Extended Essay – Chemistry

Discovering the amount of copper found in alcoholic beverages.

To what extent is there a difference between the concentration of copper in *Cachaça* according to the price of the product for Cachaças produced in Brazil measured by titration and by UV-Vis spectroscopy?

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1 OUTLINE OF THE INVESTIGATION

1.1 Aim

The aim of this extended essay will be to compare and evaluate the quantity of copper present in different prices of alcoholic beverages.

1.2 Hypothesis

According to the hypothesis, the quantity of copper present will be proportional to the cost of the *Cachaça*, meaning that the more affordable it is, the more copper will be present. This happens based on the cost of the beverage purification process, the more accurate the process is, the higher the cost of the procedure and of the final product.

1.3 Research Question

To what extent is there a difference between the concentration of copper in *Cachaça* according to the price of the product for *Cachaças* produced in Brazil measured by titration and by UV-Vis spectroscopy?

2 INTRODUCTION

Heavy metals are dense metallic elements that can be hazardous even at low concentrations, depending on the metal.¹ They are characterized by atomic densities exceeding5 g/cm^3 and atomic numbers (Z) greater than 20, as copper (Z=29). Heavy metals are non-biodegradable and tend to accumulate, so when they are introduced into the environment, they congregate and contaminate different trophic levels and different parts of the ecosystem.² Because of that, these metals are found in water, food, beverages, air, and more, and they intoxicate humans by ingestion or inhalation. Once within our bodies, heavy metals may attach

¹ "Heavy Metals - Lenntech." *Www.lenntech.com*, <u>www.lenntech.com/processes/heavy/heavy-metals/heavy-metals.htm#:~:text=The%20term%20heavy%20metal%20refers.</u>

² Briffa, Jessica, et al. "Heavy Metal Pollution in the Environment and Their Toxicological Effects on Humans." *Heliyon*, vol. 6, no. 9, Sept. 2020, p. e04691, <u>https://doi.org/10.1016/j.heliyon.2020.e04691</u>.

to proteins, changing their function and causing people harm³. Even though these chemicals are naturally occurring elements of the planet, human activities are responsible for most environmental contamination and human exposure.

Copper is a heavy metal with very high human use, mainly because of its electrical conductivity. It is also a part of our daily life and has contributed vitally to sustaining and improving society since the dawn of civilization. It has many uses in our society nowadays, being a part of many civilian tasks and part of people's work⁴. With that in mind, it can accumulate in the soil and be taken up by plants, contaminating them. Although copper is necessary to the human body in low quantities, as it helps human development and health, ingesting higher than recommended amounts of copper on daily basis can lead to severe illness, such as kidney and liver damage.

According to Vera Lucia Neves Dias⁵, copper is

[...] distributed practically throughout the entire body, but in different concentrations, which indicates its functional role. Protein activities such as tyrosinase, cytochrome oxidase and ceruloplasmin are basically governed by copper. Despite its great importance in the human body, its excess in the body is harmful due to interference in the normal catalytic activities of some enzymes. (Dias et. al.)

Humans are constantly exposed to copper since it is contained in many foods and beverages. Because copper is essential for our systems to fulfill metabolic tasks, a copper deficiency may endanger a person's health.⁶

Copper, as well as alternative heavy metals, are found in alcoholic drinks, which implies that when these beverages are taken, the individual is ingesting copper. The Food Board Nutritional (FBN) established the Recommended Dietary Allowance (RDA) for this metal is

³ Tamás, Markus, et al. "Heavy Metals and Metalloids as a Cause for Protein Misfolding and Aggregation." *Biomolecules*, vol. 4, no. 1, 25 Feb. 2014, pp. 252–267, <u>https://doi.org/10.3390/biom4010252</u>. Accessed 9 Dec. 2019.

⁴ Geology.com. "Uses of Copper | Supply, Demand, Production, Resources." *Geology.com*, 2017, <u>https://geology.com/usgs/uses-of-copper/</u>

⁵ Dias, Vera Lúcia Neves, et al. "A Importância Do Cobre Na Dieta Alimentar. Revisão." *Hig. Aliment*, 2009, pp. 28–31, <u>https://pesquisa.bvsalud.org/portal/resource/pt/vti-14058</u>

⁶ National Institutes of Health. "Office of Dietary Supplements - Copper." *Nih.gov*, 2017, <u>https://ods.od.nih.gov/factsheets/Copper-HealthProfessional/</u>

from 1.5 to 3.0 mg per day for adults.⁷

Cheap and expensive alcoholic beverages are made in different ways, with distinct procedures and, most importantly, divergent filtering processes. To produce beverages at a cheap cost, the process must be affordable enough that the corporation can still profit, implying that less expensive products are usually less safe for human health because of how the product was made, with its aim to save money.

