

Determining the Amounts of Caffeine in Foreign Black Teas

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Abstract

This study examines the efficiency of caffeine extraction and brewing across five internationally sourced black teas—British, Indian, Yemeni, Turkish, and Chinese—through controlled experimental trials. Caffeine was extracted using liquid-liquid extraction with dichloromethane and quantified via UV-Vis spectrophotometry to assess solubility, yield, and extraction kinetics. Indian tea showed the highest average yield (480.0 mg/min) and caffeine concentration (44.79 mg/g) during trials 2 and 3. Overall extraction rates ranged from 271.0 to 524.6 mg/min. Brewed tea pH values (4.0–5.0) suggest that mild acidity may influence caffeine solubility. High percent errors (~99%) indicate significant deviations from theoretical values, likely due to procedural variability and instrumental limitations such as solvent evaporation and calibration drift. Despite these errors, the relative ranking of caffeine concentration remained consistent across all teas, demonstrating the robustness of inter-tea comparisons. This research contributes to the field of beverage chemistry by offering a detailed methodology for analyzing caffeine content and optimizing brewing conditions.

Background

Caffeine, which enhances alertness by affecting neural activity, is found in black tea. Due to environmental factors like climate, geography and soil composition, caffeine content in black teas vary. Quantifying caffeine content provides insight to plant growth conditions and raises awareness of caffeine consumption, as health risks can be present with excessive intake, essential for making informed choices and managing intake, especially for individuals who are sensitive to caffeine.

When the caffeine content is isolated from tea, partition coefficients are referred to within the ratio of caffeine concentration in dichloromethane to its concentration in liquid black tea, indicating the quantified caffeine content will preferentially dissolve in the organic layer, compared to the water layer, allowing for efficient caffeine extraction from the black tea.

Partition Coefficient (K) = [Caffeine Concentration in organic phase] - [Caffeine Concentration in Aqueous Phase]

1. Molecular Structure of Caffeine



Due to its molecular structure, Caffeine absorbs ultraviolet light at 273 nm through the Beer-Lambert Law, describing

the linear relationship between the absorbance and concentration. The higher the caffeine concentration, the higher the absorbance recorded by the UV-Vis spectrophotometer.

Beer-Lambert Law Equation:

- $A = \epsilon lc$
- A = Absorbance
- ϵ = Molar absorptivity constant (100 L mol⁻¹ cm⁻¹ for caffeine at 273 nm)
- l = Path length of the cuvette (1 cm)
- c = Concentration of Caffeine (mg/L)

Memon and Idress (2024) optimized caffeine extraction from tea using dichloromethane, finding longer extraction and higher temperatures, improved efficiency up to 30 minutes and 100°C, yielding 0.089 g of caffeine from black tea. Another study made the process more simpler and accessible for education use through hot water extraction.

Within this experiment, the amounts of caffeine (mg) in foreign black teas from India, England, China, Turkey, and Yemen will be investigated, using Liquid-Liquid Extraction Method, allowing for differentiation of caffeine content world-wide. Quantified caffeine content in international teas are expected to change with the variations in origin. The control variables must be maintained in order to ensure that external variables do not hinder the results.

Independent variable	Type of Black Tea Powder (Origin)
Dependent variable	Mass of Caffeine Content Extracted from the Black Teas
Controlled variable(s)	Mass of tea powder, Volume of water used for brewing, Brew temperature (e.g., always 90-95°C), Brew time (e.g., always 5 min), Volume of dichloromethane used for extraction (e.g., always 25 mL) and Same spectrophotometer and wavelength (273 nm) for all measurements.
Research Question	How can the Caffeine Content be Quantified using Liquid-Liquid Extraction in Black Teas Originating from India, China, England, Yemen and Turkey?

It can be hypothesized that black teas from different countries will have varying levels of caffeine content because tea leaves grown in different countries can have factors that influence the composition of leaves. Differences in climate can affect the caffeine concentration. Different countries can also have different soil types containing various minerals. Black tea plants grown in fertile volcanic soil can produce more caffeine than those grown in nutrient poor soil.

Differences in caffeine content cross-continental, presented genetic variations in *Camellia sinensis* influence caffeine synthesis. Growing techniques like fertilization, pruning and shading, as well as post-harvest processes such as drying, withering and oxidation also influence caffeine levels. Although climate impacts photosynthesis and caffeine production, pH, mineral activities affect plant metabolism.

2. Procedure

Step 1: Preparing the 1000 mg/L Caffeine Stock Solution:

- Carefully crush one 200mg caffeine tablet into a powder substance using a mortar and pestle.
- Use an analytical balance and weighted boat to weigh out half of the tablet, containing 100 mg of caffeine content. Measure the actual amount of the tablet.
- Place the crushed half tablet into a graduated cylinder

for precision and pour around 10 ml of deionized water into it in order for it to dissolve.

- Place the solution into a 100 ml volumetric flask and fill it to the meniscus with deionized water.

Step 2: Preparing the Caffeine Standards:

- Take 5 clean 100 ml volumetric flasks; label them each 10 mg/L, 20 mg/L, 30 mg/L, 40 mg/L and 50 mg/L.
- For 10 mg/L → Pipette 1.0 mL of the stock solution into the flask.
- For 20 mg/L → Pipette 2.0 mL of the stock solution into the flask.
- For 30 mg/L → Pipette 3.0 mL of the stock solution into the flask.
- For 40 mg/L → Pipette 4.0 mL of the stock solution into the flask.
- For 50 mg/L → Pipette 5.0 mL of the stock solution into the flask.
- Fill each flask to the 100 ml mark with deionized water.
- Swirl the flask gently to mix everything evenly.
- Place each stock solution into a different quartz cuvette (¾) because of plastic absorption.
- Measure its absorption using the UV-IS spectrophotometer.



