IB Chemistry SL Internal Assessment

Title:

An investigation into the effect of the pH of the brewing solution on the extraction of caffeine from a cup of tea.

Research Question:

What is the effect of increasing the pH (11.238, 11.604, 11.756, 11.858, 11.917) of the aqueous brewing solution consisting of sodium carbonate (Na_2CO_3) of the tea on the yield of caffeine extracted (in mg ±2mg) from a cup of black tea using dichloromethane solvent extraction, measured by iodometric back titration against a standard solution of sodium thiosulphate.

May 2021

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Research Question

What is the effect of increasing the pH (11.23, 11.60, 11.76, 11.86, 11.92) of the aqueous brewing solution consisting of sodium carbonate (Na_2CO_3) using to make tea on the yield of caffeine extracted (in mg) from the cup of black tea using dichloromethane solvent extraction, measured by iodometric back titration against a standard solution of sodium thiosulphate.

Introduction

Pure caffeine ($C_8H_{10}N_4O_2$) is found in a white, bitter-tasting crystalline powder form. It's a member of the alkaloid class, meaning it is an organic compound containing nitrogen atoms. The caffeine molecular structure is made up from a purine ring system, and is seen in *Figure 1*[1]. The compound, however, is not commonly found as a powder, but in food grade items such as coffee, sodas, energy drinks and tea.

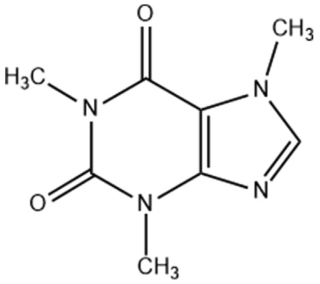

Figure 1: Caffeine Molecule

With caffeine becoming the most widely used and unregulated psychoactive drug[2], its components and the amount of such substance in our teas, coffees and energy drinks has become a subject of interest to me. Often drinking tea while studying, I saw that the tea bags I used had their caffeine content labeled on the side of the box, which piqued my curiosity about how much of the it could be experimentally extracted, and if it matched the label. Additionally, I wanted to know how I could maximize this yield through manipulating a variable in the extraction process.

This investigation aims to explore the effect of raising the pH of the aqueous brewing solution the black tea is made in on the yield of caffeine (in mg) produced from the solvent extraction process. This will be done through first a dichloromethane (CH_2Cl_2) extraction to obtain crude caffeine, then an iodometric back titration to determine the amount of pure caffeine extracted. Additionally, the analysis of the extracted substance's melting point in comparison to pure caffeine will be conducted.

Background Information:

Tea leaves are made up of 3 main constituents[3] (1) cellulose, a natural water-insoluble polymer, (2) caffeine, and (3) tannins, a type of phenolic compound.[4] In order to extract the caffeine from tea bags, it has to be separated from the other constituents through several steps.

First the cellulose is removed by extracting the water-soluble constituents through brewing the tea bag in hot water. Hot water is necessary as caffeine is not readily soluble at room temperature, having a solubility of only 16 mg/mL, but 667 mg/mL at 100°C.[5] After cooling, the caffeine then has to be extracted from the water using dichloromethane (CH_2Cl_2), an organic solvent, immiscible in water and commonly referred to as DCM. This extraction is possible due to caffeine being more soluble in DCM than in water at room temperature (solubility of 16 mg/mL in water and 140 mg/mL in DCM).[6]

However, while the cellulose is removed by this stage, the tannins are still slightly soluble in DCM. To remedy this, and facilitate the extraction of only caffeine from the solvent, the tannins need to be

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[1] "Molecular Structure of Caffeine." *Chemistry Stack Exchange*, 15 July 2016, chemistry.stackexchange.com/questions/54268/molecular-structure-of-caffeine.

[2] "Caffeine." *Centre for Addiction and Mental Health*, www.camh.ca/en/health-info/mental-illness-and-addiction-index/caffeine.

[3] Gebely, Tony. "Chemical Compounds in Tea." *Tea Epicure*, 22 Feb. 2019, teaepicure.com/tea-chemistry/.

[4] "Tannin." *Encyclopædia Britannica*, Encyclopædia Britannica, Inc., 27 Jan. 2021, www.britannica.com/science/tannin.

[5] "Caffeine (Anhydrous) Product Information." *Sigma-Aldrich*, www.sigmaaldrich.com/content/dam/sigma-aldrich/docs/Sigma-Aldrich/Product_Information_Sheet/c0750pis.pdf.

[6] Chaugule, Aniket, et al. "Extraction of Caffeine." *International Journal of Advanced Research in Chemical Science*, vol. 6, no. 9, 2019, pp. 11–19., doi:10.20431/2349-0403.0609002.

converted into salts. Since they are phenolic compounds, they are acidic enough so that the addition of sodium carbonate (Na_2CO_3), which acts as a base, can cause the deprotonation of their -OH group[7] and thus the formation of the salts of the tannins will occur. Since theses salts are now insoluble in DCM, and soluble in the polar water, caffeine is the only constituent found in the solvent and can be extracted through its evaporation.