Cachaça is a term used to define a class of alcoholic beverages in Brazil produced from sugarcane, with alcohol content from 38 to 48% by volume at 20°C, obtained by distillation of fermented sugar cane, according to Brazilian legislation, Decree n. 4851, of 10/02/2003, Art. 92^8 .

This beverage is popular in Brazil and is recognized as a "typical Brazilian beverage", as says Decree No. 4062 of 12/21/2001 and the Industrial Property Law No. 9279/96 that defines the expressions "*Cachaça*", "Brazil" and "*Cachaça* do Brasil" as a product of unique quality having in view of its natural characteristics⁹.

During the production process of a typical Brazilian *Cachaça*, the following steps can be used: preparation of the raw material (sugar cane cutting, separation of its foliage, transport and storage), juice extraction, and fermentation. The result of this fermentation is taken to distillation in an alembic of copper, and this is the stage of possible contamination by this metal.¹⁰

The purification process of alcoholic drinks is an essential and key stage in guaranteeing their safety and maintaining their quality. The standard procedure includes distillation, filtration, and sometimes the aging process in oak barrels. Nevertheless, a possible issue that

⁷ RECOMMENDED DIETARY ALOWANCES. Subcommitte on the Tenth Edition of RDAs. Washington: National Academic, 1998. Cap.10, p.195-246.

⁸ BRASIL. Lei no 9279 de 14 de Maio de 1996. Published on Diário Oficial da União de 15/05/1996. Capturedin 1/ 10/2004. Online. Available on <u>https://www.planalto.gov.br/ccivil 03/leis/19279.htm</u>

⁹ "Decreto Nº 4.062, de 21 de Dezembro de 2001.Pdf — Ministério Da Agricultura E Pecuária." Www.gov.br,www.gov.br/agricultura/pt-br/assuntos/inspecao/produtos-vegetal/legislacao-1/biblioteca-denormas-vinhos-e-bebidas/decreto-no-4-062-de-21-de-dezembro-de-2001.pdf/view.

¹⁰ Garbin, Renata, et al. "Níveis de Cobre Em Amostras de Cachaça Produzidas Na Região Noroeste Do Rio Grande Do Sul, Brasil." *Ciência Rural*, vol. 35, no. 6, Dec. 2005, pp. 1436–1440, <u>https://doi.org/10.1590/s0103-84782005000600033</u>. Accessed 8 November, 2022.

may arise throughout the purifying process is the existence of copper. The leaching of copper into liquid from various equipment, such as stills, pipelines, or aged barrels, might potentially result in health problems for consumers if the concentration surpasses permissible limits. Advanced analytical methods are used for the purpose of detecting copper contamination. Many methodologies may be used for the determination of copper content in beverages, with titration and spectrophotometry being two often employed procedures. The methods mentioned above exhibit a high level of sensitivity and possess the capability to precisely measure low concentrations of copper present in alcoholic drinks.

Titration is used to calculate an analyte's concentration. This process begins by addinga reagent with known properties, titrant, to a solution in which the concentration of a particular component is to be determined, titrate.¹¹

Spectrophotometry measures the intensity of light as it flows through a sample solution to determine how much a chemical compound absorbs light.¹² This experiment is conceivable because some molecules can absorb light in a specific wavelength¹². This allows the measurement of the amount of light emitted or absorbed. Due to this characteristic of electromagnetic radiation, it is possible to perform qualitative or quantitative analyses of a substance's chemical concentration.

In this research, the amount of copper in cheap, average-priced, and expensive *Cachaças* made in São Paulo using two distinct methods, titration and UV-Vis spectrophotometry, was analyzed.

The theme for this Extended Essay was chosen based on two reasons. The first point was the science experience; among all the disciplines in the Brazilian and the IB curriculum, the possibility of doing research in Chemistry brings a great opportunity to improve lab skills. The second reason was the interest in studying the impact of extremely cheap alcoholic

¹¹ Li, Daoliang, and Shuangyin Liu. "Titration - an Overview | ScienceDirect Topics." *Sciencedirect.com*, 2019, <u>www.sciencedirect.com/topics/agricultural-and-biological-sciences/titration</u>.