Step 3: Brewing the Teas:

- In a 250 ml beaker, add 150 ml of deionized water
- Measure 8.0g of each tea with uncertainty into each beaker using an analytical balance and weighted boats
- Place the tea into the 150 mL of water
- Heat up the water bath to 90-95 degrees celsius and place the beakers inside of them, allowing them to each heat up for around 5 minutes each
- In the bath, stir the beakers gently
- After heating, use filter paper, rubber bands and spoons to separate the tea solution from the tea particles
- After, set aside the tea solution in a separate beaker and make sure it is labeled the country that it is from
- Repeat this step for each black tea



Step 4: Finding the pH:

- Dip the stirring rod into the tea solution
- Dab it to the pH strips and wait around 5 minutes to ensure accurate results
- Compare the strip color to the colors on the pH measurement kit
- Repeat this step for each black tea



Step 5: Extracting Caffeine Using Liquid-Liquid Extraction:

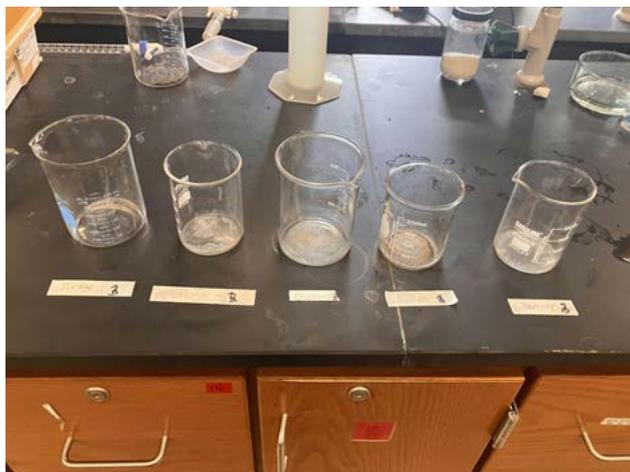
- Transfer 50 mL of each tea solution into a separatory funnel
- Add 10 mL of sodium carbonate solution to neutralize acidic compounds. This should cause the color of the solution to become darker.
- Add 25 mL of dichloromethane to the separatory funnel
- Shake gently and let the layers separate (caffeine moves into organic layer)
- Drain the bottom layer/organic layer into a beaker
- Evaporate the dichloromethane using a water bath, leaving the solid caffeine
- Weight the extracted caffeine using an analytical balance
- Repeat for each black tea

Step 6: Measuring the Amount of Caffeine:

- Take the dried caffeine from the beaker where the dichloromethane was evaporated and pour 3 mL of deionized water into the beaker.
- Swirl it around and wait for the particles to disintegrate, until the solution becomes pure liquid
- Pour 10 mL of deionized water into the beaker and pour

that solution into the volumetric flask

- Pour 25 mL of dichloromethane into the volumetric flask and swirl it very well
- In between swirling, vent the air by turning the opening of the flask
- Do this process a few times and let the volumetric flask sit for a few minutes
- After, filter out the bottom layer of dichloromethane into a smaller beaker and set it aside
- After it is dried, the white follicles on the sides and inside



of the beaker are the caffeine contents

- Place around 1 mL of water inside of the beaker and mix it until it is a solution, making sure the sides of the beaker filled with caffeine are also within the solution.
- Next, place the solution into a quartz cuvette and add deionized water in order to dilute the solution
- Finally, calibrate the spectrophotometer, recording measurements.
- Conduct three trials



Independent Variable: Type of Black Tea Powder

Dependent Variable: Mass of Caffeine Content Extracted from the Black Teas

Control Variables:

- Mass of tea powder
- Volume of water used for brewing
- Brew temperature
- Brew time
- Volume of dichloromethane used for extraction
- Same spectrophotometer and wavelength for all measurements.

Materials:

- 5 Black Teas from different countries
- Dichloromethane
- Deionized water
- UV-Vis spectrophotometer
- Caffeine tablets
- 250 mL Beakers (± 1)
- 100 mL Volumetric flasks (± 0.1)
- 10 mL Graduated Cylinders
- Water Bath
- Weighted Boats
- Filter paper
- Rubber Bands
- pH strips
- Analytical Balance (± 0.001)
- Separatory Funnel (± 0.5)
- Funnel
- Pipette (± 0.01)
- Stirring Rod
- Sodium Carbonate
- Electronic Device

- Mini Beakers (± 10)
- Labeling Tape

The Liquid-Liquid Experimentation is best for this procedure because caffeine is more soluble than other solvents compared to water, which allows for a selective extraction where caffeine is transferred to the organic phase when mixed with the tea solution, leaving behind various tea components in the aqueous phase. It is best for separating solutions and particles from each other through selectivity and uses solubility differences. Dichloromethane is used due to its selective solubility, allowing neutral caffeine to transfer organic phase with sodium carbonate, leaving other components in the aqueous layers. , Google spreadsheets were used to minimize human errors, while data was organized in tables for easy adjustments and clarity. , A linear fit was chosen for the calibration curve because of its high R2 value, confirming a strong linear relationship suitable for quantifying caffeine. According to the Beer-Lamert Law, absorbance is proportional to concentration within the measured range.