With that being said, certain variables affect the efficiency of this extraction, including the pH of the aqueous brewing solution that the tea bags will be submerged in during the first step. According to scientific literature by Kim et al., the yield of caffeine extracted "increase[s] when the pH of the extraction solution [is] increased from 4 to 7,"[8] but begins to decrease afterwards.[9] The effect of pH and this trend can be explained through the dependence of the partition coefficient (K_d) of caffeine on the pH of the solution in the system of caffeine-solvent-water. The higher the K_d, the higher the concentration of caffeine in the solvent instead of the water. According to a study conducted by Klebanov et al., the relationship between the K_d of caffeine and pH is as seen in *Figure 2.*[7]

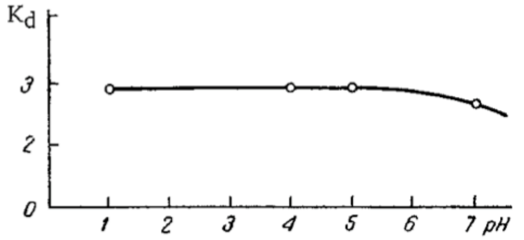

Figure 2: K_d of caffeine vs. pH

Due to this trend, I chose pH to be my independent variable to be manipulated. According to a study done by Vuong et al. is the last point before increasing the pH drops the efficiency of caffeine extraction.[10] However, their study only investigated pH values up to 10 and only on green tea. I wanted to adapt this to see if such trends continued and if they were applicable to black tea, so I set pH values above the range they investigated. My dependent variable is the mass of pure caffeine extracted which I measured in mg (±1). I also took into consideration the extraction's percentage purity after the iodometric back titration through calculating the mass of pure caffeine found in the crude extract.

Hypothesis

Since the K_d of caffeine in the solvent starts dropping off after a pH of 7, it means that a higher pH begins to decrease the concentration of caffeine dissolved in the solvent and increase the concentration in water, thus decreasing the yield extracted as some caffeine molecules will be left behind. With this theoretical background and previous research on green tea and coffee pointing to a negative correlation between pH and efficiency of caffeine extraction, I predict that the trend will continue when investigated with black tea to form a negative linear regression between the variables of pH and extracted caffeine in mg as seen in the sketch in *Figure 3.*

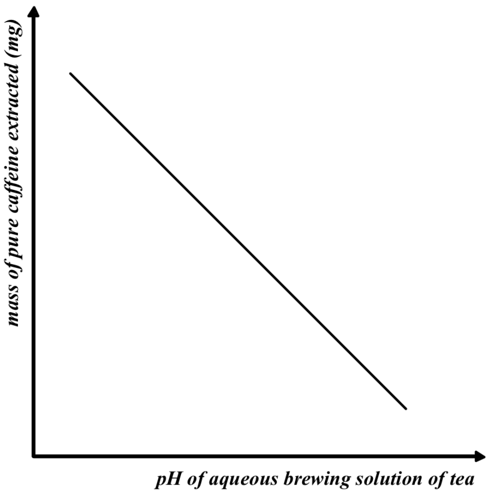

Figure 3: Predicted trend between pH & mass of pure caffeine extracted

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[7] "Tannin." *Encyclopædia Britannica*, Encyclopædia Britannica, Inc., 27 Jan. 2021, www.britannica.com/science/tannin.

[8] Kim, Sang-Hee, et al. "Effect of PH on the Green Tea Extraction." *Korean Journal of Food Science and Technology*, vol. 31, no. 4, 31 Aug. 1999, pp. 1024–1028.

[9] Klebanov, G. S., et al. "Extraction of Caffeine from Aqueous Solutions." *Pharmaceutical Chemistry Journal*, vol. 1, no. 4, 1967, pp. 221–223., doi:10.1007/bf00770195.

[10] Vuong, Quan V., et al. "Effects of Aqueous Brewing Solution PH on the Extraction of the Major Green Tea Constituents." *Food Research International*, vol. 53, no. 2, 14 Sept. 2012, pp. 713–719., doi:10.1016/j.foodres.2012.09.017.

Variables

Independent Variable	*pH of the aqueous brewing solution containing sodium carbonate (Na_2CO_3) that the black tea bags are prepared in*— The pH values were meant to be measured using a pH meter but due to the lab's meter's inaccuracy and inability to be calibrated, the pH had to be found analytically by using the mass of Na_2CO_3 added to the solutions. The pH values (11.238, 11.604, 11.765, 11.858, 11.917 ±0.004) were derived from different masses (0.980g, 5.010g, 9.987g, 15.109g, 19.876g ±0.001) of Na_2CO_3 by referring back to the HL topic 18 to carry out the set of calculations needed to find the pH of a weak base.
Dependent Variable	*The mass of the pure caffeine extracted from 2 black tea bags measured in mg (±1).* This will be measured after the extraction of crude caffeine from the solvent extraction and the iodometric back titration using a standard sodium thiosulfate ($Na_2S_2O_3$) solution of known concentration (0.005M ±0.001) has been conducted in order to find the *pure* mass.

