Base Liquid-Liquid extraction", Asian Journal of Science and Technology, 12(2), 2157,
11) Anbuselvi,S., Sarvanthi, S. and Nikhil Kumar,M. A., 2017. "Extraction of caffeine from different variety of tea samples", International Journal of Pharmacy and Technology, Coden-IJPTFI,
12) Sevdar,G., Demir, E. and Sokmen, M., 2015. "Comparison of some extraction methods for isolation of catechins and caffeine from Turkish green tea", International Journal of Secondary Metabolite, 2, 16-25.