3. Safety Concerns

- Use heat protectant gloves when handling the water bath and hot beakers
- Make sure to wear safety goggles when using the dichloromethane
- DO NOT inhale the dichloromethane, as it can cause dizziness, drowsiness, headaches and nausea
- Do not leave the tea solution out for several consecutive days, as mold has the potential to build

4. Analysis and Conclusion

How much Caffeine Content is there in Foreign Black Teas from Indian, England, Yemen, Turkey and China? India and Yemen consistently had high caffeine concentration and mass extraction levels, suggesting a higher partition coefficient, indicating it is more soluble in the organic phase. The Chinese tea has low levels of caffeine yield/extraction, suggesting lower natural caffeine content. British and Turkish teas had presented intermediate levels. For caffeine content per gram, Indian and Yemen black teas had higher levels, which supported the idea that *Camellia* grown in warmer climates have more caffeine content. However, the Chinese black tea had lower caffeine levels, which remained consistent with the literature value.

The Turkish and Indian black teas presented stable solubilities, suggesting that there was a consistent caffeine extraction. Due to experimental loss and potential contamination, Yemen black tea in trial 2 had the highest percent yield of 110.3%, while the British and Chinese black teas had the lowest percent yields, presenting potential extraction inefficiency and reinforcing their lower caffeine content. However, Indian and Turkish teas had the most effective extraction because their yields were close to 100%. The slowest caffeine rates were from the Chinese tea, aligned with their low caffeine concentration, while the fastest caffeine rate was by the Yemen and Indian teas, suggesting that caffeine dissolves more quickly. The uncertainties for the literature values are unknown.

Percent Errors

Tea Sample	Experimental Caffeine per g (mg/g) (± 0.001)	Literature Value (mg/g)	% Error (± 00.01)
Indian Tea	0.302	45	99.33%
Yemen Tea	0.280	50	99.44%
Turkish Tea	0.270	38	99.29%
Chinese Tea	0.227	25	99.09%
British Tea	0.230	35	99.34%

The results partially vary with the hypothesis, as the caffeine content in black teas differed by country of origin, but some methodological flaws limit the validity of these conclusions. The results from the Chinese teas had the lowest average at $0.047 \text{ g/L} \pm 0.005$, aligning with the literature presenting that Chinese black teas normally have lower caffeine content during processing and cultivation. The Indian tea has the highest average caffeine concentration at $0.107 \text{ g/L} \pm 0.001$. The Turkish and British teas presented intermediate caffeine concentrations of $0.064 \text{ g/L} \pm 0.001$ and $0.059 \text{ g/L} \pm 0.001$. Reflecting several experimental inconsistencies that impacted the results, Yemen's tea had a higher average caffeine content of $0.089 \text{ g/L} \pm 9.015$, but its data showed considerable variability.

The Chinese and Yemeni teas experience experimental variability through uneven mixing and solvent evaporation. It can be suggested that teas are efficient at releasing caffeine into hot water, as Indian and Yemeni black teas presented higher caffeine extraction rates. However, due to methodological inconsistencies, the results are not completely conclusive. In Trial 1, for the Yemen black tea, caffeine extraction was likely affected due to prolonged exposure to solvent where yields were reduced, leading to

mold growth. In trial 2, due to solvent contamination, the tea exceeded the expected caffeine yield. These inconsistencies question the accuracy of caffeine measurements from the Yemen tea. Further compromising the reliability of the data, human error and standard solution preparation resulted in volume exceedance. The improper flask sealing and the beaker slip for the Indian tea led to the loss in volume and might have reduced the caffeine concentration in the final solution.

Chinese tea has the lowest caffeine content, followed by Turkish and British teas showing intermediate content, and the Yemeni, then the Indian tea with the highest concentrations. However, procedural errors may have contributed to the high concentrations. A spectrophotometric outlier is suggested through the Yemeni tea fluctuations in its calibration curve, mostly caused by inconsistent solvent absorption and measurement process. The British and Turkish teas presented intermediate values, while the Indian and Yemeni teas had the highest. The lowest was found in Chinese tea. Aligning with the quantifiable caffeine content, the Chinese tea had the lowest solubility, while the Indian and Yemeni teas had the highest.

Trial 1:

Absorption vs Caffeine Concentration:

Caffeine Concentration (mg/L) (± 0.01 mg/L)	Absorption (273 nm) (± 0.001 nm)
0.00 (Distilled Water)	0.000
5.00	Inapplicable
10.00	0.060
20.00	0.022
30.00	0.045

Brewing Conditions of Black Tea Samples:

Tea Sample	Mass of Tea Used ((g) ± 0.001 g)	Brew Time (Min) $\pm (1$ minute)	Volume of Tea Brewed (mL) (± 100 mL)
British Tea	7.934	5	150
Chinese Tea	7.935	5	150
Yemen Tea	7.934	5	150
Turkish Tea	7.943	5	150
India Tea	7.932	5	150

pH Values of Black Tea Solutions:

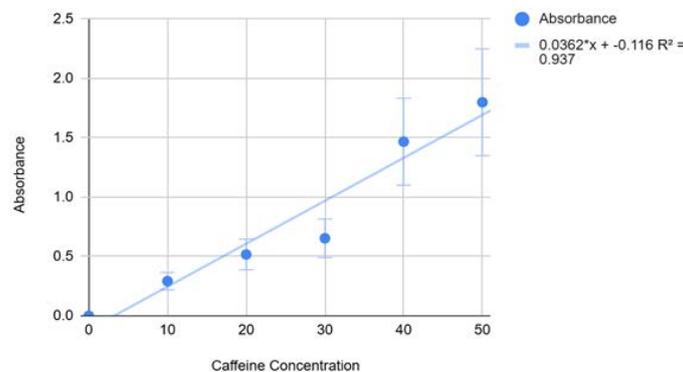
Tea Sample	pH (± 0.1)
Indian Tea	5.0
Yemen Tea	4.0
Turkish Tea	4.0
Chinese Tea	4.0
British Tea	5.0