Controlled Variables	Method of Control & Possible Effect on Results
Type of tea bag	The brand of tea bag was maintained throughout the experiment (Lipton Yellow Label Black Tea) to control the literature value of caffeine in them. If different tea bags were used for each trial, the amount of caffeine extractable would also differ and skew the end results.
Amount of water used for brewing the tea	Each tea bag was brewed in a beaker containing 150mL (±10) of water to maintain the same or as similar of a concentration of tea as possible in each sample. This would allow for a fair comparison of the caffeine extracts at the end.
Length of time tea was brewed for	Each tea bag was brewed in its water over a constant source of heat for 15.00 minutes ± 0.01minutes. This, again, is to keep a similar concentration of tea and get as much of the caffeine into the water as possible.
Amount of Na_2CO_3 solution added to brewed tea	A constant volume of 5.00mL ± 0.01mL of the alkali Na_2CO_3 solution was pipetted into the brewed tea after it was cooled, regardless of the pH of the sample.
Amount of DCM (CH_2Cl_2) used for extraction	A constant of 25.0 ± 0.1mL of DCM was measured using a graduated cylinder and used in each extraction of the tea. The amount was kept controlled in order to not disadvantage any sample with an inadequate amount of solvent that would skew the final mass to the lower pole.
Amount of DCM (CH_2Cl_2) extractions per sample	Each sample with a different pH had 2 separate DCM solvent extractions conducted on it to get the maximum amount of caffeine removed from the aqueous solution.
Length of time for DCM (CH_2Cl_2) and water to settle	After the DCM and aqueous layer were mixed, they were placed into a separating funnel for the same amount of time of 25.00 ± 0.01min in order to separate into different layers. Not keeping this time constant would have caused in the loss of some of the DCM containing caffeine mixed in the aqueous layer still in the separating funnel.
Concentration and Volume of Lugol's Iodine solution	During the iodometric back titration, the concentration of the Lugol's iodine stock solution was kept constant at 5% iodine and 10% potassium iodide to facilitate the calculations for the pure caffeine. Additionally, the volume added to the erlenmeyer flask was also constant at 25.0 ± 0.1mL measured using a graduated cylinder.

Concentration of Sodium Thiosulfate ($Na_2S_2O_3$)	The concentration of sodium thiosulfate used in the burette during the iodometric back titration was a standard one and was maintained at 0.005 ±0.001M.
Amount of Starch Indicator used	A constant of only 3 drops of the starch was used during each sample's iodometric back titration in order to not cloud up the solution at the end, resulting in a possible overshooting of the end point which is supposed to be clear at the end.

Uncontrolled	**Possible Method of Control & Possible Effects on Results**
Shaking of Separation Funnel	The length of time the separation funnel was shaken was not measured or controlled and this could have affected the results since some samples would have had increased contacted with the DCM layer due to additional mixing while others would have had limited contact. The more contact there is the greater the extraction of caffeine into the solvent. This could be controlled by allocating a number of gentle shakes to be a standard.
Temperature of Tea after cooled	The temperature of the tea after it has cooled down from brewing affects the extraction into the DCM since the solubility of caffeine in water and DCM fluctuates with the temperature. If the temperature of one sample was higher than another that means that less caffeine could have been extracted due to the caffeine still being slightly soluble in water at that temperature. A set temperature should have been set as a standard before the start of the solvent extraction.
Fluctuation of digital balance	The balance available at the lab had a tendency to fluctuate a lot and was not able to be calibrated. Due to the reliance on its accuracy, it can be said that this had an effect on the results at the end as they could be inaccurate and not precisely reflect my yield. This could be avoided and controlled by using a balance that has a shield around it, and possibly one that was accurate to 4 decimal places instead of 3 as our lab provided.

Risk Assessment & Safety

Hazard	**Control Measure**
Safety: Sodium carbonate (Na_2CO_3) causes skin and serious eye irritation	Gloves should be worn when measuring out the masses and hands should be washed thoroughly after handling the powder. Eye protection goggles should be worn.
Safety: Dichloromethane (CH_2Cl_2) causes skin irritation and serious eye irritation. It may cause drowsiness and dizziness and is slightly toxic if inhaled	Gloves should be worn while pouring out the DCM and hands should be washed thoroughly after handling. It should only be worked with under the fume hood to avoid breathing in the vapors let out while pouring and evaporating it.
Environment: Dichloromethane (CH_2Cl_2) is toxic to aquatic life	No waste streams containing DCM should be disposed of in skinks. Since most of the DCM will be evaporated, the glassware can be decontaminated with 75% ethanol and left under the fume hood to dry
Safety: Concentrated sulphuric acid (H_2SO_4) is very toxic through inhalation and highly corrosive to skin and objects	Protective gloves, protective clothing, and eye and face protection must be worn. Skin should be washed thoroughly after use and should be handled under a fume hood.
Environment: Concentrated sulphuric acid (H_2SO_4) is extremely toxic to aquatic life	All glassware has to be neutralized and no waste should be washed down the drain due to its corrosivity characteristic. Other waste should be labeled and disposed of through a special waste treatment facility