¹² "Spectrophotometry - an Overview | ScienceDirect Topics." <u>Www.sciencedirect.com</u>, www.sciencedirect.com/topics/physics-and-astronomy/spectrophotometry.

beverages on people's bodies.

Consequently, during the initial research for defining the Extended Essay topic, the possible presence of copper in Cachaças appeared and, as it is a very popular beverage among young people, it created a new aspect to the research: to what extended people drink these Cachaças without knowing about the concentration of copper that they are ingesting and is the concentration of copper in those drinks within the limits provided by the law?.

3 METHOD

3.1 Beverages Samples

My experiments required one cheap, one moderately priced, and one expensive *Cachaça* made in São Paulo. All the beverages were bought by the school and kept with them throughout the process and all the experiments were supervised by the chemistry teacher and my supervisor. No alcoholic beverages were handled without supervision or removed from the laboratory by the student.

Sample 1 - Cheap: Corote

This *Cachaça* is extremely popular across Brazil because of its low price and accessibility, making it a favorite among youngsters and those on a budget. For this experiment, two different flavors of *Corotes* were used, lemon and peach. This was done because the lemon-flavored *Corote* has a neutral color, whilst the peach-flavored *Corote* has a color that is extremely similar to the other *Cachaças*.

Titration is a very visual experiment based on the color changing of the sample. Initially, the sample has one color, and after adding a certain volume of titrant, a color change will occur depending on the indicator that will be used. It is critical to guarantee that the color of all samples does not affect the outcome.

The indicator for titration is a compound that presents different colors in different

chemical environments, for example, phenolphthalein, which is a molecule that has no color in acidic solutions, but which presents a pink color in solutions with a basic pH.

For the titration procedures used in this work, the indicator used was eriochrome black T, which complexes with metallic ions and generally presents a red color for these complexes and as EDTA (titrant) is added, the metallic ions They begin to form complexes with EDTA, no longer with eriochrome black T, changing the color of the solution.

Since the lemon *Corote* has a neutral color, it will not impact the experiment, and because the Peach *Corote* has a very similar color to the other *Cachaças*, if the experiment is altered due to the color, it will have the same changes as the other samples.

This beverage has 700ml and costs R\$ 3.15, which converts to, approximately, \$0.60. As it is a widely popular beverage, comparing *Corote* to the more costly *Cachaças* is interesting. The hypothesis for this research claims that *Corote* will have the highest concentration of copper.

Sample 2 - Average-Priced: Cachaça Engenho São Luiz Premium

This beverage was chosen since it is relatively inexpensive but is still unaffordable for many people. Therefore, following the primary hypothesis, there will be a difference between this beverage and the cheapest one. However, the difference between the concentration of copper of the cheapest and this beverage will be less significant than the cheap and expensive ones. *Cachaça Engenho São Luiz Premium* has 600ml and costs R\$120,00 (approximately \$23,17).

Sample 3 - Expensive: Cachaça Porto Vianna Gouveia Brasil

This is a very expensive *Cachaça*. According to my hypothesis, the difference in copper content between *Corote* and *Cachaça Porto Vianna* will be significant. This 750ml beverage costs R\$294,65, which is, approximately, \$ 56,61.

3.2 Titration

The first procedure before performing the experiment was to wash all the glassware with distilled water. This was done to avoid contamination with copper ions from tap water or from previous experiments performed with the same glassware.

For this experiment, a burette to hold the titrant and an Erlenmeyer flask to hold the titrate are necessary. When the burette's stopcock is opened, the chemical reaction will commence. The titration is complete when the solution in the conical flask changes color, indicating that the quantity of titrant equals the amount of analyte present. With this first experiment, I will discover the amount of copper in each beverage.

3.2.1 Procedure

To measure copper in each beverage, the titration process is based on the following reaction between Cu^{2+} and EDTA (complexing agent), as shown in Figure 1. In this process, the solution containing Cu^{2+} is light blue, a characteristic color of the complex of the bivalent copper ion with water (Figure 2)¹³, but it is essential to highlight that in the *Cachaça* samples, the initial color is caramel/honey (Figure 3) except "*Corote Lemon*" and that is the reason for use two types of *Corote* and therefore the other colors also have variations due to dithering effects.