Trial 2:

Absorption vs Caffeine Concentration:

Caffeine Concentration (mg/L) (± 0.1 mg/L)	Absorption (273 nm) (± 0.001 nm)
0.0 (Distilled Water)	0.000
10.0	0.292
20.0	0.517
30.0	0.653
40.0	1.467
50.0	1.799

Caffeine Concentration vs Absorption



Brewing Conditions of Black Tea Samples

Tea Sample	Mass of Tea Used ((g) ± 0.001 g)	Brew Time (Min) (± 1 minute)	Volume of Tea Brewed (mL) (± 100 mL)
British Tea	7.901	5	150
Chinese Tea	7.908	5	150
Yemen Tea	7.925	5	150
Turkish Tea	7.910	5	150
Indian Tea	7.915	5	150

pH Values of Black Tea Solutions:

Tea Sample	pH (± 0.1 mg/L)
Indian Tea	5.0
Yemen Tea	4.0
Turkish Tea	4.0
Chinese Tea	4.0
British Tea	5.0

Caffeine Extraction from Black Teas:

Tea Sample	Volume of Tea Used (mL) (± 10 mL)	Volume of CH ₂ Cl ₂ Used (mL) (± 10 mL)	Mass of Caffeine Extracted (g) (± 0.001 g)
British Tea	50	25	2.879
Indian Tea	50	25	3.140
Chinese Tea	50	25	0.608
Turkish Tea	50	25	2.567
Yemen Tea	50	25	4.501

Mass of Caffeine Extracted = Caffeine Concentration x Volume of Tea Solutions

Caffeine per Gram of Tea Powder = Mass of Caffeine Extracted/Mass of Tea Used

Caffeine Concentration of the Foreign Black Teas

Tea Sample	Absorbance (± 0.001)	Caffeine Concentration (mg/L) (± 0.001 mg/L)
Indian Tea	1.590	49.531
Yemen Tea	1.165	35.387
Turkish Tea	1.535	45.608
Chinese Tea	1.445	43.123
British Tea	1.205	36.492

Solubility Efficiency of Caffeine (%) = (Experimental Caffeine per g/Literature Value) x 100

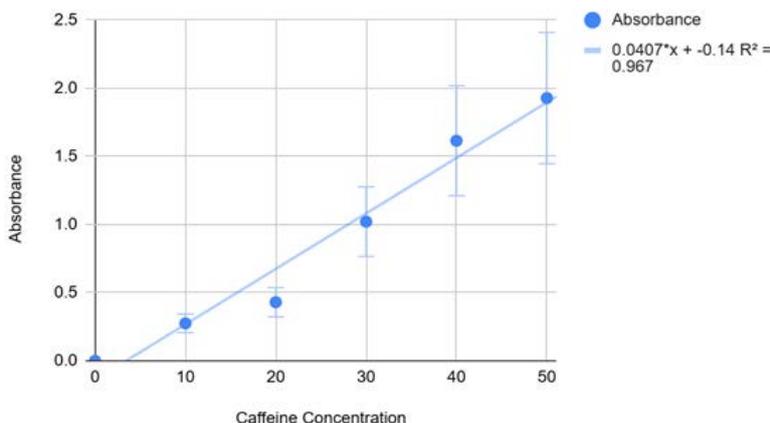
Calculate Caffeine Concentration in Tea Solutions:

- Equate the Calibration Curve's linear equation to the absorbance of each tea sample
- Solve the equation for X, which will provide the concentration of each tea

Trial 3:**Absorption vs Caffeine Concentration:**

Caffeine Concentration (mg/L) ± 0.1 mg/L	Absorption (273 nm) (± 0.001)
0.0 (Distilled Water)	0.000
10.0	0.271
20.0	0.430
30.0	1.021
40.0	1.615
50.0	1.928

Caffeine Concentration vs Absorption

**Brewing Conditions of Black Tea Samples:**

Tea Sample	Mass of Tea Used (g) (± 0.001 g)	Brew Time (Min) (± 1 minute)	Volume of Tea Brewed (ml) (± 100 mL)
British Tea	7.929	5	150
Chinese Tea	7.947	5	150
Yemen Tea	7.910	5	150
Turkish Tea	7.917	5	150
Indian Tea	7.980	5	150

pH Values of Black Tea Solutions:

Tea Sample	pH (± 0.1)
Turkish Tea	4.0
Chinese Tea	5.0
British Tea	5.0
Indian Tea	4.0
Yemen Tea	4.0
50.0	1.928

pH Values of Black Tea Solutions:

Tea Sample	Volume of Tea Used (mL) (± 10 mL)	Volume of CH_2Cl_2 Used (mL) (± 10 mL)	Mass of Caffeine Extracted (g) (± 0.001 g)
British Tea	50	25	0.045
Indian Tea	50	25	0.067
Chinese Tea	50	25	0.038
Turkish Tea	50	25	0.050
Yemen Tea	50	25	0.074

Percent Yield of Caffeine Extracted = (Mass of Caffeine Extracted (g))/(Theoretical Mass of Caffeine (g)) x 100

Brew Time vs Caffeine Extraction Rate:

Caffeine Extraction Rate = Mass of Caffeine Extracted (mg)/Brew Time (min)

Caffeine Concentration of the Foreign Black Teas:

Tea Sample	Absorbance (273) (± 0.001)	Caffeine Concentration (mg/L) (± 0.00001 mg/L)
Indian Tea	1.492	40.02451
Yemen Tea	1.684	44.73039
Turkish Tea	1.210	33.11275
Chinese Tea	0.872	24.82843
British Tea	1.135	31.27451

Standard Deviations of Absorbance:

Trials	Standard Deviations (± 0.001)
Trial 1	Inapplicable
Trial 2	0.563
Trial 3	0.644

The reliability of the data and its ability to represent true caffeine content was limited, as only three trials were conducted, considering the processing variability in tea. It made it challenging to determine if variations were actual differences or experimental errors in caffeine content, considered less significant than limitations regarding measurements. Five trials per sample should be conducted, as it is sufficient to reduce random error and increase statistical reliability within a typical lab period. This change allows for a clearer identification of anomalies or outliers and makes trends more dependable [22,23].

From spectrophotometric methods, caffeine concentrations may have been under- or overestimated because of other absorbance in similar wavelengths from tea compounds. This limitation introduced systematic errors in the absorbance readings, affecting validity. An improvement could be to prepare a control solution of caffeine-free tea, using it as a correct background absorbance, isolating caffeine absorbance more effectively and improving validity of the concentration measurements [25,26].

A random error was likely introduced with inaccuracies in preparing the standard caffeine solutions or transferring liquids, affecting accuracy and calibration in spectrometry. This limitation affected the precision and reliability of the standard calibration and final measurements, introducing random error. It contributes to inter-trial variability and its impact is lower than other limitations. The variability in measured volumes would be reduced using equipment. Accuracy and predictability would be enhanced with the consistent usage of labeled glassware and pipetting techniques. Upon comparison to the literature values and experimental results, the high caffeine content in Yemen and Indian teas was present. This was attributed to inefficiency in extraction, as higher yields potentially resulted due to the rapid dissolution of caffeine in warmer climates. Instrumental uncertainties affected the accuracy of mass measurements, presenting slight overestimation, while differences in brew times may have contributed to the variations in the findings.

Final Calculations:

Mass of Caffeine Extracted (g) = Caffeine Concentration x Volume of Tea Solution

- Given: Volume of teas solution = 50 mL

Tea Sample	Trial 2: Caffeine Concentration (mg/L) (± 0.001)	Trial 3: Caffeine Concentration (mg/L) (± 0.001)	Calculations (± 0.001)
Indian Tea	46.469	49.531	Trial 2: $(49.469 \times 50)/1000 = 2.323$ g Trial 3: $(49.531 \times 50)/1000 = 2.477$ g
Yemen Tea	52.469	36.250	Trial 2: $(52.469 \times 50)/1000 = 2.623$ g Trial 3: $(36.250 \times 50)/1000 = 1.812$ g
Turkish Tea	37.656	47.812	Trial 2: $(37.656 \times 50)/1000 = 1.883$ g Trial 3: $(47.812 \times 50)/1000 = 2.391$ g
Chinese Tea	27.094	45.000	Trial 2: $(27.094 \times 50)/1000 = 1.355$ g Trial 3: $(45.000 \times 50)/1000 = 2.250$ g
British Tea	35.312	37.500	Trial 2: $(35.312 \times 50)/1000 = 1.766$ g Trial 3: $(37.500 \times 50)/1000 = 1.875$ g

Caffeine per Gram of Tea Powder = (Mass of Caffeine Extracted)/(Mass of Tea Used)

Tea Sample Trial 2: Mass of Tea (g) (± 0.001) Trial 3: Mass of Tea (g) (± 0.001) Calculations (mg/g) (± 0.001)

Tea Sample	Trial 2: Mass of Tea (g) (± 0.001)	Trial 3: Mass of Tea (g) (± 0.001)	Calculations (mg/g) (± 0.001)
Indian Tea	7.915	7.980	Trial 2: $2.323/7.915 = 0.293$ mg/g Trial 3: $2.477/7.980 = 0.310$ mg/g
Yemen Tea	7.925	7.910	Trial 2: $2.623/7.925 = 0.331$ mg/g Trial 3: $1.812/7.910 = 0.229$ mg/g
Turkish Tea	7.910	7.917	Trial 2: $1.883/7.910 = 0.238$ mg/g Trial 3: $2.391/7.917 = 0.302$ mg/g
Chinese Tea	7.908	7.947	Trial 2: $1.355/7.908 = 0.171$ mg/g Trial 3: $2.250/7.947 = 0.283$ mg/g
British Tea	7.901	7.929	Trial 2: $1.766/7.901 = 0.224$ mg/g Trial 3: $1.875/7.929 = 0.235$ mg/g

Solubility Efficiency = (Experimental Caffeine per g/Literature Value) x 100

Tea Sample	Literature Value (mg/g)	Calculations (%) (± 0.01)
Indian Tea	45	Trial 2: $(0.293/45) \times 100 = 0.65\%$ Trial 3: $(0.310/45) \times 100 = 0.69\%$
Yemen Tea	50	Trial 2: $(0.293/45) \times 100 = 0.65\%$ Trial 3: $(0.310/45) \times 100 = 0.69\%$
Turkish Tea	38	Trial 2: $(0.238/38) \times 100 = 0.63\%$ Trial 3: $(0.302/38) \times 100 = 0.79\%$
Chinese Tea	25	Trial 2: $(0.171/25) \times 100 = 0.68\%$ Trial 3: $(0.283/25) \times 100 = 1.13\%$
British Tea	35	Trial 2: $(0.224/35) \times 100 = 0.64\%$ Trial 3: $(0.236/35) \times 100 = 0.67\%$