Safety: Lugol's iodine solution can cause irritation of the nose and throat is inhaled, and temporary eye and skin irritation	Basic protective gloves should be worn and skin should be thoroughly washed afterwards. To avoid possible inhalation, handle the solution under the fume hood.
Ethical	No ethical issues faces due to no living organisms used or directly endangered through the investigation

Materials & Apparatus

Dichloromethane Solvent Extraction:

- 5 × 10 Lipton Black Tea tea bags (22mg caffeine)
- 5 × 250 ± 10mL beakers
- Distilled Water (H_2O)
- Stopwatch
- 5 × watch glass
- Hot Plate
- Funnel
- Cotton Balls
- Electronic balance ±0.001g
- 10.00 ± 0.05ml graduated pipette
- 51.000 ± 0.001g sodium carbonate (Na_2CO_3)
- 5 × 500 ± 0.2mL volumetric flasks
- Stirring Rod
- 250mL separating funnel
- 100 ± 1mL graduated cylinder
- 5 × 50 ± 1mL dichloromethane (CH_2Cl_2)
- Dropper
- Centrifuge & × 4 centrifuge tubes
- 5 × 6.000 ± 0.001g magnesium sulphate ($MgSO_4$)
- Heating Mantle

Iodometric Back Titration:

- 5 × 250mL erlenmeyer flasks
- Distilled Water
- 5 × 10.0 ± 0.1mL sulphuric acid (H_2SO_4)
- 5 × 25.0 ± 0.1mL Lugol's iodine solution (5% iodine, 10% potassium iodide)
- Stirring rod
- Funnel
- Cotton Balls
- Sodium thiosulphate ($Na_2S_2O_3$)
- 50 ± 0.05mL burette
- Starch
- 5 × 2 × 3 drops starch

Procedure

A. Dichloromethane Solvent Extraction[11]

1. 2 Lipton Yellow Label tea bags are placed in a 250 ± 10mL beaker with 150 ± 10mL of boiling distilled water.
2. The beaker is then placed on a hot plate as seen in *Figure 4* with a watch glass on top to avoid evaporation. This is left to brew for 15.00 ±0.01 minutes then removed and left to cool down to room temperature.
3. Tea bags are then disposed of and the brew is filtered through cotton and a funnel to remove any solid particulates.
4. 5.0 ± 0.5mL of the alkali sodium carbonate (Na_2CO_3) solution is added to the brewed tea using a 10.0 ± 0.5mL pipette
    - Preparation of Sodium Carbonate Solutions:
        a. Masses of 1.000g, 5.000g, 10.000g, 15.000g and 20.000g (±0.001) of sodium carbonate (Na_2CO_3) were measured out using an electronic balance.
        b. The solid Na_2CO_3 was added into a 500mL ± 0.15 volumetric flask of distilled water and stirred.
5. The brewed tea and sodium carbonate solution is then poured into the 250mL separating funnel and 25 ± 1mLof dichloromethane (CH_2Cl_2) is added.

Figure 4: Brewing of the tea bags

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[11] Shakeel, Faiyaz, and Mohammed S. Faisal. "Caffeine: A Potential Complexing Agent for Solubility and Dissolution Enhancement of Celecoxib." *Pharmaceutical Biology*, vol. 48, no. 1, 2009, pp. 113–115., doi:10.3109/13880200903030074.

6. The funnel is shaken to allow for maximum contact between the two immiscible layers. It's allowed to relive some of the pressure build up in between shakings. This is then left to settle for 25.00 ±0.01minutes.
7. After two distinct layers are formed by the DCM and aqueous brewing solution as seen in *Figure 5*, the bottom layer, made up of DCM, is drained into a beaker.
8. The DCM at this point still contains remnants of the aqueous layer, so the drained layer is poured into centrifuge tubes and centrifuged for 3 minutes at 60 rotations per second after which the top layer of dark aqueous solution is removed using a dropper.
9. Then, the contents of the tubes are poured into a beaker where magnesium sulphate ($MgSO_4$) will be added and stirred in to act as a drying agent that will clump together with any of the excess aqueous brewing solution.
10. This is then filtered through a funnel with cotton balls to get rid of the clumped magnesium sulphate ($MgSO_4$).
11. The beaker of clean DCM is then placed on a heating mantle and gently heated until all evaporates and power crude caffeine is left behind.