 $+ H^{+}$ CuH_3In H_4In^- Sample/Analyte Free indicator Metal ion-indicator complex Light Blue Yellow-Red HY^{3-} CuY^{2-} CuH₃In Metal ion-indicator complex Metal ion-EDTA complex Titrant-EDTA ee indicator Colorless Colorless Yellow-Red Purple

Figure 1. Reaction between Cu^{2+} and EDTA. Elaborated by author.

¹³ Hattori, Shigeki, et al. "Blue Copper Model Complexes with Distorted Tetragonal Geometry Acting as Effective Electron-Transfer Mediators in Dye-Sensitized Solar Cells." *Journal of the American Chemical Society*, vol. 127, no. 26, 10 June 2005, pp. 9648–9654, <u>https://doi.org/10.1021/ja0506814</u>.



Figure 2. Structure of the copper-water light blue complex with EDTA. Elaborated by author.



Figure 3. All types of *Cachaça* used in the experiment process and their colors. Picture taken by author.

The burette was filled with EDTA 0.1 mol/L, which was purchased in the form of a standardized solution, for the experiment so that the pH of the EDTA did not affect the outcome or the color of the titrated beverage. All the *Cachaça* samples were titrated with Eriochrome black T (0,001 g, approximately, for each sample) as an indicator.

3.3 Spectrophotometry

The resources for this experiment are not available at the school; therefore, the second experiment was done at USP (Universidade de São Paulo). In this experiment, the data obtained by a standard curve will be used to compare the values from the samples and calculate the

copper concentration for each beverage.

3.3.1 Procedure

3.3.1.1 Standard Curve

Before all the measurements were taken, the cuvettes were cleaned with distilled water, and they were also washed in the same way between the measurements. The wavelength set for the whole experiment was 800nm.

A standard curve is used to determine the quantity of a chemical in an unknown substance by correlating it to a number of standard samples with established concentrations¹⁴. In this case, six solutions with a known increasing concentration of Cu^{2+} were prepared to establish the basis of the standard curve. After the three samples of the four drinks are run through the spectrophotometry experiment, they will fit into the curve, and by comparing their position to the position of the solutions, the cooper concentration in the beverages will be revealed.

Table 1: Concentration of Cu^{2+} and mass of $CuSO_4$ – Standard Curve			
Solution	Concentration (M)	CuSO4 Mass ± 0,0005 g in 250 ± 0,01 mL	
1	0.0010 ± 0.012	0.040	
2	0.0050 ± 0.0025	0.199	
3	0.010 ± 0.0012	0.399	
4	0.050 ± 0.00025	1.99	
5	0.10 ± 0.00013	3.99	
6	0.20 ± 0.000074	7.98	

Table 1. Concentration of Cu^{2+} and mass of $CuSO_4$ of the solutions used as a basis for the standard curve.

¹⁴ Delong, Robert, and Qiongqiong Zhou. "Standard Curve - an Overview | ScienceDirect Topics." Sciencedirect.com, 2010, <u>www.sciencedirect.com/topics/agricultural-and-biological-sciences/standard-curve</u>.

Absorbance is defined as the attenuation of the photon intensity passing through the solution with the absorption of photons by the substance. In the spectrophotometer, it is possible to measure the absorbance of the substance in the sample and from a standard curve to define the concentration of an unknown sample in relation to an analyte.

The Lambert-Beer Law (Equation 1) is the relation between the absorbance, defined through the fraction of light absorbed by the substance, and its concentration in the aliquot of the analyzed solution (analyte).¹⁵ All measurements were obtained on a device Shimadzu UV-1900i spectrophotometer (Figure 4).

 $A = \varepsilon bc$ (Equation 1, where A is the absorbance, ε is the molar absorptivity coefficientb is the optical path and, c is the analyte concentration).



Figure 4. Spectrophotometer Shimadzu UV-1900i. Image from Shimadzu^{®16}

4 DATA COLLECTION

4.1 Titration results

The volumes used in each of the titrations to consume the copper present in the 25mL sample are organized in Table 2, as well as the average volume of the three titrations for each brand of Cachaça.

For the titration experiments, 4 repetitions were always performed and the first measurement was discarded, to avoid possible contamination, while the other three

¹⁵ HARVEY, David. Modern Analytical Chemistry. [S.L.]: McGraw-Hill Higher Education, 2000. 798 p.