Percent Yield of Caffeine Extracted (%) = (Mass of Caffeine Extracted (g)/Theoretical Mass of Caffeine (g)) x 100

- Literature Value: 30 mg of caffeine per gram of tea

- Theoretical Caffeine = 30 x Mass of Tea Used

Trial 2:

Tea Sample	Mass of Teas Used (g) (± 0.001)	Theoretical Caffeine (mg) (± 0.1)	Mass Extracted (mg) (± 1000)	% Yield (± 0.01)
Indian Tea	7.915	$30 \times 7.915 = 237.5$	2323 mg	97.86%
Yemen Tea	7.925	$30 \times 7.925 = 237.8$	2623 mg	110.3%
Turkish Tea	7.910	$30 \times 7.910 = 237.3$	1883 mg	79.35%
Chinese Tea	7.908	$30 \times 7.908 = 237.2$	1355 mg	57.14%
British Tea	7.901	$30 \times 7.901 = 237.0$	1766 mg	74.50%

Trial 3:

Tea Sample	Mass of Tea Used (g) (± 0.001)	Theoretical Caffeine (mg) (± 0.1)	Mass Extracted (mg) (± 1000)	% Yield (± 0.1)
Indian Tea	7.980	239.4	2477 mg	103.4%
Yemen Tea	7.910	237.3	1812 mg	76.4%
Turkish Tea	7.917	237.5	2391 mg	100.7%
Chinese Tea	7.947	238.4	2250 mg	94.1%
British Tea	7.929	237.9	1875 mg	78.8%

Caffeine Extraction Rate = (Mass of Caffeine Extracted (mg)/Brew Time (min))

- Brew Time: 5 min for all samples

Trial 2:

Tea Sample	Mass Extracted (mg) (± 1000)	Brew Time (mn) (± 1 minute)	Extraction Rate (mg/min) (± 0.1)
Indian Tea	2323	5	464.6
Yemen Tea	2623	5	524.6
Turkish Tea	1883	5	376.6
Chinese Tea	1355	5	271.0
British Tea	1766	5	353.2

Trial 3:

Tea Sample	Mass Extracted (± 1000)	BrewTime (± 1 minute)	Extraction Rate (± 0.1)
Indian Tea	2477	5	495.4
Yemen Tea	1812	5	362.4
Turkish Tea	2391	5	478.2
Chinese Tea	2250	5	450.0
British Tea	1875	5	375.0

Average:**Caffeine Concentration of the Foreign Black Teas:**

Tea Sample	Average (± 0.000001)
British	33.883255
China	33.875715
Yemen	40.058695
Turkey	39.360375
India	44.777755

Brewing Conditions of Black Tea Samples (mass):

Tea Sample	Average (± 0.001)
British	7.921
China	7.930
Yemen	7.923
Turkey	7.923
India	7.942

pH Values of Black Tea Solutions:

Tea Sample	Average (± 0.001)
British	5.000
China	4.333
Yemen	4.000
Turkey	4.000
India	4.667

pH Values of Black Tea Solutions:

Tea Sample	Average (± 0.001)
British	1.462
China	0.323
Yemen	2.288
Turkey	1.301
India	1.604

Absorption vs Caffeine Concentration:

Caffeine Concentration (± 0.01)	Average (± 0.001)
0.00	0.000
5.00	Inapplicable
10.00	0.208
20.00	0.323
30.00	0.573
40.00	1.541
50.00	1.864

Range:**Caffeine Concentration of the Foreign Black Teas:**

Tea Sample	Range (± 0.00001)
British	5.21749
China	18.29457
Yemen	9.34339
Turkey	12.49525
India	9.50649

Caffeine Extraction from Black Teas:

Tea Sample	Range (± 0.001)
British	2.834
China	0.570
Yemen	4.427
Turkey	2.517
India	3.073

pH Values of Black Tea Solutions:

Tea Sample	Range (± 0.001)
British	0.000
China	1.000
Yemen	0.000
Turkey	0.000
India	1.000

Brewing Conditions of Black Tea Samples (mass):

Tea Sample	Range (± 0.001)
British	0.033
China	0.039
Yemen	0.024
Turkey	0.033
India	0.065

Absorption vs Caffeine Concentration:

Caffeine Concentration (± 0.01)	Range (± 0.001)
0.00	0.000
5.00	Inapplicable
10.00	0.232
20.00	0.495
30.00	0.976
40.00	0.148
50.00	0.129

Mass of Caffeine Extracted (g):

Tea Sample	Trial 2 (g) (± 0.001)	Trial 3 (g) (± 0.001)	Average (g) (± 0.001)	Range (g) (± 0.001)
Indian Tea	2.323	2.477	2.400	0.154
Yemen Tea	2.623	1.812	2.218	0.811
Turkish Tea	1.883	2.391	2.137	0.508
Chinese Tea	1.355	2.250	1.803	0.895
British Tea	1.755	1.875	1.821	0.109

Caffeine per Gram of Tea Powder:

Tea Sample	Trial 2 (mg/g) (± 0.001)	Trial 3 (mg/g) (± 0.001)	Average (mg/g) (± 0.001)	Range (mg/g) (± 0.001)
Indian Tea	0.293	0.310	0.302	0.017
Yemen Tea	0.331	0.229	0.280	0.102
Turkish Tea	0.238	0.302	0.270	0.064
Chinese Tea	0.171	0.283	0.227	0.112
British Tea	0.224	0.235	0.230	0.011

Solubility Efficiency (%):