Figure 5: DCM & aqueous brewing solution in separating funnel

12. The caffeine in the beaker is then dissolved in distilled water and poured onto a watch glass to crystalize.
13. The crude caffeine is scratched off the watch glass and placed into vials and weighed as seen in *Figure 6.*

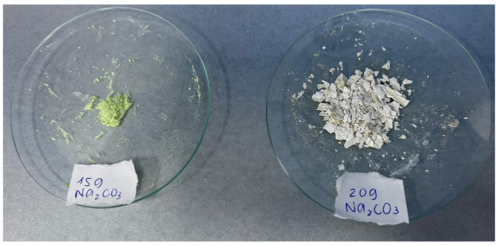

Figure 6: Crude Caffeine after crystallization

B. Iodometric Back Titration[12]

1. Transfer the contents of a vial of crude caffeine into an erlenmeyer flask with 15.0mL ± 0.1mL of distilled water.
2. Add 10.0mL ± 0.1mL of sulphuric acid into the flask.
3. Add 25.0mL ± 0.1mL of the standard Lugol's iodine solution to the flask and mix with stirring rod until a brown solid precipitate begins to form.
4. Filter the solution through a funnel with cotton balls then place the flask under a burette filled with the standard sodium thiosulphate solution.
5. Add 3 drops of starch as an indicator and begin titrating after marking the starting point. The solution should turn dark blue and be colorless at the end point.
6. Take the final measurement of the burette and repeat for the rest of the samples.

Results

A. Quantitative

Table 1: Mass of crude caffeine extracted ±2mg from the solvent extractions

pH of Na_2CO_3 (±0.002)	11.238	11.604	11.756	11.858	11.917
Mass of empty vial ±0.001g	12.421	12.407	12.406	12.450	12.414
Mass of vial with extracted crude caffeine ±0.001g	12.531	12.505	12.502	12.547	12.508
Mass of Extract ±0.002 g	0.110	0.098	0.096	0.097	0.094
Mass of Extract ±2mg	110	98	96	97	94

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[12] Wade, Peter, and J. Hannen. "Iodometric Determination of Caffeine." *Journal of the Science of Food and Agriculture*, vol. 1, no. 6, 1950, pp. 177–178., doi:10.1002/jsfa.2740010606.

Table 2: Raw quantitative data showing the effect of increasing pH on titre volume from iodometric back titration

pH of Na_2CO_3 (±0.002)	**Volume of Sodium Thiosulphate added until endpoint**										
	Titre 1			Titre 2			Titre 3			**Avg. Titre Volume ±0.1mL**	**Random △ of Avg. Titre***
	Initial reading ±0.05mL	**Final reading ±0.05mL**	**Titre volume ±0.1mL**	**Initial reading ±0.05mL**	**Final reading ±0.05mL**	**Titre volume ±0.1mL**	**Initial reading ±0.05mL**	**Final reading ±0.05mL**	**Titre volume ±0.1mL**		
11.238	0.00	35.45	35.5	0.00	35.55	35.6	0.00	35.95	36.0	35.7	0.2
11.604	0.00	35.65	35.7	0.00	35.50	35.5	0.00	36.25	36.3	35.8	0.5
11.756	0.00	36.00	36.0	0.00	36.10	36.1	0.00	36.65	36.7	36.3	0.4
11.858	0.00	36.80	36.8	0.00	36.50	36.5	0.00	37.10	37.1	36.8	0.3
11.917	0.00	36.50	36.5	0.00	36.55	36.6	0.00	36.30	36.3	36.5	0.2

**Note that the calculations for the random uncertainty will be shown in the 'Calculations and Data Processing' section*

B. Qualitative

After the crude caffeine was extracted from the DCM through evaporation then crystallization, it became clear that the powders were not white as pure caffeine is supposed to be. Almost all my samples had a strong green color visible as seen in *Figure 7*, which indicates the presence of a lot of impurities. This means that the mass of these extracts cannot be used as a measure of pure caffeine. This bolsters the choice of conducting a quantitative analysis through iodometric back titration to find the mass of the *pure* caffeine.

Figure 7: All crude caffeine extracted in vials

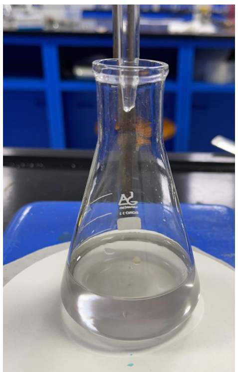


Figure 8: End point for iodometric back titration

The end point of the iodometric back titration is met when the flask's contents had a visible color change from the dark brown-red of iodine to a dark green-blue to the clear solution found in *Figure 8.*