¹⁶ "Espectrofotômetro UV-VIS UV-1900i : SHIMADZU." <u>Www.shimadzu.com.br</u>, www.shimadzu.com.br/analitica/produtos/spectro/uv/uv-1900-1.shtml. Accessed 10 January. 2023.

measurements are presented in the data tables.

Table 2: Volume(V) of EDTA used for titration of each beverage and each sample.				
SAMPLE	<i>Corote Lemon</i> ± 0,05 mL	<i>Corote Peach</i> ± 0,05 mL	Cachaça São Luiz Premium ± 0,05 mL	Cachaça Gouveia Brasil Porto Vianna ± 0,05 mL
First titration	V ₁ : 9.3ml	V ₁ : 10.5ml	V ₁ : 0.40ml	V ₁ : 1.9ml
Second titration	V ₂ : 9.6ml	V ₂ : 10.5ml	V ₂ : 0.40ml	V ₂ : 1.9ml
Third titration	V ₃ : 9.6ml	V ₃ : 10.1ml	V3: 0.50ml	V ₃ : 2.1ml
Average	V _{average} :	V _{average} :	V _{average} :	V _{average} :
	9.5 ± 0.05 ml	10.4 ± 0.05 ml	0.50 ± 0.05 ml	2.0 ± 0.05 ml

|--|



Figure 5. Photo of one of the beverages (*Corote Lemon*) before and after the titillation. Photo by author.

In Figure 5, the sample on the left demonstrates the light blue/purple color of the titrated liquid, while the sample on the right demonstrates the yellow-red color of the liquid's reaction to the free indicator before being titrated.

Figure 6 illustrates all the equipment used for the titration experiment and how

everything was prepared before the start of any measure.



Figure 6. Materials used for the titration experiment. Photo by author.

4.2 Spectrophotometry results

4.2.1 Standard curve of CuSO₄

To calibrate the spectrophotometer measurements, a standard curve was created with six copper samples with known concentrations and a white sample without copper. The absorbance measurements of the standard curve samples are listed in Table 3. The range of the standard curve was determined by the references from the literature.¹⁵

Table 3 – Absorption (800nm) value for each concentration of Cu^{2+}				
Solution	Concentration (M) \pm 0,05	Absorption (a.u.) ± 0,00005		
Blank	0,000	0,0000		
1	0,001	0,0033		
2	0,005	0,0199		
3	0,010	0,0501		
4	0,050	0,2032		
5	0,100	0,4319		
6	0,200	0,8677		

Table 3. Absorption value for each concentration of Cu²⁺

4.2.2 Beverages results

The absorbance measurements of each of the samples are listed in Table 4.

Table 4 - Copper absorption on each beverage sample				
Samples	Samples Corote Corote Lemon Peach L		Cachaça São Luiz Premium	Cachaça Gouveia Brasil Porto Vianna
Copper Absorption (800nm, a.u.) ± 0,00005	0,1719	0,1849	0,0073	0,0346

 Table 4. Copper absorption on each beverage sample

5 DATA ANALYSIS

5.1 Titration results

The relation between the concentration of copper in the beverage samples and the amount of EDTA used in the experiment is directly proportional. This is because the quantity

of EDTA required to form complexes with all the copper ions in the samples is indicative of their concentration. Thus, from the titration and the concentration of EDTA used (0.1 mol/L) it is possible to determine how much copper there is in each beverage.

In order to get an accurate result in any experiment, it is essential to conduct many samples and compare the findings. By repeating the procedure three times throughout the titration, any process mistakes, such as visualization problems, were minimized. After completing the titration in triplicate, a mean was computed, resulting in a more accurate result.

At the beginning of the titration, 25mL of each *Cachaça* sample was collected using a volumetric pipette. This volume was transferred to a quantitatively identified Erlenmeyer flask.

To determine the concentration of copper in each beverage, the average volume spent in the titration was used. These volumes are shown in Table 2.