Tea Sample	Trial 2 (%) (± 0.01)	Trial 3 (%) (± 0.01)	Average (%) (± 0.01)	Range (%) (0.01)
Indian Tea	0.65	0.69	0.67	0.04
Yemen Tea	0.66	0.46	0.56	0.20
Turkish Tea	0.63	0.79	0.71	0.16
Chinese Tea	0.68	1.13	0.91	0.45
British Tea	0.64	0.67	0.66	0.03

Percentage Yield of Caffeine Extracted (%):

Tea Sample	Trial 2 (%) (± 0.01)	Trial 3 (%) (± 0.1)	Average (%) (± 0.01)	Range (%) (± 0.01)
Indian Tea	97.86	103.4	100.60	5.54
Yemeni Tea	110.30	76.4	93.34	33.92
Turkish Tea	79.35	100.7	90.03	21.35
Chinese Tea	57.14	94.1	75.62	36.96
British Tea	74.50	78.8	76.65	4.30

Caffeine Extraction Rate (mg/min):

Tea Sample	Trial 2 (mg/min) (± 0.1)	Trial 3 (mg/min) (± 0.1)	Average (mg/min) (± 0.1)	Range (mg/min) (± 0.1)
Indian Tea	464.6	495.4	480.0	30.8
Yemen Tea	524.6	362.4	443.5	162.2
Turkish Tea	376.6	478.2	427.4	101.6
Chinese Tea	271.0	450.0	360.5	179.0
British Tea	353.2	375.0	364.1	21.8

Comparison with the Theoretical Value:

Tea Sample	Experimental Caffeine per g (mg/g) (Avg.) (± 0.001)	Literature Value (mg/g) (± 10)	% Error (± 0.01)
Indian Tea	0.302	45	99.33%
Yemen Tea	0.280	50	99.44%
Turkish Tea	0.270	38	99.29%
Chinese Tea	0.227	25	99.09%
British Tea	0.230	35	99.34%

Propagation of Uncertainty in Caffeine Concentration:

Tea Sample	Absorbance (273 nm) (± 0.001)	Uncertainty in Absorbance	Caffeine Concentration (mg/L) (± 0.00001)	Uncertainty in Caffeine Concentration (mg/L)
Indian Tea	1.492	± 0.001	40.02451	± 0.00001
Yemen Tea	1.684	± 0.001	44.73039	± 0.00001
Turkish Tea	1.210	± 0.001	33.11275	± 0.00001
Chinese Tea	0.872	± 0.001	24.82843	± 0.00001
British Tea	1.135	± 0.001	31.27451	± 0.00001

Propagation of Uncertainty in Caffeine per Gram:

Tea Sample	Mass of Caffeine Extracted (g) (± 0.001)	Uncertainty in Caffeine Extracted (g) (± 0.001)	Mass of Tea Used (g) (± 0.001)	Uncertainty in Mass of Tea Used (g)	Caffeine per Gram (g) (± 0.00001)	Uncertainty in Caffeine per Gram (g)
Indian Tea	0.067	± 0.001	7.980	± 0.001	0.00839	± 0.000125
Yemen Tea	0.074	± 0.001	7.910	± 0.001	0.00936	± 0.000124
Turkish Tea	0.050	± 0.001	7.917	± 0.001	0.00631	± 0.000122
Chinese Tea	0.038	± 0.001	7.947	± 0.001	0.00478	± 0.000121
British Tea	0.045	± 0.001	7.929	± 0.001	0.00567	± 0.000122

Propagation for Caffeine per Gram:

$$\frac{\Delta \text{Caffeine per gram}}{\text{Caffeine per gram}} = \sqrt{\left(\frac{\Delta \text{Caffeine Extracted}}{\text{Caffeine Extracted}}\right)^2 + \left(\frac{\Delta \text{Mass of Tea Used}}{\text{Mass of Tea Used}}\right)^2}$$

$$\frac{\Delta \text{Caffeine per gram}}{\text{Caffeine per gram}} = \sqrt{(0.04)^2 + (0.05)^2}$$

$$\frac{\Delta \text{Caffeine per gram}}{\text{Caffeine per gram}} = \sqrt{0.0016 + 0.0025} = \sqrt{0.0041} \approx 0.064$$

Propagation of Uncertainty in Caffeine Extraction Rate:

Tea Sample	Mass of Caffeine Extracted (g)	Brew Time (min)	Uncertainty in Caffeine Extracted (g)	Uncertainty in Brew Time (min)	Caffeine Extraction Rate (mg/min)	Uncertainty in Caffeine Extraction Rate (mg/min)
Indian Tea	0.067	5	± 0.001	± 1	13.4	± 2.7
Yemen Tea	0.074	5	± 0.001	± 1	14.8	± 3.0
Turkish Tea	0.050	5	± 0.001	± 1	10.0	± 2.0
Chinese Tea	0.038	5	± 0.001	± 1	7.6	± 1.5
British Tea	0.045	5	± 0.001	± 1	9.0	± 1.8

Propagation for Caffeine Extraction Rate:

$$\frac{\Delta \text{Extraction Rate}}{\text{Extraction Rate}} = \sqrt{\left(\frac{\Delta \text{Caffeine Extracted}}{\text{Caffeine Extracted}}\right)^2 + \left(\frac{\Delta \text{Brew Time}}{\text{Brew Time}}\right)^2}$$

$$\frac{\Delta \text{Extraction Rate}}{\text{Extraction Rate}} = \sqrt{(0.04)^2 + (0.04)^2}$$

$$\frac{\Delta \text{Extraction Rate}}{\text{Extraction Rate}} = \sqrt{0.0016 + 0.0016} = \sqrt{0.0032} \approx 0.056$$