Calculations & Data Processing

A. Finding the pH of the brewing solution analytically

Part I: Preparing the concentrations of sodium carbonate

Table 3: Concentrations of Sodium Carbonate Solutions Prepared

Sodium Carbonate (Na_2CO_3) in 500 ± 0.2mL						
Mass (m) ±0.001g	$\triangle m$	$\% \triangle m$*	Volume (V) ±0.2ml	$\% \triangle V$	Concentration (M)	$\% \triangle C$
0.980	0.001	0.1	500.0	0.04	1.85×10^{-2}	0.1
5.010	0.001	0.02	500.0	0.04	9.45×10^{-2}	0.1
9.987	0.001	0.01	500.0	0.04	1.88×10^{-1}	0.1
15.109	0.001	0.01	500.0	0.04	2.85×10^{-1}	0.05

$$\% \triangle m = \frac{\triangle m}{m} \times 100$$
$$\% \triangle V = \frac{\triangle V}{V} \times 100$$
$$C = \frac{m}{M \times V} \text{ and } \% \triangle C = \sum (\% \triangle V)(\% \triangle m)$$

Note that the percentage uncertainty of the molar mass is insignificant and thus will not be included in the propagation of error

Part II: Calculating the pH

Since the pH sensor at the lab was unable to be calibrated properly and continued giving fluctuating, illogical values for such a weak base, the pH had to be calculated analytically from the values calculated in *Table 3*.

$$CO_3^{2-}(aq) + H_2O(l) \rightleftharpoons OH^-(aq) + HCO_3^-(aq)$$

Since the CO_3^{2-} in Na_2CO_3 acts as a Brønsted–Lowry base, I expanded on my SL curriculum to find the analytical way of calculating the pHs of weak bases in water.

Since the ratio is 1:1, it can be considered that the concentration of Na_2CO_3 = $[CO_3^{2-}]$

pKb for CO_3^{2-} = 3.75

Below is a sample calculation of the pH=11.238 sample

Table 4: Calculating ions

	CO_3^{2-}	H_2O	OH^-	$HCO3^-$
Initial concentration /M	1.85×10^{-2}	—	0	0
Concertation at equilibrium/ M	$1.85 \times 10^{-2} - x$	—	x	x

$$K_b = \frac{[OH^-][HCO_3^-]}{[CO_3^{2-}][OH^-]}$$
$$K_b = 10^{-pK_b} = 1.78 \times 10^{-4}$$
$$\text{Equilibrium constant expression:} K_b = \frac{[x][x]}{[0.018492 - x]} = \frac{x^2}{[0.018492 - x]} = 1.78 \times 10^{-4}$$

Then it can be said that $x = [OH^-] = 1.7 \times 10^{-3}$M

$$pOH = -\log[OH^-] = -(\log 1.7 \times 10^{-3}) = 2.76$$
$$pH = 14.00 - pOH = 14.00 - 2.76 = 11.24$$

Since the ratio of concentration to OH- ions is 1:1, it can be considered that they have the same uncertainties.

Table 5: calculation of pH and uncertainties

$([OH^-] \times 10^{-3})/\text{M}$	$\% \triangle [OH^-]$	$\triangle [OH^-]/\text{M}$	pOH	pH	$\triangle$ pH
1.730	0.1	0.002	2.762	11.238	0.002
4.014	0.1	0.004	2.396	11.604	0.004
5.700	0.1	0.006	2.244	11.756	0.006
7.210	0.05	0.004	2.142	11.858	0.004
8.260	0.05	0.004	2.083	11.917	0.004

To find the uncertainty of the pOH which uses logarithms, I will have to use the half range method to then determine the uncertainty in the processed pH of the varying concentrations of sodium carbonate solutions prepared. The resulting uncertainties are viewed in *Table 5* above.

B. Calculating the mass of pure caffeine

(One sample of the calculations produced from the iodometric back titrations is found below, while the final amounts are found in Table 4)

Reaction #1: $I_2 + 2Na_2CO_3 \rightarrow 2NaI + Na_2S_4O_6$

Reaction #2: $C_8H_{10}N_4O_2 + 2\,I_2 + KI + H_2SO_4 \rightarrow C_8H_{10}N_4O_2.HI.I_4 + KHSO_4$