1. Calculation of concentration for the *Corote lemon* sample:

EDTA:

0.1mol --- 1000ml

Xmol --- 9.3ml

Xmol= 9.3 . 0.1/1000 → Xmol= 9.3 . 10^{-4} mol EDTA

N°EDTA = N°Cu²⁺ → N°Cu²⁺ = 9.3 . 10⁻⁴mol [Cu²⁺] = N°Cu²⁺/Vsample → 9.5 . 10⁻⁴/25 . 10⁻³

 $[Cu^{2+}] = 3.8 \cdot 10^{-2} \pm 0.012 \text{ M}$

2. Calculation of concentration for the *Corote peach* sample:

EDTA:

0.1mol --- 1000ml

Xmol --- 10.37

Xmol= 10.37 . 0.1/1000 → Xmol= 1.037 . 10^{-3} mol EDTA

3. Calculation of concentration for the São Luiz Premium sample:

EDTA:

0.1mol --- 1000ml

Xmol --- 0.45

Xmol= 0.45 . 0.1/1000 → Xmol= 4.5 . 10^{-5} mol EDTA

N°EDTA = N°Cu²⁺ → N°Cu²= 4.5 . 10^{-5} mol

 $[Cu^{2+}] = N^{\circ}Cu^{2+}/V \text{ sample } \rightarrow 4.5 . 10^{-5}/25 . 10^{-3}$

 $[Cu^{2+}] = 1.8 \cdot 10^{-3} \pm 0.061 \text{ M}$

4. Calculation of concentration for the Gouveia Brasil sample:

EDTA:

0.1mol --- 1000ml

Xmol --- 1.97

Xmol= 1.97 . 0.1/1000 → Xmol= 1.97 . 10⁻⁴mol EDTA

N°EDTA = N°Cu²⁺ → N°Cu²⁺ = 1.97 . 10⁻⁴mol [Cu²⁺] = N°Cu²⁺/Vsample → 1.97 . 10⁻⁴/25 . 10⁻³

$[Cu^{2+}] = 7.9 . 10^{-3} \pm 0.053 M$

In order to establish a correlation between the copper concentration of the samples, the peach-flavored *Corote* was chosen to compare with the other *Cachaças*.

Analyzing the results presented in Table 5, compared to *Corote* Peach, *Caçhaça Porto Vianna* contains five times less copper, and compared to *Corote* Peach, *Cachaça São Luiz* Premium has 24 times less Cu^{2+.}

By using these data, it is possible to prove that the theory is accurate since there is a variation in copper concentration depending on the price of the beverage. The least expensive beverage (*Corote*) has substantially more copper than the other two samples. Contrary to my expectations, the most expensive *Cachaça* brand has a higher copper concentration than the average-price brand.

5.2 Spectrophotometry results

The absorbance measurements obtained with the standard curve solutions were used to establish an equation that related the copper concentration of a sample to the measured absorbance for that same sample. The equation obtained was y = 4.3323x - 00014 (r²=0,9996), where y means the absorbance and x is the concentration value.

Figure 7 is a graphic representation of the standard curve, with the equation and the r^2 factor. It is possible to affirm that this equation represents a linear curve since the r^2 is very close to 1, and visually the curve is linear.



Figure 7. Standard curve for Cu^{2+} concentration measured by spectrophotometer.

Using the curve equation, it was possible to calculate the concentration for each sample, replacing the value of "y" with the value obtained in the spectrophotometer equipment:

y = 4,3323x - 0,0014

Corote Lemon:

 $0,1719 = 4,3323x - 0,0014 \iff 4,3323x = 0,1719 + 0,0014 \iff x = 4.0 \cdot 10^{-2} M$

Corote Peach:

 $0,1849 = 4,3323x - 0,0014 \le 4,3323x = y + 0,0014 \le x = 4.3 \cdot 10^{-2} M$

Cachaça São Luiz:

 $0,0073 = 4,3323x - 0,0014 \le 4,3323x = y + 0,0014 \le x = 2.0 \cdot 10^{-3} M$

Cachaça Gouveia Brasil:

 $0,0346 = 4,3323x - 0,0014 \le 4,3323x = y + 0,0014 \le x = 8.3 \cdot 10^{-3} M$

5.3 Comparison

The results of both the spectrophotometry method and the titration method, which were both effective at identifying the copper concentration present in the beverages, are shown in Table 6. The table also shows the percentage difference between the measurements made using the spectrophotometry and titration methods.

The numerical difference between the two methods was insignificantly small for almost all the beverage samples, especially because it is preserved the same order of magnitude. The percentage for *Cachaça São Luiz*, however, is considerable (10%) and demonstrates a much higher value for the spectrophotometry method (Table 5).