Evaluation**Uncertainties:**

Measurement	Uncertainty	Source of Uncertainty	Instrument Used
Tea Mass	± 0.001 g	Balance Precision	Analytical balance
Brew Time	± 1 s	Human Reaction Error	Manual Timing
Water Volume	± 1 mL	Meniscus Reading Error	Graduated Cylinder
pH	± 0.1	Subjective Interpretation	pH strips
Tea & Dichloromethane Volume	± 0.001 g	Balance Precision	Analytical Balance
UV-Vis Absorbance	± 0.1 mg/L	Calibration Curve Limitations	UV-Vis Spectrophotometer

Equipment Uncertainties:

Equipment	Analytical Balance	Graduated Cylinder	Spectrophotometer (Absorbance)
Uncertainty	± 0.001 g	± 1 ml	± 0.001 nm

The error bars for the calibration curves were determined using the averages of the standard solutions. Regarding the calibration curve, the Beer-Lambert Law forms a linear relationship between absorbance and concentration. Highly concentrated deviations can lead to many inaccuracies. Systematic uncertainty might have also been introduced by errors in the standard caffeine concentrations. Lower than expected yield rates might have also resulted from the evaporation of dichloromethane before the caffeine had been completely isolated. ,

Caffeine concentration calculations may have been impacted by small absorbance errors during spectrophotometer readings. Uncertainties in extraction and spectrophotometry resulted in high percent errors through caffeine content trends. Measurement precision errors and calibration errors many have compounded, while variability in extraction led to artificially low yields in some trials.

Mass measurements came from the analytical balance ± 0.001 precision, leading to noticeable percent uncertainty, affecting caffeine yield from small extracted mass. Caffeine concentration confidence was limited by these uncertainties. Minor uncertainties in absorbance readings, concentration calculations and mass measurements influenced the final results. The actual extracted caffeine varied because of uncertainties in absorbance and mass, propagated through caffeine concentration calculations from the calibration

curve. Though there were smaller impacted absorbance uncertainties, larger discrepancies existed.

It can be stated that random errors did not affect the trend observed, significantly, as suggested by the low uncertainties in caffeine concentration and absorbance. However, caffeine yield calculations could have been marginally affected, uncertainty in mass measurements was more significant. Compared to larger systematic errors, percent errors were already extremely high, the impact of small uncertainty values on the final conclusions was minor. The uncertainty range of this experiment was not precise enough to ensure that the caffeine concentrations were accurate, but was small enough to support the observed caffeine trends.

Despite the smaller individual uncertainties, the overall percent error was larger and reduced the confidence in the absolute caffeine content values. The high error suggests that systematic errors had more of a significant impact on accuracy than random uncertainties. The experiment's reliability would have been lower if uncertainty values had been larger, as the range of possible values would increase. The experiment's accuracy can be seen to have been limited by methodological errors because uncertainties were relatively small instead of random variations in measurements. Due to major systematic errors, the overall confidence in the extracted caffeine values are low, while observed trends were not drastically affected by small uncertainties.

Uncertainty in Calibration Curve:

Trials	Slope Uncertainty (± 0.0001)	Intercept Uncertainty (± 0.001)
Trial 1	Unapplicable	Unapplicable
Trial 2	± 0.0021	± 0.010
Trial 3	± 0.0052	± 0.015

Percent Uncertainty for Individual Measurements:

Measurements	Uncertainties	Percentage Uncertainty (± 0.001)	Impact of Results
Mass of Caffeine Extracted	± 0.001 g	$\sim 2.220\%$	Small uncertainty, but high percentage errors suggest larger bonuses in accuracy
UV-Vis Absorbance Readings	± 0.001 nm	$\sim 0.086\%$	Minimal impact on caffeine concentration
Caffeine Concentration (mg/L)	Propagated from absorbance uncertainty	$\sim 0.084\%$	Slight variability in calculated caffeine values

While the limitations of measurement devices and methods are present through percent uncertainties, discrepancies are highlighted in percent errors between literature and experimental results. Factors beyond measurement uncertainty have likely contributed to the significant deviation. Some level of precision loss can be presented in the high uncertainty in the mass of caffeine extracted. However, the percent error suggests that precision does not solely account for the inaccuracy within the literature and experimental values. The significant differences between the errors and uncertainties propose how the percent errors can be presented through the experimental setup, which reflects the limitations and percent uncertainties. The percent error in caffeine content cannot be attributed to concentration or absorbance calculations, suggesting the error comes from other issues. Issues like improper sample handling and caffeine extraction caused discrepancies, despite precision.

How much Caffeine Content is there in Foreign Black Teas from Indian, England, Yemen, Turkey and China? While Chinese tea had lower concentrations and yields, Indian and Yemen tea had higher levels, suggesting lower natural caffeine. Per gram, Yemen and Indian teas had higher caffeine, supporting how *Camellia* grown in warmer climates presents more caffeine. Turkish and British teas had intermediate caffeine levels, while Chinese had lower, aligning with literature values. With extraction inconsistencies, Turkish and Indian teas had stable solubilities and effectiveness from yields suggesting consistent caffeine extraction. Chinese tea consisted of its low caffeine, as it had the slowest extraction rate, while Indian and Yemen teas had the fastest rates, suggesting quicker caffeine dissolution.

In general, the overall trend in caffeine content across the black teas remains clear, which supports the validity of general conclusions. The findings should be viewed with caution because although they support main conclusions, the absolute caffeine content may not reflect the caffeine measured, due to the high percent errors from extraction inconsistencies and methodological flaws.

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