Table 6: Sample calculations for mass & Purity of caffeine extracted

Variable/Unit	**Sample Calculations**	**Absolute Uncertainty (±)**	**Percentage Uncertainty (%)**
Concentration of Sodium Thiosulphate ($Na_2S_2O_3$)/ M	0.005	0.001	20
Volume of Sodium Thiosulphate ($Na_2S_2O_3$) from titre (mL)	35.7	0.2	0.6
Moles of Sodium Thiosulphate ($Na_2S_2O_3$)reacted/ mol	$n = \frac{C \times V}{1000}$ $n = \frac{0.005 \times 35.7}{1000} = 1.79 \times 10^{-4}$	0.4×10^{-4}	$20 + 0.6 = 20.6 \approx 21$
Concentration of I_2/ M	0.0121	0.0001	0.8
Volume of I_2/mL	25.0	0.1	0.4
Moles of I_2 originally added/ mol	$n = \frac{C \times V}{1000}$ $n = \frac{0.0121 \times 25}{1000} = 3.03 \times 10^{-4}$ mol	0.03×10^{-4}	$0.8 + 0.4 = 1.2 \approx 1$
Moles of I2 reacted with Sodium Thiosulphate ($Na_2S_2O_3$) in reaction #1 / mol	$n = \frac{1.79 \times 10^{-4}}{2} = 8.95 \times 10^{-5}\text{mol}$	$\frac{4.0 \times 10^{-5}}{2} = 2 \times 10^{-5}$	22
Number of moles of I2 left for reaction #2/ mol	$n = (3.03 \times 10^{-4}) - (8.95 \times 10^{-5})$ $= 2.14 \times 10^{-4}\text{mol}$	$(0.03 \times 10^{-4}) + (2 \times 10^{-5})10^{-5}$ $= 2.3 \times 10^{-5} \approx 0.2 \times 10^{-4}$	9.5
Moles of pure caffeine reacted in reaction #2/ mol	$n = \frac{2.14 \times 10^{-4}}{2} = 1.07 \times 10^{-4}$	$\frac{2.0 \times 10^{-5}}{2} = 0.1 \times 10^{-4}$	9
Mass of pure caffeine/ mg	$m = (1.07 \times 10^{-4})(194.19) \times 1000 = 20.78$	0.01	0.05
% Purity of caffeine in the extracted sample	$\frac{20.78}{110} \times 100 = 18.9\,\%$	0.4	$0.05 + 2 = 2$

These same calculations were conducted for the other 4 samples. The final values are seen below in *Table 7:*

Table 7: Crude caffeine vs pure caffeine

pH of Na_2CO_3	Crude Caffeine ±2mg	% △ Crude Caffeine	Pure Caffeine ±0.05mg	% △ Pure Caffeine	Percentage Purity	Percentage Uncertainty of Percentage Purity
11.238	110	1.82%	20.78	2.00%	18.89%	3.82%
11.604	98	2.04%	20.77	2.00%	21.19%	4.04%
11.756	96	2.08%	20.58	2.00%	21.44%	4.08%
11.858	97	2.06%	20.49	2.00%	21.12%	4.06%
11.917	94	2.13%	20.47	2.00%	21.78%	4.13%

C. Calculating random uncertainty of average titre

Avergae Volume for pH 11.238 = 35.65mL

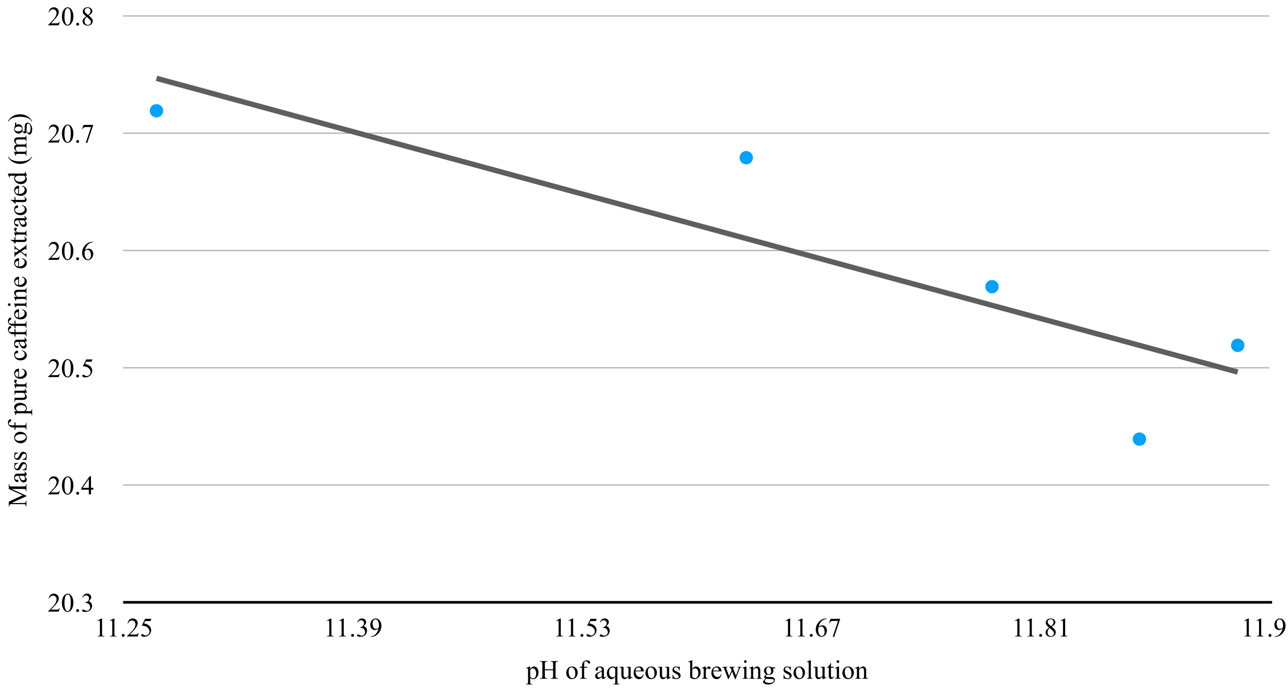
$$\triangle V = V_{max} - V_{average} = 35.95 - 35.65 = 0.3$$

$$\triangle V = V_{average} - V_{min} - = 35.65 - 35.45 = 0.2$$

The larger random uncertainty is taken, in this case ±0.2mL. In this case the random uncertainty is greater than the burette's uncertainty (±0.1mL), I will be ignoring the instrumental uncertainty for the random uncertainty calculated.