Table 5: Copper concentration on each beverage for each method				
Samples	Corote Lemon	Corote Peach	Cachaça São Luiz Premium	Cachaça Gouveia Brasil Porto Vianna
Copper Concentration (M) Titration	$3.8.10^{-2} \pm 0.012$	$4.1.10^{-2}\pm 0.070$	$1.8.10^{-3} \pm 0.061$	$7.9.10^{-3} \pm 0.053$
Copper Concentration (M) Spectrophotometry	$4.0.10^{-2}\pm 0.0005$	$4.3.10^{-2} \pm 0.0005$	$2.0.10^{-3} \pm 0.0005$	8.3. $10^{-3} \pm 0.0005$
Difference (%)	5.00	3.49	10.0	5.06
Copper Concentration (mg/L) Titration	2414 ± 0.2500	2637 ± 0.4800	114.4 ± 0.0330	500.7 ± 0.6200
0Copper Concentration (mg/L) Spectrophotometry	2541 ± 0.0005	2728 ± 0.0005	126.9 ± 0.0005	526,68 ± 0.0005
Difference (%)	5.00	3.33	9.87	4.92

Table 6. Copper concentration calculated for each beverage for both methods used in the experiment.

Figure 8 provides a graphic comparison of the two methods employed to measure the amount of copper in each beverage, as well as a comparison of the results to the number that represents the legal limit in Brazilian Legislation. All the *Cachaça* samples are over the allowed limit, as can be observed.

In this graphic, the strong black line represents the Brazilian limit imposed by the legislation for cupper in beverages, the blue data represents the concentration obtained by the absorption method, and the orange data represents the concentration calculated by the titration experiments.



Figure 8. Graphic comparison of the two methods employed to measure the amount of copper in each beverageto the number that represents the legal limit in Brazilian Legislation.

6 CONCLUSION

In conclusion, the hypothesis that the concentration of copper in *Cachaças* is proportional to the price of the beverage, implying that the lower the price, the more copper it contains, is proved correct. While the concentration of copper in *Corote* is much higher, validating my theory, the portion of my hypothesis predicting that the most expensive *Cachaça* would have the lowest concentration of copper was proved incorrect. This occurred because the drink with the average price contained less copper than the more expensive one. Yet, since the *Corote* had the greatest percentage of copper, the main part of the theory, stating that the cheaper beverage contained more copper, was proved correct.

After analyzing the experimental data and comprehending how they support my hypothesis, it is concluded that both methods can be utilized to determine the copper concentration in a sample solution. The results of the two experiments were very similar, indicating the functioning of both, and they provided the essential evidence to support my hypothesis.

There is a numerical disparity between the values discovered in the titration and the values found in the spectrophotometry, as shown in Table 6. Although the difference in most of the beverages is insignificant, determining why there was a difference in the findings is impossible. Even though the cause cannot be determined, there is a significant probability that the difference in the results was caused by titration mistakes since this is a visual experiment that relies on human observation. The possible resolution of this issue may have been accomplished by deciding to include duplicates in the spectrophotometric analysis. Moreover, the use of statistical techniques, as the computation of standard deviation and error propagation, suggests that the resulting are statistically the same.

The analyses of copper levels (Table 5) revealed that all samples of *Cachaça* had levels of this metal above the maximum limit allowed by the legislation (5mg/L). This is very worrying because the legislation is made to ensure the safety of consumers, and with these levels above the allowed, consumption of these products by the population can become dangerous.

The results obtained for *Cachaças* from other parts of Brazil in the works^{17,18} cited as

¹⁷ Stanzani, Fernanda, et al. UNIVERSIDADE FEDERAL FLUMINENSE ESCOLA de ENGENHARIA ENGENHARIA QUÍMICA.

¹⁸ "Níveis de Cobre Em Amostras de Cachaça Produzidas Na Região Noroeste Do Rio Grande Do Sul, Brasil Levels of Copper in "Cachaça" Samples Produced in the Northwest Region of Rio Grande Do Sul, Brazil." *Ciência Rural*, vol. 6, no. 6, 2005, pp. 1436–1440, www.scielo.br/j/cr/a/cNGDwBbBcxXRpRd4JXL685D/?format=pdf&lang=pt.

references for this investigation were not as high for this metal. Nevertheless, only approximately 11% of the samples analyzed in the study published in the *Jornal Ciência Rural* were over the authorized limit, indicating that there may have been an error in the experiment carried out at the school or in the sample collection.

While the values discovered through experiments in the samples differ significantly from those found in the reference material, there is consistency in the value obtained between the triplicate measurements and the two methods. Its consistency ensures that the processes were carried out appropriately and generated precise results. To ensure that no factor influences the outcome of the experiments, a second wash of the glassware with distilled water before and between experiments could be performed.

In future analyses, to obtain a more consistent and accurate result, additional methods to identify the concentration of copper in the beverages could have been performed. An example of a method that could have been used is precipitation titration with analyte reactants.

In conclusion, it is critical to recognize the importance of a proper purification procedure, especially when the final product is a food or beverage. This procedure is in charge of eliminating chemicals, heavy metals, and other undesired substances from the product, guaranteeing that its consumption is safe¹⁹. Products with inefficient purifying processes often include more undesirable chemicals and may be harmful to human consumption. To address the issue of copper contamination in alcoholic beverages the integration of rigorous regulatory measures and severe fines is necessary. This would provide a viable opportunity to mitigate the prevalence of copper contamination within this drinks and ensure the protection of the general populace.

It is essential to emphasize the importance of not consuming alcoholic drinks as a young adolescent since their bodies are still developing and there are dangers to growth or sexual

¹⁹ "How Does Water Treatment in the Food and Beverage Industry Work?" New Food Magazine, www.newfoodmagazine.com/article/89676/water-treatment-industrywork/#:~:text=This%20process%20can%20remove%20both. Accessed 15 February. 2023

development²⁰, as well as an increased chance of accidents because they lose control of their actions. It is important to ensure that teenagers do not drink since they tend to purchase cheaper drinks because they are generally attempting to hide it or do not have money. When adolescents consume cheaper drinks, there is a greater potential for a poor purification procedure and, as a result, a greater danger to their health.

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²⁰ "Underage Drinking | CDC." Www.cdc.gov, 28 Oct. 2020, www.cdc.gov/alcohol/fact-sheets/underagedrinking.htm#:~:text=Disruption%20of%20normal%20growth%20or.

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8 APPENDIX



Figure 1: All material used to produce the solutions with a known copper concentration. Elaborated by author.



Figure 2: Process of collecting the *Corote Lemon* sample. Elaborated by author.



Figure 3: Corote Lemon sample. Elaborated by author.



Figure 4: Process of collecting the *Corote Peach* sample. Elaborated by author.



Figure 5: Corote Peach sample. Elaborated by author.



Figure 6: Process of collecting the Cachaça Engenho São Luiz sample. Elaborated by author.



Figure 7: Cachaça Engenho São Luiz sample. Elaborated by author.



Figure 8: Process of collecting the Cachaça Porto do Vianna sample. Elaborated by author.



Figure 9: Cachaça Porto do Vianna sample. Elaborated by author.



Figure 10: Samples of *Cachaças* used in the titration experiment. Elaborated by author.



Figure 11: Buret used for the titration experiment. Elaborated by author.



Figure 12: Measurements of every Cachaça's pH. Elaborated by author.



Figure 13: All the three samples of each *Cachaça* after reacting with Eriochrome black T. Elaborated by author.



Figure 14: Three samples of *Cachaça Engenho São Luiz* after reacting with Eriochrome black T. Elaborated by author.



Figure 15: Three samples of *Corote Peach* after reacting with Eriochrome black T. Elaborated by author.



Figure 16: Three samples of *Corote Lemon* after reacting with Eriochrome black T. Elaborated by author.



Figure 17: Three samples of Cachaça Porto Vianna after reacting with Eriochrome black T. Elaborated by author.



Figure 18: All copper samples with known concentrations. Elaborated by author.



Figure 19: Corote Peach before the titration. Elaborated by author.



Figure 20: : *Corote Peach* after the titration. Elaborated by author.



Figure 21: Corote Peach before (right) and after (left) the titration. Elaborated by author.



Figure 22: Cachaça Engenho Porto Vianna before the titration. Elaborated by author.



Figure 23: Cachaça Engenho Porto Vianna after the titration. Elaborated by author.



Figure 24: Cachaça Engenho Porto Vianna before (right) and after (left) the titration. Elaborated by author.



Figure 25: Cachaça São Luiz before the titration. Elaborated by author.



Figure 26: Cachaça São Luiz after the titration. Elaborated by author.



Figure 27: Cachaça Engenho Porto Vianna before (left) and after (right) the titration. Elaborated by author.



Figure 28: Corote Lemon before the titration. Elaborated by author.



Figure 29: Corote Lemon after the titration. Elaborated by author.



Figure 30: Corote Lemon before (right) and after (left) the titration. Elaborated by author.