D. Processed Data Representation

Figure 9: mass of pure caffeine vs pH of aqueous brewing solution



Conclusion & Analysis

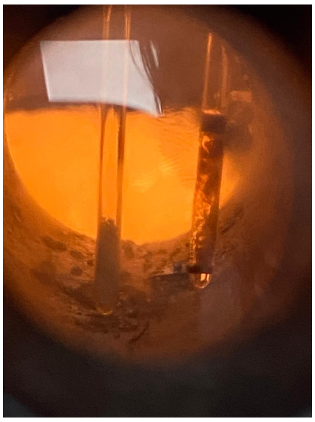


Figure 10: extract in melting point apparatus #1

The negative correlation represented by the graphed data in *Figure 9* with the linear line of best fit sloping down, shows that as the pH of the aqueous solution the tea bags is are brewed in increases from a value of 11.238 to 11.917, the mass of caffeine extracted through the DCM solvent extraction and later calculated through iodometric back titration decreases.

The results obtained strongly suggest that the initial hypothesis, which stated that the variables would be inversely proportional to one another, is correct and sound and even matches the predicted sketch of the results in *Figure 3*. These results also follow the general trend found amongst literature that investigated the effect of pH on other caffeinated beverages within lower pH levels.

The pattern seen within the data might be caused by pH's influence on caffeine's K_d in a solvent-water system. Due to a higher pH leading to a lower K_d value, it can be deduced that this means there is a higher concentration of caffeine in water as seen in the expressions below, which will lead to a lower yield of extraction due to some of it not being present in the solvent. This assumption and theoretical background fits the trends seen in the experimental results.

$$C_8H_{10}N_4O_2\ _{(H_2O)} \rightleftharpoons C_8H_{10}N_4O_2\ _{(DCM)}$$

$$K_d = \frac{[C_8H_{10}N_4O_2]_{(DCM)}}{[C_8H_{10}N_4O_2]_{(H_2O)}}$$

Evaluation

Almost all measures of percentage uncertainties are below 2% therefore almost inconsequential, therefore bolstering the validity of the conclusions made and the analysis of them. Additionally, there was no clear outlier point or anomalous result with the exception of the sample corresponding to the pH value of 11.858 which had a relatively lower yield compared to its surrounding points. This could be due to inconsistencies in the shaking of the DCM as the aqueous layer could have not had enough contact with the solvent.

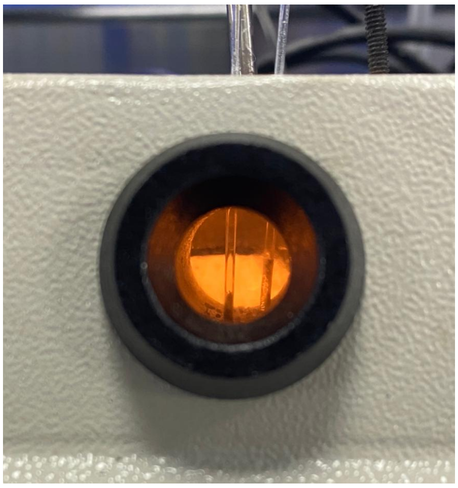


Figure 11: extract in melting point apparatus #2

Additionally, after the iodometric back titration, a small amount of the caffeine from the sample with the highest yield (pH value of 11.27) was placed into a capillary and investigated using a melting point apparatus to compare it to the melting point of a standard caffeine sample. This is seen in *Figure 10* and *Figure 11.* While the melting point of pure caffeine is 228 ±1°C, my sample's melting point was 237 ±1°C which increases the validity of the extracted solid actually being caffeine.

However, my results could benefit from further validation through the use of infrared spectroscopy to analyze both a sample from my extractions and one of pure caffeine and comparing the two fingerprints using technology. This machinery is not accessible, but future investigations could benefit from this.

My results's validity are also inherently flawed as the amount fo caffeine in each tea bag could differ greatly due to manufacturing inaccuracies on Lipton's part. This is as a result of lenient food labeling regulations that allow for a large margin of error, leading to my calculations on percentage yield being skewed and inaccurate. However, my experiment benefited from the reduction of random errors throughout its duration due to to the use of repeated trials and repeated measurements of substances. This improves the validity of my results and the conclusions found.

To further investigate this theory, a wider range of pHs would be analyzed, ranging from acidic to neutral to basic. More types of caffeinated items could also be tested to see if the trend seen within black tea is generalizable to other substances. Additionally, more advanced technology could be used to verify that the extract produced is actually caffeine such as the use of IR spectroscopy.

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